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Vox Sanguinis

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ABSTRACT

Local day session

New organizational needs that affect us

LD01-L01 | New european regulation on substances of human origin (SoHO)

A de Celis-Miguélez¹

¹DG of Public Health and Health Equity, Ministry of Health, Madrid, Spain

In October 2020, the European Commission (EC) starts the revision of Directives 2002/98/EC and 2004/23/EC on the safety and quality of human blood and components and tissues and cells. The drafting process ends in July 2022 and the EC publishes its proposal and sets in motion the co-decision process between the European Parliament and the Council. On 14 December 2023, under the Spanish Presidency of the Council, the co-legislators reached a political agreement.

This Regulation establishes a common regulatory framework for all SoHO, with the exception of organs.

The main objectives are:

- To reinforce protection for donors, recipients and offspring of human assisted reproduction.
- Facilitate access to SoHO therapies based on high safety and quality standards and updated technical rules.
- Improve harmonization between Member States.
- Contribute to the self-sufficiency of critical SoHOs and improve crisis preparedness.
- Consolidate the ethical principles of altruism, voluntariness and non-profit in line with the Charter of Fundamental Rights.

Main developments and challenges. In our country, annually more than 1.7 million donations are carried out, 1.9 million transfusions are performed and 413,000 L of plasma are sent to the fractionation industry for the manufacture of critical medicines. At a national level, and following the creation of the National System for Transfusion Safety, the transfusional medicine unit of the DG Public Health, coordinates, promotes and establishes the actions established in the National Hemotherapy Plan together with the Scientific Committee for Transfusional Safety and the National Hemotherapy Commission. Being also the Spanish Competent Authority for blood and components in the EC and other organizations. This Regulation will imply: The registration, supervision, and authorization and inspection in certain cases, of all

entities that carry out any activity, with an impact on quality and safety. The authorization of new preparations that will require the assessment and monitoring of quality, safety and effectiveness aspects, including the implementation of clinical studies. New digital platforms that require increased data reporting. New regulatory coordination, consultation and decision-making structures at European level, such as the SoHO Coordination Board. A crucial aspect is the management of the supply of critical SoHOs, defined as those whose shortage may endanger the health of recipients or the manufacturing of products developed from SoHO, including plasma. The Regulation calls on Member States (MS) to have a sufficient, adequate and resilient supply of critical SoHOs aimed to appropriately meet recipients' needs and to contribute to EU self-sufficiency. MS shall facilitate public sector involvement in donation activities on the basis of the principle of VUD, to ensure a broad and resilient SoHO donor base capable of dealing with crises or disease outbreaks. All these measures are in line with the joint declaration of the 27 heads of state and government, Declaración de Granada (October 2023), which addressed the development of the concept of "open strategic autonomy in Europe" and reaffirmed the high strategic value of plasma in the general interest of public health.

Conclusion: This Regulation aims to strengthen the existing legal framework while allowing the flexibility to incorporate the necessary technical and scientific advances so that patients and recipients benefit from SoHO therapies and prepares the EU for crisis and resilience to safeguard access to SoHO therapies.

LD01-L02 | Plasmapheresis and plasma donation - challenges in the blood/plasma supply chain

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Plasma donation and the availability of blood products is a balance between citizens' rights and duties. Patients have the right to receive the blood products needed to treat their disease. At the same time, citizens in good health have a duty to give and the right to be treated with respect by blood banks. Blood banks have a duty to obtain the plasma needed to cover the consumption of blood products and to treat donors with the utmost respect. Patients are motivated by their need in the face of illness and blood banks by the mission they have been entrusted with. In non-remunerated models, donors are motivated by the desire of solidarity. In paid donation models, donors may be motivated by this reason, but are primarily motivated by financial need. If, as has occurred in the last two decades, the consumption of blood products increases dramatically, plasma donation must increase in the same proportion. In view of this, we should ask ourselves whether

solidarity is sufficient to cover the enormous increase in IgG consumption. The paid plasma donation model is effective, but not necessarily the only possible approach and it should be analysed to see whether it has a negative influence on other types of donation. Strategies should be implemented to increase plasma donations while maintaining current levels of whole blood donation. Strategies based on: information on plasma donation and making donation a pleasant and effective experience. Respecting donors, which means preventing unfavourable effects of continued plasma donation and avoiding excessive pressure to donate. For this purpose, it is essential that IgG consumption does not exceed clinically justified needs. Altruistic, compensated and remunerated donation. There is sometimes little separation between altruistic, compensated and remunerated donation. Perhaps pure altruistic donation does not exist; donors are compensated or gratified by the satisfaction of fulfilling a duty, social recognition, refreshments, personal treatment and small gifts. Financial compensation that covers the donor's justifiable expenses for making the donation is completely licit. To suggest it should cover time spent, fear and other discomforts is possibly entering the field of remuneration. As a general rule, blood banks cannot buy donors' willingness or induce donation through financial compensation, but they should be grateful for the effort they make. Donation of plasma in Spain. Spain, like most European countries, has a plasma deficit and a clear dependence on blood product from other countries, where they are obtained for a fee. In 2023, 404,842 litres of plasma were fractionated to obtain albumin and IVIG. With a yield of 26.5 g/L of albumin and 4.3 g/L of IVIG, a total of 10,730,752 g of albumin and 1,734,249 g of IVIG were obtained, representing 59% and 36% coverage, respectively. If the goal were to reach 50% IVIG sufficiency, a total of 172,420 L more plasma would have to be obtained in Spain than is currently used for fractionation. This would mean performing 287,366 more plasmaphereses than at present. The National Plan must provide the tool to achieve these objectives.

Local day session

Transfusion medicine in our environment: Opportunities and threats

LD02-L01 | Consequences of the new composition of plasticizers on blood components

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Dispensable bag systems for blood and blood component collection and processing are medical devices that should comply with the requirements of the relevant regulations (such as EU directives,

the European Pharmacopoeia and ISO standards). Materials used in the manufacture of these plastic containers should be carefully chosen to minimize the risks arising from the leaching of chemical constituents into the product. Particular attention should be given to the toxicity of the materials used and the biological compatibility of the plastic containers with the products. There is concern that exposure to diethylhexylphthalate (DEHP) may be toxic to humans, as in vivo animal studies have shown reproductive and developmental impairment to occur when chronically exposed to high doses of DEHP. On the basis of these studies, DEHP is classified as category 1B for reproductive toxicity according to CLP-Regulation (EC) No 1272/2008. Consequently, the European Medical Device Regulations (EU MDR 2017/745) banned the use of DEHP in medical devices above 0.1% (w/w). The European Chemicals Agency (ECHA) has proposed to the European Commission to terminate the medical device exemption for DEHP. As a result, Commission Regulation (EU) 2023/2482 of 13 November 2023 amending Annex XIV to Regulation (EC) No 1907/2006 sets 1 July 2030 as the sunset date and 1 January 2029 as the latest application date for the use of DEHP in medical devices. Plasticizers such as di(isononyl) cyclohexane-1,2-dicarboxylate (DINCH), butyryl trihexyl citrate (BTHC), di(2-ethylhexyl) terephthalate (DEHT), di(2-ethylhexyl) 4-cyclohexene-1,2-dicarboxylate (DOTH), and 4-cyclohexene-1,2-dicarboxylic acid dinonyl ester (DL9TH) have been evaluated under different conditions, including different preparation methods, product volumes, storage conditions, and additive solutions, with varying results. Alternative materials are being developed as potential replacements for DEHP, but more data are needed to better understand their leaching properties and, in some cases, their potential toxicity, as well as their impact on the medical effectiveness of treatments. The removal of DEHP from the entire blood bag set (including bags, tubing and connectors) requires the validation of red blood cell (RBC), platelet, and plasma quality by both blood centers and blood bag system manufacturers. The RBC components will require extensive validation because their quality is directly affected by the absence of DEHP, as opposed to platelet and plasma components. Therefore, ongoing research is required to identify and optimize the best possible combination of storage solution and plasticizer. European blood alliance (EBA) members have recently established recommendations for the quality evaluation of blood components obtained with non-DEHP devices to serve as scientific support in the authorization process specific to their jurisdiction or for their internal validation use. In parallel to the non-DEHP transition, anticoagulant and storage solution-containing blood bag systems are classified from class 2b to class 3. This means that clinical evaluation is needed, but it is currently unclear what type of clinical evidence will be required for newly CE-marked medical devices. Manufacturers, blood suppliers and competent authorities need to work together to prevent missing deadlines again and for an orderly transition to DEHP-free blood collecting systems without threatening the blood and blood product supply.

LD02-L02 | Epidemiología de los brotes del WNV en España y sus consecuencias para la donación de sangre

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West Nile virus is a mosquito borne virus that replicates in birds and may produce disease in humans. While 80% of the infections in humans are asymptomatic, 1% of the cases can result in severe disease including meningoencephalitis and death. Transmission to humans can occur through the bite on an infected mosquito, but also by transfusion of blood or organ transplant from a viraemic person. Evidence in birds, mosquitoes and horses indicates that the virus is present in Spain at least since 2003, but only six cases of human disease were notified until 2020, one in 2004, two in 2010 and 3 in 2016. In 2020 a large outbreak with 77 cases of severe disease and 8 deaths occurred in western Andalucía and southern Extremadura. Virus amplification occurred in natural areas but also in urban areas, and virus antibodies were detected in blackbirds and house sparrows captured in the parks of the Coria and Puebla del Rio. Since then, new cases have been notified each year in Spain. In 2021 we started an entomological surveillance program monitoring the abundance of the main mosquito vectors and the presence of West Nile virus in the mosquitoes by Real Time PCR. Mosquitoes are captured, identified to species level and pools of up to 50 females are analysed by virus detection. West Nile virus infection is detected mainly in *Culex perexiguus*, but also in *Culex pipiens*, *Culex modestus* and *Culex laticinctus*. The first detection of virus in mosquitoes occurs four weeks in advance to the reporting of the first human cases. This system has been incorporated by Andalusian regional government (Junta de Andalucía) as a surveillance program to reduce West Nile virus incidence. Mosquitoes are monitored at weekly intervals at 26 localities, and virus detection results in notifications sent to local authorities to reinforce communication of mosquito prevention measures to population, alert local health authorities and hospitals and implement screening of blood and organ donations originating from areas with reported cases of West Nile virus infection in mosquitoes, birds, horses or humans. During the 2023 season, 19 of West Nile virus infection were reported in different regions of Spain, 13 presented neurological symptoms, 1 was febrile and 5 were detected after the screening of blood donations originating from areas with already declared West Nile virus circulation. Overall the results show that virological surveillance in mosquitoes are a useful tool for early detection of West Nile virus circulation.

LD02-L03 | Estado actual de la transfusión domiciliar en España

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Home transfusion is a well established reality in Spain. Since the late 80s, hospital-at-home units in the main teaching hospitals have

received transfusion support. However, even if this can be considered a collective success that has allowed a new paradigm of patient management through process de-hospitalization, still many hospitals do not offer home transfusion. There are several reasons for this: lack of resources, lack of interest, and lack of confidence due to safety concerns. However, the number of hospitals that perform home transfusion or are getting ready for it increases yearly. We will show the result of the survey we conducted among hospital members of the Spanish Blood Transfusion Society and will have a look at other forms of out of hospital transfusion. We will also analyze the causes for not implementation at the moment. What we find is an active landscape of home transfusion, where the prospect of transfusing at the home is widely accepted as a therapeutic tool. We will also have a look at the research options that arise in this context. We can say that home transfusion is in good health in Spain, and that if resources are available, it will be capable of increasing its presence out of the hospital, where the focus of attention is being put due to technological improvements.

LD02-L04 | Transfusión y calidad de vida—Quality of life and transfusion

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The outcomes of transfusion are, from a theoretical point of view, quite clear. For the transfusion of packed red cells it is to keep or increase the oxygen supply to tissues. Since this parameter cannot be easily measured on a daily basis, the level of haemoglobin is used instead and there is a number of papers evaluating its thresholds to prescribe a transfusion. Evaluating if these thresholds have an impact on survival and in the quality of life is still a challenge. When evaluating quality of life there are several hurdles. The first one is the difficulty in assessing such a subjective and complex parameter as quality of life. In this sense the World Health Organization is making efforts in defining the different domains some of which are particularly affected by the health status such as fatigue, pain, dependence and mobility. Some societies are also making efforts in standardising questionnaires for assessing quality of life. When reviewing transfusion guidelines, there are several medical conditions that require a chronic transfusion and in which the aim of treatments should be to improve health-related quality of life. Publications in myelodysplastic syndromes indicate that quality of life is improved with a liberal transfusion policy. Nevertheless, in this disease there are a number of other factors with an impact on quality of life. The perception of disease in children and adults with major thalassemia differs and their quality of life, despite being transfusion dependent, varies. Regarding sickle cell patients, transfusion is also associated with different quality of life in adults and in children. In conclusion efforts in research of quality of life, also related to transfusion, and in moving it forward to the daily practice will be beneficial for patients.

Local day session

A look into the future

LD03-L01 | Adoptive immunotherapy with virus-specific T cells in immunocompromised patients

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Infusion of immune competent donor lymphocytes (DLI) offers a possibility of treatment to immunocompromised hosts although it carries risks such as triggering the graft-versus-host reaction (GVHD). To decrease risk, cell selection methods have been developed to improve specificity and safety. Some are based on negative cell selection, eliminating naïve lymphocytes that triggers alloreactivity (i.e., CD45RA depletion), or by functional methods such as photodepletion after mixed lymphocyte culture or conventional (DLI modified with suicide genes). However, the strategies of positive selection of specific lymphocytes seem safer. Methods to select or enrich virus-specific T (VST) cells are based in direct selection methods using specific tetramers or by selecting those interferon-gamma positive after activation with virus peptides or through ex vivo expansion involving the repeated stimulation with donor antigen-presenting cells challenged with viral peptides. These advanced therapy technologies have evolved during the last years, and scientific literature show VST are a good option for the treatment of refractory CMV, EBV and adenoviral infections and for several other viruses related to transplants. Authors shown to be a safe therapy with low rates of GVHD (usually below 10%) and good responses (ranging up to 70%-80%). For a more universal therapy, availability of third-party healthy donors allows for the immediate use of cells for allogeneic therapy in cases where patients lack an appropriate donor. We will discuss on the creation of cell donor registries of HLA typed donor tested for presence of VST for the most prevalent opportunistic viruses. Creation of the REDOCEL registry and data on clinical outcomes of a VST therapy based in direct selection will be presented.

LD03-L02 | A look into the future - artificial intelligence in transfusional medicine

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Artificial intelligence (AI) refers to the theory and development of computer systems able to perform tasks commonly associated with intelligent beings, such as solving problems, making decisions, recognizing speech, detecting patterns and above all the ability to learn. Natural language processing (NLP), machine learning and deep learning are technologies normally included in the term AI. Machine learning (ML) is a subset of AI

based on the study of computer systems that learn automatically from experience but without being intentionally programmed. ML ranges from simple linear correlations or logistic regression, perhaps combined with dimensionality reduction techniques, to clustering techniques or tree-based methods. ML can be trained on small data sets or with Big Data databases. Deep learning (DL) is a machine learning technique that use artificial neural networks that imitate the structure of the human brain. It makes non-linear and complex correlations and need large amounts of data to train. Natural language processing (NLP) is a subfield of artificial intelligence, linguistics, and computer science concentrate on making human communication, such as text and speech, understandable to computers. In recent years there has been an extraordinary expansion of the literature on ML applying to healthcare, and specifically to transfusion medicine. ML could integrate complicated and diverse databases and could help making decisions, support individualized care, and along with additional design, enhance patient outcomes. Most studies on ML and transfusion medicine utilizes supervised learning so the model is trained on input features and labeled output features to facilitate predictions on hidden examples. However, there are other ML methods: unsupervised ML which identified patterns in unlabeled data and reinforcement ML that learns through trial and error. Many of the investigations on the application of ML to transfusion medicine used supervised ML while a few utilize unsupervised ML. Some use deep learning. None of the studies used reinforcement ML. Studies on ML and transfusion medicine were concentrated on prediction of blood transfusion, hemovigilance, hospital blood bank inventory management and helping transfusion decisions. Within prediction of blood transfusion, a significant majority of research were in surgery and trauma patients. Additionally, recent advances in NLP may involve the development of systems that could integrate decision-making and structures data from electronic health records with clinical notes or verbal instructions. It is very encouraging to think that artificial intelligence can help improve transfusion medicine and close the gap between recommended blood use and clinical practice. This is a field full of interests and expectations as well as challenges. Hopefully artificial intelligence helps us make medicine more humane.

Presidential award session

Novel developments in the use of platelet biomaterials

PL01-L01 | Platelet lysates and extracellular vesicles in cell therapy and regenerative medicine

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Platelets, traditionally recognized for their critical role in hemostasis, are now at the forefront of pioneering developments in cell therapy

and regenerative medicine, where the blood transfusion community should play a vital role. Quality-assured allogeneic platelet concentrates, sourced from healthy donors, serve not only to prevent or control bleeding in patients with thrombocytopenia or platelet dysfunction but are also increasingly recognized as a cornerstone source material for advancing biotherapeutic applications. These applications leverage the fundamental physiological role of platelets in immunity, inflammation, wound healing, and tissue repair, thanks to their content of alpha-granules, dense granules, mitochondria, lysosomes, which are rich in growth factors, cytokines, antioxidants, enzymes, and signaling molecules. The production of pooled human platelet lysates (HPLs) from outdated allogeneic platelet concentrates, which are no longer suitable for transfusion represents a significant innovation in recent years. These HPLs, replacing fetal bovine serum, offer an efficient, clinical-grade, xeno-free growth media supplement for the expansion of therapeutic human cells (including mesenchymal stromal cells and others) for transplantation and applications ranging from regenerative therapies to cancer immunotherapy. Moreover, the growing exploration into allogeneic platelet concentrates for generating tailor-made single-donation or pooled HPLs and HPL-derived extracellular vesicles (p-EVs) reflects a potential transformative approach to biotherapies. These preparations are under investigation in both pre-clinical and clinical studies for their potential to treat a variety of degenerative diseases and traumas, including ocular surface disorders, wound care, osteoarthritis, and even neurodegenerative diseases. Since platelet concentrates are already established as medicinal products and are readily available, their use as a source material for biotherapies may expedite the pre-clinical research and clinical translation process more efficiently than other cellular sources. Of paramount importance in this emerging field is the quality and safety of the starting platelet concentrates, as well as ensuring the pathogen safety of the resulting platelet-derived preparations, especially those when resulting from pooling of multiple platelet concentrates. This underscores the indispensable role of blood organizations and research institutions worldwide in not only ensuring these standards but also in driving the development of safe platelet lysates as ancillary materials for cell culture, as well as well-characterized, virally-safe platelet lysates and p-EVs as novel “platelet-derived medicinal products (PDMPs)”, tailored for evidence-based clinical applications. This presentation aims to provide a comprehensive update on the translational applications of allogeneic platelet-derived biotherapies, highlighting the expanding role of blood products beyond traditional transfusion practices into a realm where they serve as key components in the development of novel therapeutic modalities.

PL01-L02 | The pathogen safety of pooled blood products—lessons to be learned for platelet products

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The use of medicinal products derived from human blood is well established in many different clinical applications and, in the context of the current state of the art, is indispensable for the foreseeable future. The risk of transmitting an infectious disease agent through these therapies is, however, equally a fact, with regular reminders provided by the emergence or global transposition of viruses. This applies even more to products that are made from the donations of several individuals, as the risk can be statistically collected and therefore potentially be higher. Plasma-derived medicinal products (PDMPs) are manufactured from the plasma donations from many thousands of donors yet are now successfully safeguarded by the selection of low risk donors, the testing of donations and plasma pools, and their manufacturing processes that have been designed to inactivate or remove any potentially present viruses. The pathogen safety measures deployed for PDMPs will be discussed, with a specific emphasis on how they can also be used for platelets and platelet-derived products.

PL01-L03 | Human platelet lysates, a new treatment for neurodegenerative diseases?

L Buée

Abstract not available

Plenary session

Ideas changing transfusion medicine

PL02-L01 | Augmented reality in transfusion medicine

R Fine

Abstract not available

PL02-L02 | CRISPR gene editing and blood disease (Casgevy + SCD / B-thal)

S Corbacioglu

Abstract not available

PL02-L03 | Glycosidases and universal blood

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The enzymatic conversion of A and B antigens on the surface of red blood cells (RBCs) to the universal group O, has long been sought as an attractive solution to simplify blood logistics, avoid potentially fatal ABO-mismatches and to minimise spill of expired blood units. The concept of enzymatically converted group O (ECO) RBCs was coined by Goldstein et al. in 1982, using an α -galactosidase from coffee bean to convert group B RBCs to group O, which was transfused into volunteers to demonstrate the safety and potential of the concept. Since then, increasingly more efficient exoglycosidases of microbial origin against A and B antigens were reported, with recent work demonstrating the potential of the human gut microbiota as a source of ECO enzymes. Despite these major advances, translation of the ECO concept to clinical practice has yet to materialise, in part due to safety concerns raised in subsequent clinical trials, revealing unexplained weak crossmatches when group B converted RBCs were transfused into A and O recipients. Common to all hitherto reported enzymatic efforts to generate ABO-universal blood, is the focus on enzyme efficiency towards the A and B antigens. To date, the underlying assumption has been that the complete removal of these antigens will yield ABO-compatible RBCs. Notably, three different extensions of the A antigen (Gal-A, H type 3 and A type 3) contribute to the complexity of the A phenotype, especially in the A₁ subgroup. More recently, Olsson et al. reported the first extension of the B antigen by a $\beta(1,3)$ GalNAc, officially designated ExtB. Notably, no enzymes targeting the extended A or B antigens have been reported, and the impact of these glycans on the ECO concept has not been previously investigated. We hypothesized that mucus-associated members of the human gut microbiota, which are exposed to ABO-decorated mucins, are likely to have evolved enzymatic routes to harness these abundant and constantly supplied glycoconjugate nutrients. This focus on the mucus ecological niche led us to select the dedicated mucin-degrading specialist *Akkermansia muciniphila* for enzyme discovery. Biochemical evaluation of 23 *Akkermansia* exoglycosidases, led to the discovery of enzymes, which transformed both the A and B antigens, in addition to their four known extensions. Sequential and one-pot treatment strategies guided the formulation of enzyme blends for group A and B RBCs, and the resulting cells crossmatch reactivity with 100 group O plasmas was evaluated. Removal of both the A and B antigens as well as their extended forms led to a significant increase in compatibility of the ECO-RBCs with the O plasmas, as compared to conversion of A or B antigens alone. Structural analyses of the converting enzymes allowed us to analyse the structural elements that underpin high efficiency of the discovered enzymes on RBC surface glycoconjugates.

Plenary session

Implementation science

PL03-L01 | Learning health systems

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Clinical evidence that can cut avoidable deaths and enhance quality of life does not reliably find its way into everyday patient care. There are failures to introduce effective new clinical practices quickly enough, consistently use those already proven to be effective, or stop using those found to be ineffective or even harmful. The gap between evidence and practice is a strategically important problem for policy-makers, healthcare systems and research funders because it limits the health, social and economic impacts of clinical research. This gap also applies to transfusion medicine. Implementation science frameworks and evidence can help close such gaps. There is a growing evidence base on the effectiveness and value for money of implementation strategies, which transfusion medicine can adopt or adapt. For example, audit and feedback involves providing healthcare professionals with summaries of their clinical performance over a specified period of time with the intention of motivating improvement. Computerised decision support systems (CDSSs) within electronic health records aim to improve safety by reminding clinicians to deliver recommended care and reducing errors in decision-making. Both of these strategies generally have modest if worthwhile effects and are scalable but their effects can vary greatly by setting and targeted clinical behaviour. Therefore, it is important to further develop and evaluate implementation strategies to enhance their effectiveness in transfusion medicine. This requires well-conducted randomised trials but individual studies can be expensive and time-consuming. However, we have demonstrated the feasibility and utility of embedding trials within existing large-scale initiatives that use existing data, such as national clinical audit programmes. Such work represents the evolving features of a learning health system, using rigorous, data-driven methods to continuously improve transfusion practice and integrating research and quality improvement. Secure funding, interdisciplinary collaboration and ambitious goals are essential to underpin the evolution of data-driven learning health systems for blood transfusion.

PL03-L02 | Implementation science in transfusion medicine

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All transfusion teams will be aware of a gap that exists between evidence-based recommendations and what happens in practice. This may be manifested as variation in transfusion practice that cannot be explained by case-mix alone. Transfusion teams devote considerable

time and resources to the design and implementation of different strategies to improve transfusion practice to be more aligned to national standards and recommendations. These tools can include education, clinical guidelines at local and national levels, and audit and feedback. These approaches may be delivered either as single or multimodal interventions. More recently approaches based on electronic systems such as computerised decision support systems have been suggested as tools for effective change in transfusion prescribing behaviour. Yet the effectiveness (and crucially cost-effectiveness) of these interventions is unclear in transfusion medicine. These interventions can (and should) be subjected to research scrutiny. Systematic reviews suggest limited and inconsistent impacts for many implementation initiatives in transfusion medicine. Most published studies are not randomised trials. The AFFINITIE programme (Audit and Feedback INterventions to Increase evidence-based Transfusion practice) in UK provides an example of a robust evaluation of audit and feedback. Initially a series of interviews and surveys were conducted to better understand the challenges of feedback delivery and effectiveness at hospital sites. The AFFINITIE research team then developed two empirically and theoretically informed feedback interventions, 'enhanced content' to improve feedback clarity and usability and 'enhanced support' for hospital staff to act on feedback, on transfusion appropriateness. The modified interventions were then tested in two linked 2×2 cluster-randomised trials in national audits of transfusion for surgery (135 hospitals) and haematological disorders (134 hospitals) respectively. However, the enhanced feedback interventions were found to be no more effective than standard feedback. This study highlights the need to avoid assumptions about the effectiveness of apparent improvements to current interventions. There is a need to continually evaluate our approaches to improving transfusion practice, and ideally in the context of robust clinical studies. Audit and feedback generally works but those effects may be limited and we need a better understanding of the reasons for inconsistency and how to improve its impact. Reasons for lack of effect in AFFINITIE potentially included a lack of credibility of the audit standards, concerns about the data validity collected by the largely manual-based processes and evidence of variable (and often poor) enactment of the intended feedback at hospital sites. Further talks in this session and the workshop will explore these issues in more detail, and offer opportunities for advancing knowledge on how to accelerate implementation in transfusion medicine.

PL03-L03 | Practical challenges in hospital based implementation

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The ability to develop (and then execute) a roadmap for implementing change within healthcare is a core skillset for transfusion medicine professionals. This session delves into the intricacies of implementing quality improvement (QI) projects within hospital

settings, addressing practical challenges encountered throughout the process. Beginning with project selection, the session emphasizes the importance of aligning QI initiatives with organizational priorities and patient needs. Common QI project drivers include: patient safety issues (e.g., unnecessary transfusion of red blood cells leading to transfusion reactions), supply concerns (e.g., recurrent shortages of platelets), patient concerns (e.g., implementation of low vacuum sample collection tubes to reduce diagnosis blood loss), and cost (e.g., de-prescribing efforts for intravenous albumin to reduce supply costs). The selection of an interdisciplinary team is explored, highlighting the significance of diverse expertise and effective collaboration. The team needs to include both local champions from the physician, nursing, and technologist groups, but also key stakeholders that are concerned about the change being implemented. By including "slow adopters" on your project team you will ensure that you identify as many obstacles as possible before you select your interventions. It is important to submit the project plan to the local research or QI review board to allow for later sharing of results through academic presentations or articles. Conducting a thorough baseline audit is pivotal for accurate project assessment. Strategies for data collection, analysis, and interpretation ensure a solid foundation for intervention planning. A local biostatistician should be identified to assist with project planning, drafting the statistical analysis plan, and providing assistance with the graphical representation of the data (e.g., run charts for the publication or for monthly feedback to the clinical team). The QI project needs to identify an appropriate primary outcome measure, process measures, and balancing measures. The QI team needs to set, a priori, the threshold for achieving the targeted improvement in the primary outcome. Choosing appropriate interventions involves a critical evaluation of evidence-based practices tailored to the hospital's unique context. Interventions can be deployed as a bundle or sequentially over time. The session provides insights into overcoming obstacles faced during implementation, including resistance to change, resource constraints, and communication barriers. Success celebration strategies are outlined to sustain motivation and reinforce positive outcomes. Guidance on publishing project results is discussed, emphasizing the critical importance of the dissemination of lessons learned to contribute to broader healthcare QI efforts.

Award session

Jean Julliard Prize session

PL04-L01 | Anti-Platelet Factor 4 (PF4)-antibodies in VITT and beyond

L Schönborn

Abstract not available

PL04-L02 | Platelet extracellular vesicles and DAMPs in immunity and transfusion

G Marcoux

Abstract not available

Plenary session**Risks of an aging donor population****PL05-L01 | Global aging challenges to blood collection**Y Fung¹¹School of Health, University of the Sunshine Coast, Sippy Downs, Australia

Many countries across the globe are experiencing growth in the proportion of people aged over 60 years in their population. This demographic change is not limited to high income countries, as WHO reports that by 2050, 80% of older people will be living in low and middle-income countries (WHO Aging & Health 2022). Population aging has a direct impact on the practice of blood collection because the donation criteria of most blood collection agencies (BCAs) specify lower and upper age limits. With a maximum donation age, older donors will have to retire and be lost from the donor pool. To address this loss, many BCAs have activities directed at recruiting younger donors to meet blood supply needs even though the proportion of younger people relative to the whole population is shrinking. A few countries now have no upper age limit for regular/repeat donors. Compounding this problem, is evidence from developed countries which show that older patients use a disproportionately higher number of red cells (>50%). Hence, the aging population may not only reduce the blood donor pool, but they may also increase the demand for red cells. This talk will review recent data from Asian BCAs which spans countries with younger populations as well as those with older populations (Fung et al Vox Sanguinis 2023). While blood collection is a relatively common practice, it is also important to note that blood donors are a heterogeneous cohort even within each BCA. The height and weight of each donor determines their total blood volume, and this in turn has implications for the volume of blood that can be safely donated. Data from BCAs in Asia will be used to provide examples of how BCAs customise their donation criteria to their population. The data also raises questions on the appropriateness of general global blood donation criteria such as minimum Hb levels. And highlights the limited volume of evidence to develop local donation criteria outside the Americas, Europe and Australasia. With the ease of travel and immigration, there is growing ethnic diversity in many populations across the world. Hence, would customising blood donation criteria to ethnic groups or to an individual within each BCA, contribute to improving donor safety and extend or remove the age limit to continue donating.

PL05-L02 | Ageing in the hematopoietic stem cells nicheM C Florian^{1,2}¹ICREA, ²Regenerative Medicine, IDIBELL, Barcelona, Spain

One goal of regenerative medicine is to rejuvenate tissues and extend lifespan by restoring the function of endogenous aged stem cells. However, evidence that somatic stem cells can be targeted in vivo to extend lifespan is still lacking. Recently, we provided proof-of-concept evidence that a short systemic treatment with a specific CD42 inhibitor (CASIN) rejuvenates aged blood stem cells in vivo, exerting a broad systemic effect sufficient to extend murine health and lifespan. Moreover, our data show that the bone marrow niche is remodeled upon aging and we report on critical alterations of the vasculature and of the arteriolar vessels. Overall, our data supports that increasing the regenerative potential of endogenous aged somatic stem cells is possible and represents an important strategy for rejuvenating tissues and improving health- and lifespan in the elderly.

PL05-L03 | Young versus old blood—myths versus evidence-based science in a rapidly aging worldT Kanias¹¹Vitalant Research Institute, Denver, United States

Background: Advancements in medical care coupled with low birth rates in some developed countries have resulted in increasingly aging population worldwide. This is relevant to transfusion medicine because age is a biological variable that contributes to inter-donor variability in the quality of blood components including red blood cell (RBC) concentrates.

Objectives: (1) To review the global changes in blood donor demographics related to age. (2) To discuss the current understanding of age-related risk factors for donation. (3) To discuss age-related differences in blood product quality. (4) To review donor-recipient linkage studies that evaluated blood transfusions from younger versus older donors.

Methods: We reviewed data and outcomes from studies that evaluated trends in donor age demographics, linked donor age with stored RBC/plasma quality, or with RBC transfusion effectiveness/outcomes measured by hemoglobin increments or adverse events.

Results: Table 1 summarizes key findings from cumulative studies.

Conclusions: Average blood donor age is increasing worldwide, while younger blood donor recruitment remains challenging. Donor age may vary among different ethnic groups and may reflect cultural differences and recent immigration of potentially younger donors in the US. Aging is associated with changes in RBC biology and characteristics, which may impact RBC product quality. Understanding the clinical effects of donor age on transfusion outcomes is an essential step towards precision transfusion medicine.

PL05-L03 Table 1: Studies that demonstrated the effect of age and aging on blood donor demographics, blood product quality, and transfusion outcomes.

Objective #	Reported findings	Sources
1. Trends in donor age demographics	Median donor age has gradually increased (1–4 years) in last two decades. This observation is coupled with the global increase in the proportion of people over 60 years. COVID-19 further contributed to the age dichotomy resulting in increased (40.7%) donations from donors ≥65 years and significant losses (–60.7%) of donors <18 years in the US. Ethnicity-associations with mean American donor age. European White (~47 years), African American (~40 years), Asian (~35 years), Hispanic (~33 years).	PMID: 18005327, 28871601, 29034365. America's Blood Centers (2019–2021 survey). PMID: 29034365
2. Age-related risk factors for donation	Teenage donors: Highest risk of phlebotomy reactions iron deficiency, cognitive effects Senior donors: Anemia, iron deficiency.	PMID: 30633813, 28258073, 34085375, 25254019
3. Age effects on blood product quality	Stored RBCs: Aging is associated with significant changes in RBC antioxidant capacity, hemolysis, and estimated median densities. RBC units from teenage donors contain smaller RBCs, higher RBC concentration, and increased RBC expression of enzymes required for 2,3-diphosphoglycerate synthesis. Plasma: Rejuvenation studies in mice created a 'gold rush' for young donors plasma. No strong evidence for significant age differences in the quality of plasma products.	PMID: 37387566, 38420746, 38418415, 32241843, 29034365 PMID: 31572771, 21752027
4. Donor age associations with RBC transfusion outcomes	There are conflicting data regarding the effects of donor age on RBC transfusion effectiveness or posttransfusion complications including morbidity, with possible associations between younger donor age and RBC posttransfusion reactions.	PMID: 34477851, 31350268, 28437543

Academy day session

Blood banking during natural disaster

AD01-L01 | Weathering stormy times in the blood bank—A different perspective on Island life

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Hospital blood banks require reliable and timely deliveries of blood components and plasma products to support patient transfusion needs. Extreme weather events may cause temporary shortages of blood and can interfere with blood supplier deliveries, particularly for hospitals located in remote or hard to access areas. Vancouver Island is the largest island on the west coast of North America, covering 31,285 square kilometers and hosts a population of approximately 900,000. Thirteen hospitals across the island have on-site blood inventories and are supplied from a central blood supplier on the mainland. Given their vulnerability to blood delivery suspensions during extreme weather, Vancouver Island hospitals have developed strategies and contingency measures to ensure that patient transfusion needs can be met during these times.

AD01-L02 | Blood banking in Jakarta blood centre during the flooding

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Indonesia is an archipelago country and has a tropical climate, with two distinct monsoonal wet and dry seasons. Flooding and earthquake are the two major disasters. Based on national disaster management authority (BNPB) reports in 2022, Jakarta as Indonesia's capital city included in the high-risk group of flooding with the water level height 10–400 cm whenever the rain season comes usually in January and February because Jakarta is a lowland with the average height of 8 m below the sea level. Jakarta also located on the seafloor, so when the sea level rise due to global warming, the likelihood of flooding is increasing. The Jakarta government has made efforts to reduce the affected areas by flooding. The blood centres should also prepare an emergency and contingency plan to assure that the blood system could work during the flooding. According to WHO Guidance on ensuring a sufficient supply of safe blood and blood components during emergencies, we did the risk assessment. The flood would block the road which risks cancellation of blood donation activity in the affected area and no donor came to the blood centre. The further impact was the shortage of blood stock if it lasted more than 2 days. Previously the fixed site for donating blood only one in Jakarta, which

was located at central Jakarta. Although this site was not affected with flood but most of the donors lived in suburb of Jakarta. So, one of the mitigations was bringing the blood collection centre closer to the donors. We established a blood collection centres in each district of Jakarta province. The second mitigations, increasing the blood donor activity from 1000 to 1200 donations per day to 1400–1500 per day since October until December 2023 to anticipate the shortage of blood stock during flood in January and February 2024. The third mitigations about blood stock management in blood bank hospital. If they ran out of blood stock, they should have minimum one unit of each blood group for emergency cases and prioritize it while our team asked or visited the blood donors to donate their blood. We also had a good networking with other blood centres around Jakarta to provide blood to the hospital blood bank if the flood in uncontrolled. Other risks were disrupted supply of critical materials and utilities. So, we also secured and ordered additional stock of critical materials. We also regularly checked the backup of electricity and water supply. For the staffing, in this situation, they were ready to carry out the task of their friends who trapped in the flood. With these strategies, we could perform business as usual in February 2024 even the flood reduced the number of donations to only 25% of the target donations but we still had enough blood stocks and no shortage of blood stock. Hospitals still could deliver blood on time to patients. Of course, these strategies needed support from government, societies and clinicians.

AD01-L03 | Iceland Vulcano

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Challenges: Iceland is an active volcano island and it has shown more activity in recent years. However, it is not only the risk of a volcano eruptions that can endanger blood supply in the country. It is also the unique and isolated geographical location of Iceland, which is located in the middle of the North-Atlantic Ocean, extreme weather conditions and earthquakes. These are some of the major challenges we are facing in Iceland. Iceland is 103,000 km² sparsely populated island with almost 390,000 inhabitants. Majority of the people live around the capital city, Reykjavík or around 240,000. Although these numbers are not high it is important to take into account the constant increase in numbers of visitors, with over 2 million visitors in 2023.

Current activities: There is one nationwide blood bank establishment in Iceland, with two fixed collection sites, one mobile collection unit and one production site. Approximately 11 thousand whole blood units are collected annually from volunteer and non-remunerated donors. The Blood Bank aims to be self-sufficient regarding red blood cells, plasma and platelets for patient transfusions. The Icelandic Blood Bank aims to secure sustainable blood services and meet the unknown demand and fluctuations to secure sufficient blood supply. To secure accessibility of blood supply in rural areas small emergency

stocks of red blood cells are located in 7 different hospitals around Iceland. An effective monitoring system is extremely important, that includes daily meetings where representatives from all relevant departments of The Blood Bank, analyse current stock levels, daily usage of blood components, human resources, and other affecting factors. Appropriate actions are taken, if needed, according to predefined blood supply levels and resources. The Blood Bank has in place contingency and emergency preparedness plans available for different kind of scenarios. Emphasis is placed on having a good cooperation with stakeholders. This is achieved by using tools from the quality management system, for example, risk assessments and supplier evaluation. It is a well-known risk to have one production site and even though we have good relationships with suppliers for equipment, it is important to maintain good cooperation with other European countries. Implementation of the ISBT-128 standards simplifies in need of an emergency the process of purchasing blood components from other European Blood Banks.

Summary: It is important to review and update contingency and emergency preparedness plans on regular intervals with active participation of stakeholders. Despite the challenges we are facing, due to the size and location of the island, it can also be beneficial. For example, the communication channels with all stakeholders are short due to the proximity. The Icelandic Blood Bank has implemented international standards to facilitate cooperation. Maintaining good relationship with other blood establishments in Europe is also an important factor.

Academy day session

Controversies in prenatal serological testing

AD02-L01 | Is anti-D prophylaxis required for events before 12 weeks gestation?

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Background: The risk of maternal exposure to the fetal D blood group antigen increases with gestation as both the fetal blood volume and the risk of feto-maternal haemorrhage (FMH) increase. Limited evidence exists to quantify the risk of alloimmunisation against D in early pregnancy. This has resulted in variation in recommendations internationally.

Aims: To review current international practice and the evidence base for offering anti-D immunoglobulin (anti-DIg) in the first 12 weeks of pregnancy. Focussing on gaps in knowledge and discrepancies in approach between different guideline writing groups.

AD02-L01—Table 1: Recommendations from a selection of guidelines relating to offering anti-D following termination / abortion

Anti-D use prior to 12 weeks	Medical termination	Surgical termination
BSH (CSH) 2014	Yes (any gestation)	Yes (any gestation)
SOGC 2018	Yes	Yes
WHO 2022	No	No
NICE 2023	Yes (but not <10 + 0 weeks)	Yes (including <10 + 0 weeks)
ACOG 2023 (& 2018)	Yes (any gestation)	Yes (any gestation)

Methods: A literature search was performed to identify research, audit, guidelines and related evidence which referred to the use of anti-DIg in the first 12 weeks of pregnancy. Published guidelines were reviewed to identify the strength of evidence behind recommendations and potential reasons for any discrepancies between different nations / professional groups.

Results: Published guidelines consistently and appropriately described the low level of evidence behind any recommendations made. Recommendations regarding termination of pregnancy in a selection of published guidelines are shown in table 1.

Conclusions: Similar, low levels of evidence were quoted in all guidance. Differences in recommendation appeared to be influenced by The methods used for consultation / achieving consensus, The perceived simplicity of the recommendations in terms of decision points. The social context in which care is being provided. Individual practice will be governed by the guidance applied where the practitioner lives. A simple, blanket approach based on gestation is easiest to understand from a health care professional view point but social and logistical factors are particularly important to pregnant women. The oldest two published guidelines quoted in table 1 are in the process of being rewritten which have different approaches to achieving consensus. Ongoing international collaboration, improving the evidence where possible and moving towards increased consistency will hopefully simplify this area of practice.

AD02-L02 | Controversies in laboratory management of a pregnancy with anti-K

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Background: There is controversy on critical cut-off values of laboratory testing to select pregnancies at increased risk for anti-Kell (K) mediated HDFN (hemolytic disease of the fetus and newborn). Without early detection and treatment, Anti-K mediated HDFN may result in progressive fetal anemia, fetal hydrops, asphyxia and perinatal death.

Objective: The value of repeated anti-K titer determination and biological activity measurement using the antibody-dependent cellular

cytotoxicity (ADCC) test determination in the management of pregnancies at risk for anti-K mediated HDFN will be discussed. In addition, the latest international research data and the controversies in the various national guidelines are explained.

Content: First, the pathophysiology of K-mediated HDFN will be described. Daily practice and the controversies of this practice are discussed based on case studies and a large Dutch retrospective cohort study of pregnancies with anti-K and a K-positive fetus, identified from January 1999 until April 2015. The applicability of the updated version of the Dutch guideline with regard to a pregnancy with anti-K will be discussed in the light of organization of care and availability of resources.

Discussion: The organization of screening, detection and monitoring of a pregnancy with anti-K depends on the prevalence of K-alloimmunization in a country. It also depends on quality and possibilities to diagnose fetus or neonate with anemia using ultrasound and clinical assessment. Considerations that are made include the use of a fetal K-antigen typing, critical cut-off value of the titer and options for measuring biological activity.

AD02-L03 | Is mid pregnancy and pre delivery type and screen necessary?

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Background: During pregnancy, the group and screen (G&S) is an important test performed during the first trimester to identify clinically significant antibodies (CSA). Typically, this test is performed at the diagnosis of pregnancy, repeated at 28 weeks, and sometimes prior to delivery. Practices differ internationally and existing guidelines on repeat G&S testing at 28 weeks vary, including different recommendations for RhD negative and positive patients. The objectives of this test are to identify CSA that may lead to hemolytic disease of the fetus/newborn (HDFN), to determine an RhD negative individual's eligibility for anti-D immunoglobulin (RhIg) and/or to ensure availability of red blood cells (RBCs) at delivery. The role of G&S for routine low-risk pregnancies will be discussed.

28-week Group and Screen: The historical reasoning for repeating the G&S at 28 weeks is twofold; (1) To reassess for alloanti-D following the first-trimester test; and (2) to identify whether a patient has developed a new, non-RhD CSA since the time of initial testing. If a new CSA is identified, follow up titration testing and potential referral to maternal fetal medicine would be warranted. Additionally, if a new alloanti-D is identified, RhIg administration for alloimmunization prevention is no longer appropriate. The prevalence of CSA during pregnancy, is variable depending on availability of RhIg and is <1% where robust prophylaxis is available. This estimate includes older literature

when testing was performed by saline indirect antiglobulin tube tests. With current sensitive testing methodologies, most CSAs should be detectable at the first trimester screen.

Repeat Group and Screen at Delivery: A G&S is sometimes repeated pre-delivery due to concern about the possible need for a peripartum transfusion. Literature suggests that peripartum transfusion rates average 1%–2%. A G&S at delivery is only indicated if the likelihood of a transfusion for either the patient or neonate is high, if the patient did not undergo the initial, routine G&S, or if the patient may need additional blood bank testing or specialized blood products (i.e., known alloantibodies, complex transfusion needs). Universal, prophylactic G&S testing is not necessary for low-risk patients and a risk assessment tool may guide appropriate G&S testing at delivery.

Benefits of Reducing Unnecessary Group and Screen: If 28-week and delivery G&S were reserved for patients at risk for alloimmunization and transfusion, respectively, the reduced testing could lead to time and cost savings for the laboratory and health care's system. During this presentation, the pros and cons of performing the group and screen test at 28 weeks and at delivery will be discussed.

Academy day session

Donors & donations

AD03-L01 | Towards a data-driven donor-deferral policy - improving donor health and eradicating on-site low Hb deferrals globally

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On-site donor deferral for low Hb has been a concern of blood banks for many years. Not only does it imply the immediate loss of a donation, but it also results in a less efficient use of testing materials and staff time. Most importantly, however, it demotivates donors to return for donation and therefore leads to loss of future blood donations as well. The rationale behind the deferral threshold for donors is to ensure sufficient Hb content in blood products and to prevent donors from donating whilst having too low Hb levels, ensuring sufficient recovery of the donor from donation. However, from the substantial amount of data collected on subsequent Hb levels measured in blood donors it can be concluded that the current donor deferral policy is not only failing to protect donors from becoming iron deficient, but it also shows that it is insufficiently protecting donors with low Hb levels from donating. When looking at the Hb levels of donors between subsequent donation there are two interesting observations to be made: (1) the average change in Hb over time (expressed on a logarithmic scale) is linear, and therefore easily predictable; and (2) the level of variation in measured Hb-levels between subsequent donations is a multitude of the actual change in Hb over time. Note that the latter is true for both venous and fingerprick measurements. The

fact that we use a single, highly variable Hb measurement to decide on a donor's eligibility causes (random) measurement variability to be the primary underlying mechanism for on-site donor deferral. As variability is inherent to any measurement, and particularly when measuring biomarkers, within the field of laboratory medicine the term "reference change value" (RCV) is commonly used. The RCV indicates whether the difference between two subsequent measurements is large enough to conclude that this difference can be attributed to a change in the biomarker considered or whether it is due to (random) variation. The RCV consists of both biological (biomarker dependent) and (pre)analytical (sampling and measurement method dependent) contributors. Therefore, instead of applying a fixed threshold for the interpretation of an outcome that is variable, we should interpret this outcome relative to a reference value. A simple but meaningful reference value for Hb would be the average Hb level of past donations. It can be shown that even when using the average historical Hb level as a reference, a substantial reduction in the number of on-site low Hb deferrals may be achieved. There is a second motivation for using a donor's historical average Hb level as a reference: routinely collected Hb and ferritin measurement data show that there is a striking association between low ferritin levels and the drop in Hb relative to the donor's homeostatic Hb level. This means that by monitoring structural changes in Hb levels in donors over time (instead of applying one deferral threshold for all donors) we may prevent donors from becoming iron deficient from subsequent donations. Systematic analysis of routinely collected biomarker data in blood donors provides extremely useful insights that can be used to guide and shape future donor deferral- and donor management strategies. Further research as well as discussion with various stakeholders is needed to ensure that donor safety is guaranteed and that alternative approaches are practicable and supported by the blood banking community at large.

AD03-L02 | Association of blood donor characteristics with blood component quality and storage properties

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Background: Blood component quality is of utmost importance for effective transfusion outcome. Modern processing techniques have standardized blood component preparation with better process control. However, inherent differences in donor characteristics may still introduce product variability.

Aim: To determine the association of blood donor characteristics with packed red blood cell (PRBC) and platelet concentrate quality and storage properties

Methods: A series ($n = 4$) of prospective cross-sectional studies were conducted at our center. Blood donor characteristics were analyzed for correlation with total haemoglobin (Hb) content of PRBCs and leucoreduced PRBCs in two separate studies. The association of donor characteristics, pre-donation Hb and uric acid (UA) levels of blood

donors with PRBC storage properties (potassium, lactate dehydrogenase (LDH), plasma Hb and percentage hemolysis) was analyzed in another study. An association between donor characteristics and platelet storage properties (platelet count, mean platelet volume, pH, glucose and lactate levels) was also determined in another separate study.

Results: The mean total Hb content was found to be significantly higher in PRBCs prepared from male (48 g/unit vs 41.8 g/unit, $p \leq 0.001$) and non-vegetarian (48.2 g/unit vs. 46.7 g/unit, $p = 0.04$) blood donors. A strong positive correlation was observed between donor pre-donation Hb and total Hb content of both PRBC and leucoreduced PRBC ($r = 1.000$; $p = 0.0001$ both). When analyzed for the association of blood donor characteristics with storage properties of PRBCs, significantly higher mean hemolysis was observed in PRBCs prepared from donors with UA levels ≤ 6 mg/dL on day 35 of storage (0.22 ± 0.11 vs 0.18 ± 0.07 , $p = 0.03$). A significant positive correlation was also observed between donor UA levels and LDH levels of PRBCs on day 35 of storage (β coefficient 715.52, $p = 0.003$) on multiple regression analysis. In another study where the association between donor characteristics and in vitro storage platelet properties was analyzed, platelets from donors with high HbA_{1c} levels ($\geq 6\%$) had lower median pH (7.31 vs. 7.37, $p = 0.024$) and higher median glucose levels on day 1 of storage (358 mmol L^{-1} vs. 311 mmol L^{-1} , $p = 0.0001$) while higher median lactate levels were observed throughout the platelet storage period (Day 1: 7 mmol L^{-1} vs. 5.7 mmol L^{-1} , $p = 0.037$ and Day 5: 16 mmol L^{-1} vs. 12.2 mmol L^{-1} , $p = 0.032$).

Conclusions: Our studies demonstrate that blood donor characteristics do impact the quality and storage properties of PRBCs and platelet concentrates. These findings emphasize the importance of considering donor characteristics in optimizing blood component therapy and improving transfusion outcome.

AD03-L03 | Data driven insights on strategic youth-based blood donor programmes for sustainable growth

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Background: Youth-based blood donor programmes are vital for blood availability and safety in low-medium income countries (LMICs) like Zimbabwe. Increasingly, developed countries are also focusing on youth donation for stable contributions to blood adequacy and safety. Data-driven insights are crucial for structuring and monitoring these programmes effectively. In Zimbabwe, the blood donor programme consists of a three-layered approach involving in-school youth programs (peer promoters), out-of-school initiatives (Pledge 25, Club 25), and an adult segment. This paper provides insights into the importance of data analytics in understanding the dynamics of managing these programmes, including donor recruitment algorithms, indicators for retention, loss to follow-up, and sustainability.

Aims: This study aims to share integral insights from data to inform the dynamics of youth-based blood donor programmes.

Methods: Using Zimbabwe as a case study, we explore the structured blood donor programme, particularly focusing on vibrant youth participation. The segments studied include peer promoters and the Pledge 25 programme. We examine the governance structure of these youth initiatives and the data analytics used to monitor, evaluate, and learn from their strategic implementation.

Results: Annual blood collections in Zimbabwe average 80,000 units, with approximately 70% from youths below 25 years old. In 1994, the National Blood Service Zimbabwe (NBSZ) introduced two youth-led initiatives: peer promoters and Pledge 25. Stakeholder engagement was crucial, especially for obtaining blood in school settings. Blood collection and safety analytics revealed the effectiveness and safety of collecting blood through in-school programmes. The peer promoters' programme emphasized peer influence and safety parameters, leading to its success. An out-of-school programme focused on youth retention strategies, with emphasis on the appropriate robust analytics. Youth-led programmes empowered young donors and bridged donation gaps during school holidays with provision of appropriate incentives and recognition system. A transitioning framework to the adult donor segment was established. Repeat donations averaged 60%, with year-on-year donor retention around 40% (all) and 70% for Pledge 25 over five years. The Blood Supply Management Status (BSMS) and demand/supply indicators have remained satisfactory, reflecting the success and sustainability of the youth program. Residual risk estimates for HIV, HBV, and HCV were satisfactory, although HBV burden remained high over a ten-year study. Collaborations, youth empowerment projects, and social media analytics have all been positive contributors.

Summary/Conclusions: Designing effective youth blood donor programmes requires defined data analytics for continued monitoring and sustainability assessments. Youth-led governance structures, when empowered with key sustainability indicators, effectively manage these programs for long-term success and blood supply stability.

Academy day session

Update on immunohaematology

AD04-L01 | Revisiting soluble blood group proteins

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The detection and identification of antibodies to red blood cell (RBC) antigens is critical in patient pre and post transfusion laboratory testing. Conventional serological testing methods for antibody identification can be challenging especially for routine, non-specialised immunohaematology laboratories and the required antibody identification could be problematic and could ultimately delay transfusion. The presence of an

autoantibody, multiple alloantibodies, or a suspected antibody to a high-prevalence antigen in patient samples pose the greatest challenges in such cases. A common tool in our immunohaematology toolbox to aid in antibody resolution for many years has been the use of inhibition or neutralization studies. Blood group substances such as ABO in soluble form in body fluids or secretions have been known for nearly 100 years. Inhibition tests using ABH substances or body fluids for ABO and Lewis were done as early as the 1940's. Inhibition with other soluble antigens such as P1 in hydatid cyst fluid or Sd^a in urine followed in the 1950s and 1970s. Since then inhibition studies using soluble antigens, some being commercially available, have been widely used in serological testing. In the 1990s soluble CR1 made by recombinant DNA technology was used successfully to identify Knops blood group system antibodies. Since then, soluble blood group proteins (rBGPs) have been shown to be effective in haemagglutination inhibition assays used in the identification of red cell antibodies. Of note, the use of a rBGP aided in the classification of the new blood group antigen CD59 in 2014. rBGPs can be expressed in either prokaryotic or eukaryotic vector systems although the latter is often preferred because it allows for posttranslational modification such as glycosylation or disulphide bonding that might be required for the correct folding for the formation of the three-dimensional epitope. Today, there are roughly 18 soluble rBGPs carrying 92 distinct blood group antigens available for inhibition studies in serological testing. These rBGPs can interact with serum or plasma in haemagglutination testing assays in a wide application from traditional tube and column agglutination technology (CAT) testing to solid-phase red cell adherence assay (SPRCA)-based methods. The currently available rBGPs have been extensively tested and validated and have been proven to be successful in aiding in identification of antibodies to common high-prevalence antigens expressed on single pass glycoproteins or glycosylphosphatidylinositol-linked (GPI) proteins. A more recent application of rBGPs could be mitigation of plasma interference from anti-CD38 or anti-CD47 therapy. As anti-CD38 or anti-CD47 interference are drug specific, use of sCD38 or sCD47 could be advantageous over the development or use of anti-idiotypic antibodies as these are drug-specific and therefore rBGPs would potentially have a wider scope of use. The first section of this presentation will provide a background on rBGPs and provide an update on the currently available rBGPs. The second section of the presentation will walk through the potential application of rBGPs in the immunohaematology laboratory and the fundamentals of their use in serological testing. The third section of this presentation will include the presentation of case studies where the use of rBGPs was pivotal in aiding in antibody resolution as a supplement to conventional RBC panel-based antibody identification methods.

AD04-L02 | Practical and clinical implications of Del phenotypes

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Del (D-eluate) phenotype was first identified in 1984 in serological D-negative individuals through serologically adsorption and elution tests. It is a very weak form of D that cannot form effective agglutination in routine serological RhD typing tests, consequently, to be commonly typed as a primary D-negative phenotype. Approximately 9%–33% of the individuals with a serologic D– phenotype in Chinese, Korean, Japanese, Thai, Myanmar, and Indonesian populations carry a Del phenotype but not a true D– phenotype. Although more than 40 distinct genetic variants underlying the DEL phenotype have been recognized thus far, *RHD*01EL.01* (*RHD*1227A*) was found predominantly in Del individuals (>95%) from these populations in East and Southeast Asian regions. Thus, it was termed Asian-type DEL in 2010. In China, the USA and the European Union (EU), approximately 96%, 77% and 9% of the Del population, respectively, carry the Asian-type DEL allele representing approximately 1.7 million people in China, 90,000 in the USA, and 10,000 in the EU. From the recipient's perspective, it was first described that Asian-type DEL recipients may not develop alloanti-D after exposure to RhD-positive (D+) RBCs as early as 2006. Some evidence, namely, that no alloanti-D immunization was detected in the pregnant women with Asian-type DEL after carrying a D+ fetus, and in few Asian-type DEL cases post D+ RBC transfusion, supported this speculation. In 2023, several lines of strong clinical, epidemiological and molecular functional evidence that individuals with Asian-type DEL do not produce alloanti-D when exposed to D+ RBCs during pregnancy or transfusion were obtained to support routine D+ blood for transfusions in all Asian-type DEL recipients (Blood. 2023;141(17):2141-2150). Besides, RhIG immunoprophylaxis in women with Asian-type DEL can be dropped, and that anti-D immunization monitoring during any Asian-type DEL pregnancy can be stopped. For the donor's perspective, Asian-type DEL RBCs can elicit primary or secondary alloanti-D immunization in true D– recipients, albeit rarely with fewer than twenty case reports to date. If we label DEL RBCs as D+ RBCs, as is practiced by several blood services in Europe, the US, and Brazil, all alloanti-D immunization possibly caused by Asian-type DEL RBCs will be avoided. However, for East and Southeast countries, this expedient approach would absolutely exacerbate the shortage of D– blood due to low frequency distribution of D-negative phenotype (0.3%–0.4%). Currently, to avoid transfusion of Asian-type Del RBCs to the recipients with alloanti-D and the women with child-bearing age may be a feasible and transitional strategy. Finally, some further studies about Asian-type DEL, such as the distribution of Asian-type DEL in the unreported Southeast Asian populations and the molecular mechanism of the more commonly identified specific weak D phenotype in Asian-type DEL allele carriers, are all needed in the future.

AD04-L03 | A thousand and one—practical strategies for finding rare blood donors

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Usually, red cell units are available from the blood centre when needed. But if the patient has a rare blood group, an antibody to a high incidence antigen or multiple common alloantibodies, there will be challenges if you have not prepared in advance. On a practical level, rare blood is something that is not readily available when needed. In the most difficult cases, there may be less than 10 donors of a certain rare blood group registered in the entire world. Basic pheno- and genotyping together with antibody screening will recognize most of these. The most efficient approach to find donors is to recruit patients who have been identified to have a rare phenotype according to an antibody finding. Similarly, antibody findings lead to possible rare donors in antenatal samples and antibody screening of blood donors. Siblings of a rare blood group person are more likely to have the same rarity. Donor typing can be targeted to regular donors, starting from group O donors. In addition, typing can be focused on the donors from certain country of birth or ethnic background. Jk(a-b-) phenotype donors are more likely to find in Finnish, Polynesian and Japanese population than in others. It is important to carry out Rh and K phenotyping at large scale to meet the requirements of patients with common blood groups and blood group antibodies; luckily the extremely rare null phenotypes are found simultaneously. With Rh and K phenotyping including k typing from K+ donors, it is possible to find rarities like k- and Rh_{null}. Extended typing including Fy, Ss, Jk might find Fy(a-b-), Jk(a-b-) and S-s- donors. It can be done either by pheno- or genotyping. Phenotyping can be further extended to antigens that have antisera available like Kpa and Lua, and when finding antigen positive donors, test for Kpb and Lub antigens. The Finnish Red Cross Blood Service FRCBS typing process is shown in table 1. Furthermore,

FRCBS operates a biobank containing genome data from approximately 58,000 blood donors. By screening these donors for gene markers indicating rare blood groups, have been identified several rare donors, such as Vel-, Kpb-, and Lub-. In the perfect scenario, phenotyping will be confirmed by genotyping and vice versa. Donors typed by both methods are the most valuable, especially when exporting units. The genetic background of almost all rare blood groups is known. Commercial genotyping platforms include several rarities. Yet, null phenotypes can be found by serological methods and routine genotyping kits do not recognize all variants. Finally, there is a need for database where to find one in a thousand donor, when needed. When rare blood is not available nationally, the ISBT International Rare Donor Panel, managed by the International Blood Group Reference Laboratory maintains rare blood lists provided by the ISBT WP rare donor members to ensure that rare blood is available worldwide to patients who need it.

AD04-L03 – Table 1. Finnish Red Cross Blood Service typing process.

Method	Antigen	Donors	Tests / month
Antibody screening		1. time donors, possible immunized	7000
Automated phenotyping	Rh K(k)	1. time donors	2772
Automated phenotyping	Jk Fy Ss M	Regular donors, A B O K- no R1R2	319
Manual phenotyping	Ula LWa/b Lsa WESa Cx Coa/b Lua/b Cw Kpa/b Wra	Part of extensively phenotyped donors	48
Genotyping	RH KEL JK FY MNS DI DO CO YT LU	Part of extensivelyphenotyped and African origin donors	54

Academy day session

Granulocyte disorders - diagnosis and transfusion therapy

AD05-L01 | HNA investigations what, why & where

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The binding epitopes for all HNAs are located on five glycoproteins and are divided into five human neutrophil antigens (HNA-1 to -5). Several polymorphisms in the coding sequence of these glycoproteins generate allelic determinants for each HNA. Incompatibilities of some HNAs during pregnancy or during transfusions or transplantations lead to immunization and thus to the formation of alloantibodies. The binding of alloantibodies to the target antigen in the fetus during pregnancy leads to activation/elimination of the antibody-labeled cells and thus to neonatal alloimmune neutropenia (NAIN). It is also known that the presence of these alloantibodies can lead to rejection of the antigen-positive organ in organ recipients. Similarly, the presence of HNA alloantibodies in transfused blood products can cause transfusion-related acute lung injury (TRALI) in antigen-positive recipients. Autoimmune neutropenia of infancy (AINI) is another clinical disorder mediated by autoantibodies against HNAs. Although it is self-limiting, AINI is very common. As biological structures, HNAs are important not only for antigenicity but also for the function of the cells expressing them. This summary is intended to provide an overview of the structure of HNAs, their antibodies and their role in various diseases.

AD05-L02 | Update on the practice of granulocyte transfusions

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Granulocyte transfusions are used to treat refractory infection in patients with profound neutropenia. There is wide variation in manufacture and clinical use worldwide. This can at least in part be attributed to the lack of conclusive evidence from clinical trials. Beyond their use in treatment of infection, pooled granulocytes have also been shown to have an immunomodulatory effect, potentially improving outcomes in children with high-risk leukaemia. Whilst there is significant uncertainty regarding the role of granulocyte transfusion, use of the component continues and potential for its development remains. Granulocytes for transfusion can be produced by apheresis; donations may be directed or non-directed and donors may be stimulated with steroids and/or

granulocyte-colony stimulating factor. An alternative component can be made by pooling buffy coats from whole blood. Logistical difficulties can arise with apheresis granulocytes including timely recruitment and testing of suitable donors. Typically, the yield of stimulated apheresis granulocytes is higher than the pooled component. However, typically patients are unable to receive apheresis granulocytes daily or for a prolonged duration. Due to relative ease of manufacture of pooled granulocytes, these are beginning to be adopted in some centres worldwide. The Resolving Infection with Neutropenia with Granulocytes (RING) study is the largest randomised controlled trial of granulocyte transfusions. There was no difference in the primary composite outcome of survival and resolution of infection between the treatment (granulocytes) and control (no granulocytes) arms. The trial was compromised both on recruitment and ability to deliver target doses. Published studies suggest that infection continues to be a significant burden in haematology patients undergoing intensive chemotherapy, and that only a small proportion of patients eligible for granulocyte transfusions receive them. Alternative approaches to traditional trials have been employed. Other challenges with granulocyte transfusion include manufacture of a high-yield component that can be stored for clinical use. Donor-derived neutrophils have short lifespans *in vitro* and require irradiation. There is risk of alloimmunisation and there are risks of fever, transfusion-related acute lung injury (TRALI) and cytomegalovirus transmission. Additional roles of granulocyte transfusions include a potential role in immune priming. In a small prospective study of children undergoing cord stem cell transplants for relapsed, refractory acute leukaemia and receiving pooled granulocytes, an early polyclonal CD8 T-cell expansion was seen with high rates of MRD (minimal residual disease) negativity (8/10). There are many avenues for development of the component; optimisation to address challenges of manufacture and deliverability may include growing neutrophils from stem cells. Further possibilities with regards utility include manipulation of neutrophils to enhance or direct their function. Granulocyte transfusions continue to be used predominantly for treatment of refractory infection in the setting of severe neutropenia. Conventional randomised trials seem unlikely to be replicated and alternative study designs need to be considered. However, all studies are limited by the shelf life and viability of current products. Future uses of granulocyte transfusions may include a role in immune priming.

AD05-L03 | Neonatal alloimmune neutropenia: The Brazilian experience

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Background: Neonatal alloimmune neutropenia (NAIN) is a potentially lethal disorder resulting from maternal alloimmunization to fetal human neutrophil antigens (HNAs). The alloantibodies most frequently involved in NAIN are against the HNA-1 and -2 systems;

AD05-L03 Table 1. Clinical and serologic profile of the NAIN cases.

NAIN Cases	Gestational age, weeks	Neutrophil count in cord blood, $\times 10^9/L$	Neonate clinical condition	Antibody specificity in mother serum
1	38.0	1.390	Thrombocytopenia ($56,000/mm^3$)	HNA-1b / HLA-I
2	26.6	1.130	RDS, PDA, Sepsis, PIH	HNA-1a / HNA-1d / HLA-I
3	30.0	1.276	None	HNA-1b / HLA-I
4	34.0	1.175	SGA	HNA-3b
5	36.4	1.280	None	HNA-3b / HLA-I/II
6	30.9	0.660	Thrombocytopenia ($82,000/mm^3$), PJ	HNA-3b / HLA-I/II
7	30.5	0.507	Pneumonia	HNA-3b
8	36.9	1.435	None	HNA-3b

Abbreviations: PIH, peri-intraventricular hemorrhage; PDA, patent ductus arteriosus; PJ, physiological jaundice; RDS, respiratory distress syndrome; SGA, small for gestational age.

however, the HNA-3, -4 and -5 systems have also been associated. Its frequency is not well established since many cases go undiagnosed, and the limited number of specialized laboratories worldwide is perhaps one of the main obstacles.

Aims: To describe the prevalence of NAIN in a Brazilian cohort by investigating the maternal-fetal HNA incompatibility, the presence of maternal HNA and HLA alloantibodies, and the clinical profile of newborns (NBs) involved in this condition.

Methods: Neonatal neutropenia, defined as neutrophil count $<1.5 \times 10^9/L$ in cord blood, was examined in samples from 10,000 unselected neonates over a two-year period, resulting in 88 neutropenic NBs and their 83 mothers. Peripheral blood samples were collected from these mothers and NBs for serologic and molecular tests. Genotyping was performed by PCR-SSP (HNA-1/-4) and PCR-RFLP (HNA-3/-5). Serologic studies were conducted using granulocyte agglutination and immunofluorescence tests (GAT and GIFT) and LABScreen-Multi-HNA-Kit (LSM) (OneLambda).

Results: Neonatal neutropenia was identified in 88/10,000 (0.9%) NBs. Genotyping revealed 53/88 (60.2%) maternal-fetal HNA incompatibilities (31.8% for HNA-1; 14.8% for HNA-3; 15.9% for HNA-4; 21.6% for HNA-5). Serologic studies revealed 31/83 (37.3%) mothers with positive results with at least one technique for anti-HNA and/or anti-HLA antibodies. The anti-HNA specificities were confirmed in eight positive cases (8/10,000; 0.08%), related to HNA-1a/-1d (1), -1b (2), and -3b (5) epitopes. Anti-HLA antibodies were detected in 18/31 mothers (11 anti-HLA class I, 7 anti-HLA class I and II). The eight NAIN cases presented variable clinical manifestations, with infections observed in two NBs, one with respiratory distress syndrome and sepsis, and the other with pneumonia (Table 1).

Conclusions: This large Brazilian study investigated antibodies against all five HNA systems in mothers of neutropenic NBs, showing that NAIN affects 1/1250 neonates. Among the HNA antibodies identified, we highlight the presence of anti-HNA-1d and anti-HNA-3b antibodies, rarely related to NAIN cases. NAIN symptoms may range from asymptomatic to serious infections, and is one of the causes of neonatal neutropenia, occurring alone or concomitantly with other causes, worsening the clinical evolution of the neonate.

Academy day session

Blood service finance 101

AD06-L01 | Understanding financial reports

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The success of any business, company or institution, whether operating on a for profit or not for profit basis, is critically dependent on a sustainable business model. Financial sustainability is a critical element for overall sustainability and the ability of management to manage finances and understand financial reports is essential for ensuring that the company is a going concern. Financial statements offer a view to the health of a company at any given time and is used by management to make operational and strategic decisions. They are a number of financial tools and reports and four reports will be discussed, with examples, in the presentation. These are the: Annual budget—The budget is a forward looking projection of revenue, expenses and potential profit/surplus or loss. The budget is prepared considering income from operations as well as expenses incurred in generating the revenue. Various internal and external factors need to be analysed when preparing the budget. For a Blood Service this includes analysis of demand for products, staff costs, operational expenses, provision for bad debts, depreciation. The financial budget is then prepared projecting the income and expenses. (1). Income statement—An important management report that summarises all income and expenses over a given period. Income statements are prepared on a monthly basis and, put simply, show how much money a company made and spent over a period of time. Management regularly review income statements to understand how well a business is doing at a point in time. Is revenue on target? Are expenses below or above target? Is the company making a surplus or loss? This information can then be used to make operational adjustments going forward. Balance Sheet—A balance sheet conveys what the company is worth or the “book value”, expressing what the company owns and owes at a fixed

point. It shows that assets, liabilities and shareholders equity of the company. Assets are things that a company owns that have value. Assets include physical property, such as plants, equipment, inventory, accounts receivable, trademarks, patents, cash and investments. (1). Liabilities are amounts of money that a company owes to others. This can include all kinds of obligations, like money owing to a bank, rent for use of a building and money owed to suppliers. (2). Shareholders' equity is sometimes called capital or net worth. It's the money that would be left if a company sold all of its assets and paid off all of its liabilities. This leftover money belongs to the shareholders, or the owners, of the company. For a not for profit company this is the equity that could be transferred to another entity. (1). Cash Flow Statement—A cash flow statement provides a detailed picture of what happened to a company's cash during a specified period, usually a month or quarter. It demonstrates an organization's ability to operate in the short and long term, based on how much cash is flowing into and out of the business. Understanding a cash flow statement is important to ensure that the company is able to meet its payment commitments to staff and creditors.

AD06-L02 | Economics in the blood supply chain

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The Sanquin Blood Establishment example

Mission statement Sanquin Blood Supply:

Together with the donor, we ensure better life for patients.

From the perspective of an economist the Sanquin goal is to realise low cost levels and invest the savings in R&D to further optimize the blood supply chain.

Financing the Sanquin Blood Services

The external budget of Sanquin Blood Service and tariffs for blood components are approved by the MoH. The goal of the budget model is to stimulate effectivity and efficiency while guaranteeing a safe blood supply in the Netherlands. The financial model should support the mission of Sanquin and create incentives for business improvement (quality, effectivity, efficiency and R&D). 50% of the budget is fixed and 50% of the budget is variable based on the level of activities. New safety measures have to be approved by MoH. After approval extra budget becomes available.

Continuous business improvement

The role of economics and setting targets

The goal of the budget model inside a blood service is identical: to guarantee high quality and service levels; to stimulate effectivity and efficiency; and create resources for R&D. Managers are responsible for those costs that they can influence. Fixed costs can be reduced by reorganisations like consolidations or process improvements. Starting in 1998 with 22 Blood services the consolidations resulted in 2015 in 1 consolidated blood establishment in Amsterdam, 1 processing unit in Nijmegen and many fixed donor centers and mobile teams. Goal of these consolidations was to optimize and standardize all processes and create an optimal use of the economies of scale. Variable costs

can be reduced by optimal allocation of staff, equipment and disposables. Input was used from the EBA Benchmarking project, using Lean Management and the implementation of the results of scientific research in Sanquin. Targets for business improvement are set and the internal budgeting system created incentives.

Conclusion: Process improvement programmes should never stop, Participate active in benchmarking, use Lean management and Pull systems, optimize the supply chain, Invest in donor and transfusion Research.

AD06-L03 | Finance considerations and challenges in my blood service

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Introduction

The Blood Services Group (BSG) of the Health Sciences Authority plays a critical role in running the National Blood Programme and ensuring that the nation's blood supply meets the healthcare needs of Singapore's population.

Financial Considerations and Challenges

To maintain this crucial service, BSG faces multifaceted financial considerations and challenges. Understanding and effectively addressing these challenges is imperative for the sustainability of the blood supply. One major consideration is the cost of blood collection, testing, and processing. This encompasses various operational expenses such as staff salaries, equipment, maintenance, and laboratory facility costs. Balancing quality and safety while managing these costs is essential. Additionally, maintaining the cold chain for blood storage and transportation requires significant investment in refrigeration systems, cold rooms, and logistics. Another challenge is donor recruitment and retention, which involves costs for public awareness campaigns, educational programs, and recognition schemes necessary to attract and retain voluntary, non-remunerated blood donors. Additionally, stringent quality assurance and compliance measures, including testing for infectious diseases, necessitate investments in audits, inspections, and quality management systems. Staff training and retention also pose financial challenges, requiring continuous investment in education and development programs. Balancing inventory levels to meet demand without excess or shortage is another financial consideration. This requires investments in technology, storage facilities and logistics to optimize inventory management. Financial sustainability is a persistent challenge, involving covering operational costs while ensuring the affordability of blood products. Keeping pace with technological advancements and preparing for emergencies, such as pandemics, also requires financial resources.

Strategies for Addressing Financial Challenges

To navigate these financial challenges effectively, strategic planning and budgeting are crucial. Cost optimization and efficiency measures can help identify areas for improvement and cost-saving opportunities. Enhanced donor recruitment and retention efforts, partnership with the Singapore Red Cross to leverage the latter's experience and strengths as a charity organisation, enabling more cost-effective blood donor

recruitment and retention efforts, and investments in technology and infrastructure, are also essential. Continuous quality improvement, and staff development are crucial. Utilizing data analytics for decision-making, conducting risk assessments, adopting a risk-based decision-making framework for blood safety, and maintaining transparent financial reporting are important measures to ensure financial sustainability.

Conclusion

By implementing these strategies, BSG can ensure financial sustainability while fulfilling its critical mission of providing safe and adequate blood products for patient care.

Academy day session

Social media in transfusion medicine

AD07-L01 | Connecting with a professional community on Instagram and other formats

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Introduction: Social media, such as Instagram, Facebook, X and LinkedIn, has become powerful tools in transfusion medicine. These platforms allow users to share knowledge, engage a community, advocate for change, or develop professionally. Through collaborative efforts between ISBT Marketing and the Young Professionals Council (YPC), the @isbtinternational Instagram currently has 4136 followers, mainly comprising of those in the age groups of 25 to 35 years old (42.4%) and 35 to 44 years (33.4%). The Facebook audience is similar to Instagram with 48.7% aged between 24 and 35 years old. To help create a community among this majority age group, the hashtag #ISBTYoungBlood was used as a tool to collate posts from young professionals in transfusion medicine across multiple platforms. Our aim is to continue expanding the ISBT community by engaging with young professionals in transfusion medicine from around the world through social media.

Methods: Posts were created collaboratively between ISBT Marketing and YPC members, occasionally featuring colleagues or other members of the ISBT community. Regular content planned for the posting schedule, included article spotlights, special planned events and promotional posts for live journal clubs. For Congresses, WhatsApp groups were created, and Instagram content, such as a post, a reel, stories or reposts, were uploaded on a day-to-day basis to increase engagement, attendance, and feedback for certain events.

Results: The hashtag #ISBTYoungBlood has been used in 315 posts on Instagram, 59 posts on Twitter and 23 posts on LinkedIn. The most popular post based on the number of Instagram likes was a static post with three group photos of young professionals during ISBT Gothenburg 2023 (246 likes, 27 comments, 4 saves). Following this post in popularity

was a “Did You Know?” Article (211 likes, 72 comments, 37 saves), a reel showing highlights from the Legendary Leadership workshop with Judith Chapman at ISBT Gothenburg 2023 (131 likes, 5 comments, 3 saves), an article highlighting the discovery of a new antigen in the Rh-associated glycoprotein blood group system (129 likes, 67 comments, 17 saves) and an interview about tackling blood shortages during the COVID-19 pandemic (116 likes, 31 comments, 17 saves). For ISBT Gothenburg 2023, the WhatsApp group allowed young professionals to stay up to date on events, raise queries/issues and plan social gatherings. There were 82 Instagram stories posted during ISBT Gothenburg with a reach ranging from 248 to 407 accounts. In 2024 so far, the YPC has contributed to five posts for an Article Spotlight, International Day of Girls and Women in Science and sharing of a recent YPC publication on blood supply and demand via a poll.

Conclusion: Social media metrics from ISBT accounts, primarily those from Instagram, has revealed there is an active young professionals community who engage in posts about journal articles, congress updates and career experiences. This collaborative initiative with ISBT Marketing provides opportunities for young professionals to showcase their skills, learn more about transfusion, connect with a community and develop professionally as young leaders in the field.

Acknowledgements: A big thanks to all those who have been involved in the ISBT community of social media so far. I am grateful to ISBT Marketing for helping with the preparation of this abstract, including the social media metrics reported.

AD07-L02 | Influencing people to do good—the role of social media in donor behaviour

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Background: Social media have shown great potential for producing significant changes in behavior and have become the cornerstone for many public health communication efforts. The nonprofit sector—including blood collection agencies—has adopted social media to aid their cause and reach their goals, that is, recruiting, engaging, and binding donors. However, despite the tremendous impact of social media on society and its promising role for donor recruitment and retention, it has been overlooked in blood donor research.

Aims: This presentation will give an overview of our recent research with and about social media as a tool and resource for understanding, explaining, predicting, and influencing donor behaviour.

Methods: First, I showcase an inductive computational method to make sense of vast amounts of dynamic unstructured blood donation text data that exists on social media. Second, I present experimental evidence on how social media interaction with new donors influences their donation attitudes and behaviour. Lastly, I present qualitative interview data from 22 communication and marketing professionals across blood banks in 13 countries on how they use social media in their work.

Results: First, we applied structural topic modeling to Dutch Facebook and Twitter-data about the topic blood donation by the general public

(2012–2018). We found 25 topics clustered in 6 distinct categories. Over time, there was a substantial reduction of messages in which donors announce their donations. On the contrary, topics that emphasize the positives of blood donation (e.g., contributing to public good), including donor identity-related topics, are rising. Second, our field randomized controlled trial offers initial evidence that regular engagement with a blood bank Facebook page can have a lasting impact on donor behavior. Lastly, our interview study showed that social media is an important component of the communication strategies employed by blood banks in Europe, used not only for engaging with current and potential donors but also for societal positioning and establishing collaborations with external entities, such as public institutions and private companies. Key platforms used by blood banks include traditional ones such as Facebook and LinkedIn, alongside newer ones like TikTok and Spotify.

Summary/Conclusion: To make optimal use of social media for information, recruitment and retention campaigns, blood collection agencies should recognize the turbulent and dynamic environment of social media. Monitoring public opinions about blood donation can help blood collection agencies make strategic choices and utilize social media more effectively. Engaging with (potential donors) through social media channels can impact donor attitudes, stimulate donation, and contribute to the reputation of blood banks.

AD07-L03 | Blood cells and blue ticks in Indian blood banking

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The landscape of human behavior and communication is swiftly transforming with the advent of information technology. In India, instant messenger applications (IMAs) have transcended their traditional role, becoming an integral part of daily life. Notably, their significance extends beyond personal interactions, as evidenced by their burgeoning role in medical education and clinical teaching, as explored by Shrivastava SR et. al. in 2024. IMAs have become indispensable tools for the blood banking community in India. Their application has evolved significantly, with blood centers increasingly relying on them for seamless communication among clinical teams, hospitals, and within blood centers. Staff members utilize IMAs to coordinate daily activities, and these platforms offer features such as chat groups that facilitate connections, communication, and the recruitment and retention of blood donors. Furthermore, IMAs play a pivotal role in scheduling donation appointments and locating rare blood group donors during emergencies. Some IMAs even enable the broadcasting of urgent messages regarding specific blood types, complete with geotagging to pinpoint donor locations in proximity to blood centers. Beyond these practical functions, IMAs are becoming instrumental in disseminating educational content, sharing recent research updates, fostering discussions, and facilitating training programs within the blood banking community. Within blood centers, IMAs serve as platforms for adverse event reporting, ensuring prompt responses and maintaining quality assurance. Their utility extends to remote consultations for

complex immunohematology cases, addressing issues in routine blood center operations, and providing support for patients experiencing massive bleeding. Additionally, IMAs offer a versatile platform for sharing digital multimedia, documents, and vital information among healthcare professionals in the blood banking domain. With so much popularity and utility these IMAs have their drawbacks of privacy concerns, security issues with end-to-end encryption, dependency on internet connection, and other social media platform integrations. Increasing the use of social media and IMAs has reduced the attention span of humans. This has not only bombarded humans with information overload but has also resulted in addiction, distraction, reduced face-to-face interaction as well as cyberbullying and increased social pressure. It is important to set boundaries and balance both online and offline interactions.

Academy day session

Career possibilities in transfusion medicine

AD08-L01 | New frontiers in transfusion science—higher specialist scientific training

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The Higher Specialist Scientist Training (HSST) programme is a five-year NHS England (NHSE) funded training programme supported by a Doctoral level academic award (DClinSci). The HSST programme provides opportunities for healthcare scientists in England to train to become eligible to apply for available consultant scientist posts in four main pathological divisions: Life Sciences, Physiological Sciences, Physical Sciences and Biomedical Engineering and Bioinformatics. The HSST programme in Clinical and Laboratory Transfusion Science, launched in 2015 under the Life Sciences division, comprises of 3 main areas of focus.

1. A postgraduate diploma (PgDip) in Leadership in Healthcare,
2. A Specialist Curricula assessed by Royal College of Pathologists (RCPATH) Fellowship examination,
3. A Doctoral level research project.

HSST offers a blended approach to training. It comprises of essential skills, including leadership, innovation, research, and higher specialist scientific and clinical knowledge. All required in senior scientific roles in the NHS. Trainees also network with other HSST trainees and have the opportunity to learn from and collaborate with experts in their field throughout the programme of study. In addition to the formal structure of lectures and the curriculum, the programme is mainly self-directed and the rate of progress through the programme is individual to each student, depending on their prior experience and

current / future job role. Consultant Clinical Scientists working in the field of transfusion alongside medically qualified colleagues are not novel. With Consultant Clinical Scientists working in Histocompatibility & Immunogenetics and other transfusion and transplantation-associated disciplines for many years. The development of a role for Consultant-level scientific practice within the specific field of transfusion, with a red cell immunohaematology focus, is a relatively recent development and one that has been welcomed in multidisciplinary patient care pathways, where the gradual change in patient demographic has meant that more transfusion dependent patients, with challenging transfusion requirements, such as those with haemoglobinopathies, require expert transfusion input from Consultant Clinical Scientists. The presentation describes the journey to Consultant Clinical Scientist from the perspective of one of the first Scientists to qualify via this exciting new training route.

AD08-L02 | From pipettes to pens & press

M Vermeulen

Abstract not available

AD08-L03 | From a national blood service to the WHO

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Transfusion medicine is the branch of medicine that encompasses all aspects of the transfusion of blood including aspects related to hemovigilance. It includes issues of blood donation, immunohematology, and other laboratory testing for transfusion-transmitted infectious agents (TTIAs), management of clinical transfusion practices, patient blood management, therapeutic apheresis, stem cell collections, cellular therapy, and coagulation. In many countries, transfusion medicine knowledge has not been fully introduced in medical education curricula or other health worker education. Therefore, the knowledge of transfusion medicine possessed can vary. For medical graduates, a widely understood career choice is to become a clinical specialist, health management expert, or scientist. I have started working in the field of transfusion medicine 30 years ago, as the head of the Infectious Transfusion Unit (TTI), head of Blood Component Production Division and director of the Indonesian Red Cross Central Blood Center. During this period I learned a scientific-based, management-based and clinical knowledge of transfusion medicines. After 2 period as Director of the Blood Center, I have been transferred to work in the Ministry of Health for 4 years. My role is to assist the Ministry of Health to develop Minister of Health Regulation on the national blood services organization, national technical standards of blood services and regulations for plasma fractionation. In addition, during my 25-year career in the field of blood, I was worked as a lecturer in a 3-year academy program for

blood technicians and 2-year master's degree program on transfusion medicine. Finally, 1 year before I came to retirement age, I applied for a job at WHO headquarters, and has started working as a team leader for Blood and Other Products of Human Origin. My role is to develop written global guidelines on blood and provide support to countries. The work of developing global guidelines led me to a new learning process on how to interact with global experts, global blood stakeholders and serve countries that need WHO support. In conclusion, transfusion medicine consists of multi knowledge and skills, from scientific, technical, management, clinical and even diplomacy. This rich knowledge and skills will open up opportunities to have more challenging works. Challenges will make our lives more interesting.

Parallel session—donors and donation

Hurdles to donor recruitment

PA01-L01 | Ensuring blood supply for the world's largest population—India

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Background: While blood transfusion remains an important tool in modern medicine and benefit patients from all age groups, it is not without risks. Given the lack of artificial substitutes, blood transfusion services worldwide continue to rely on human donors to maintain inventory and assure the supply. Risk reduction begins with donor selection and culminates in bedside procedures to ensure safety. According to the WHO, (WHO, 2021) the quantum of collection of blood is disproportionate to regional populations. There is a real deficit of blood supply especially in Low-income countries (LIC) and Lower middle income countries (LMIC). Systematic study of the need and demand for blood in the country is essential to design strategic approaches to ensure adequacy and reduce wastage.

Aims: In response to a request from the National AIDS Control Organization (NACO) of India, we designed and conducted a series of studies to estimate national need, clinical demand and supply of blood. (Mammen et al 2022).

Methods: We used review of literature and the Delphi method among groups of experts to estimate national need for blood. Demand was determined through a national level cross-sectional study in five randomly selected states from five representative regions of the country. We estimated the national demand by extrapolating the study data (demand and beds) from 251 facilities to the total number of estimated beds in the country. Utilization data was also collated from these facilities. Data regarding supply for the year studied was obtained from the National Blood Transfusion Council (NBTC) of India.

Results: The need for blood, is based on incidence or prevalence of morbidities that require replacement of blood/components and the quantum of blood required, was estimated at 26.2 million units per annum (95% CI; 17.9–38.0). National demand was estimated at 14.6 million units per annum (95% CI: 14.59–14.62) and achieving this should be the immediate focus of the Blood Transfusion System (BTS). The gap between supply and demand is 2.5 donations per 1000 persons, which is around one million units.

Conclusion: There is a gap between demand and supply, and the Blood Transfusion Services at the national level need to act strategically to focus programmatic efforts to ensure adequate supply and access to blood. There is a need to develop models that provide guidance tailored for their local demand. Investment into voluntary blood donor programs that will increase awareness in the community is essential to secure a safe source. Regions where there are chronic shortages (blood deserts) need to be mapped and novel modes need to be explored to ensure access to blood. (Raykar, 2024) This requires a systematic approach, enabling regulations and collaboration between authorities and like-minded organizations while focussing on grass root level efforts to address problems in a contextually relevant and cost-effective manner.

PA01-L02 | Dissecting factors required for achieving 100% voluntary non-remunerated blood donation: A qualitative study of international interview survey using the PESTELE framework

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Background: The WHO has revised the goal of achieving 100% voluntary, non-remunerated donation (VNRD) by 2025, however, the achievement seems difficult especially for low-income countries. The Global Blood Safety Working Party (GBS-WP) of the International Society of Blood Transfusion (ISBT) initiated an international interview survey.

Aims: To identify factors required for achieving 100% VNRD.

Methods: Representatives from each country were recruited by snowball sampling based on recommendations from ISBT GBS-WP members. Consent was obtained individually. The main topics of the semi-structured paper-based interview were types of blood donation

and their rates (#1), and the opinion regarding factors required for achieving 100% VNRD (#2). According to response #1, countries were classified into two groups; achieving countries with a VNRD rate of 90% or more and underachieving countries with that below 90%. The response text about #2 was coded according to the PESTELE framework, an evaluation matrix consisting of seven external factors (Political, Economic, Social, Technological, Environmental, Legal, and Ethical). Thematic analysis with iterative categorisation was then applied to the codes to identify key elements in each factor.

Results: Responses were obtained from a total of 13 interviewees from 10 countries. The achieving countries were Finland (100%), Japan (100%), Oman (96%), Thailand (90%–100%), and Türkiye (94%), while the underachieving countries were Brazil (65%), Indonesia (66%–92%), Georgia (36%), Ghana (25%), and Saudi Arabia (40%). The number of elements identified in each PESTELE factor were 5, 5, 10, 8, 5, 4, and 4, respectively. Among them, a total of 15 elements were common to both the achieving and the underachieving; “Policy making by the government” as a political element, “Awareness about the importance of VNRD”, “Public education”, “Community engagement”, “Reward”, and “Desire to improve blood safety” as social elements, “Testing for blood safety” and “Social media” as technological elements, “Construction of blood services” and “Community generation” as environmental elements, “Regulating remunerated donation”, “Regulating the obligation of blood services”, and “Legal reward” as legal elements, and “Nonmaleficence” and “Fidelity” as ethical elements. Compared to elements identified in the achieving, those in the underachieving were more variable in number (Table).

Summary / Conclusions: The fact that each PESTELE factor has more diverse elements in the underachieving may imply a big hurdle faced by these countries in transitioning to VNRD. From a political standpoint, the results suggest that government oversight by making specific regulations in addition to related policies would be effective for the underachieving countries. In addition to dealing with technological and environmental issues, social actions to foster “Awareness about the importance of VNRD” and “Desire to improve blood safety” through “Public education” and “Community engagement” seem very important. Collectively, this study showed that multiple factors with a wide variety of elements are required for achieving 100% VNRD,

PA01-L02 Table. The number of elements identified in the PESTELE factors by the qualitative analysis

Factors	Only in the Achieving	Common	Only in the Underachieving	total
Political	0	1	4	5
Economic	2	0	3	5
Social	1	5	4	10
Technological	2	2	4	8
Environmental	1	2	2	5
Legal	0	3	1	4
Ethical	0	2	2	4

underscoring the usefulness of the PESTLE framework to collect basic qualitative data for developing countermeasures to globalise 100% VNRD.

PA01-L03 | Removing upper age restrictions for returning donors and increasing the new donor upper age; donor safety findings utilizing a comprehensive donor vigilance system

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Background: In July 2019, Australia removed the upper age limit for returning donors (previously eligible until 81st birthday) and increased the upper age for new donors from 70 to 75 years. This change considered international experience with less conservative age criteria and increasing Australian life expectancy. Lifeblood collects data on all adverse events reported within 24 h of donation, affording a comprehensive, sensitive donor vigilance system. This review utilises our reporting system strengths to assess the safety of our policy change and includes an analysis of vasovagal reactions (VVR), phlebotomy-related events and apheresis specific adverse events.

Aims: To compare adverse event rates for new and returned donors who have become eligible under the new policy (upper age donors) with younger cohorts (18 to 70 years for new donors and 18 to 80 years for returning donors), including younger inexperienced donors who have historically proven to be at highest risk of VVR.

Methods: All whole blood and plasmapheresis collections from 14 July 2019 to 30 June 2023 were included in the analysis, together with all reported adverse events associated with those collections reported by 31 July 2023. The analysis determined the relative risk of individual adverse events in upper age donors compared with younger cohorts by gender and donation experience. Subgroup analysis determined the comparative risk of VVRs with potential for more serious outcomes including those associated with loss of consciousness (LOC), occurring offsite, and/or requiring outside medical care.

Results: There were 4529 upper age donors who made 8000 donations (6780 whole blood and 1220 plasmapheresis) in the reporting period. Males contributed 64.6% of the donations. A total of 119 VVRs (148.75 per 10,000 donations) were reported in upper age donors. The overall VVR rates for upper age new and returned donors were significantly lower compared with the younger cohort (RR 0.37; 95% CI 0.30-0.45; $p < 0.0001$ and RR 0.42; 95% CI 0.27-0.65; $p < 0.001$ respectively), consistent with known international data. However, we detected the upper age cohort had higher rates of VVR associated with LOC, requiring outside medical care and/or occurring offsite. Compared with younger donors; returning upper age donors had a significantly higher rate of LOC (RR 2.62; 95% CI 1.41-4.86; $p < 0.01$) and new upper age donors had a significantly higher rate of offsite VVRs (RR 1.60; 95% CI 1.08-2.37; $p = 0.02$). Both new and returning upper age donors had significantly higher rates of VVR requiring outside medical care than younger donors (RR: 2.48; 95% CI: 1.28-4.79; $p < 0.01$ and RR 4.45; 95% CI

2.00-9.91; $p < 0.001$ respectively). There were no significant differences for painful arm, nerve injury or citrate reactions. Infiltration rates were significantly higher in the upper age cohort; however analysis suggested the increase was associated with less serious low volume events.

Summary / Conclusions: The findings support historical findings that overall VVR rates in general decline with age independent of experience. However, this analysis provides a further level of insight and suggests upper age donors are at higher risk of serious VVRs, although the data available to date cannot definitively link risk to gender, experience or donation type. While the sample size is small and continued monitoring is needed, additional mitigations will be implemented to minimise risk, given this finding.

PA01-L04 | Has the switch to sexual risk behaviour screening impacted deferrals for PrEP/PEP therapy?

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Background: Individuals who become infected with HIV while on pre-exposure prophylaxis (PrEP) or post-exposure prophylaxis (PEP) or shortly after discontinuing therapy may have altered viral kinetics, with a prolonged window period for both antibody and nucleic acid (NAT) testing. For this reason, donors are asked about PrEP/PEP use in the last 4 months and deferred while on treatment and for 4 months after treatment cessation. In 2022, time based deferrals for men who have sex with men were replaced by sexual risk behaviour criteria; all donors are deferred if they had a new partner and anal sex or more than one partner and anal sex in the last 3 months.

Aims: We assessed donor deferrals for PrEP/PEP use in the 12 months before and after the criteria change.

Methods: Data on donors with a deferral code for PrEP/PEP was extracted from the Canadian Blood Services National Epidemiology Donor Database (NEDD). The donation file of these donors was reviewed to assess if additional HIV risk deferrals were applied on the same donation. Deferral rates pre-implementation (Sept 11, 2021-September 10, 2022, period 1) and post-implementation (September 11, 2022-September 10, 2023, period 2) were compared and p values calculated using Fisher's Exact Test.

Results: A deferral code for PrEP/PEP use was applied to 73 out of 787,312 donations screened in period 1 (9.3/100,000), and 88 out of 772,764 donations screened in period 2 (11.4/100,000 $p = 0.2075$). The percentage of coded donors deferred for PrEP use alone was 56% in period 1 (29 out of 52 donors coded for PrEP use) and 57% in period 2 (46 out of 80 donors coded for PrEP use), the deferral rate for PrEP use alone increased very slightly from 3.7/100,000 in period 1 to 5.9/100,000 in period 2 ($p = 0.0151$). Donors coded for PrEP use were predominantly male (94% in period 1, 95% in period 2), and younger than other donors (about 45% under 30, vs 16% of all donors). Deferral rates for PEP use remained low and stable; less than 15% of these donors did not have other reasons for deferral. Most donors coded for PEP use were female (80% in period 1, 70% in period 2).

Summary / Conclusions: The number of donors with deferral codes for PrEP use alone or in combination with other risk factor deferrals increased very slightly after implementation of sexual risk behaviour criteria, with slightly over half of these donors being deferred for answering yes to PrEP use and no to all other risk factors. There was no change in PEP deferrals. It is possible that individuals who are on PrEP/PEP and otherwise eligible for donation are self-deferring, and people may not be aware of the recent change to sexual risk behaviour criteria. Use of PrEP in the general population is increasing. At the present time, deferrals for PrEP/PEP have a very small impact on donor deferrals, but this may increase in the future.

PA01-L05 | For the Assessment of Individualised Risk (FAIR)—low numbers of on-session deferrals are seen under the more individualised policy in England

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Background: Since June 2021 in England, blood donation eligibility includes an assessment of individualised risks (FAIR) which replaced the 3-month deferrals relating to gay and bisexual men who have sex with men (GBMSM). Under FAIR, the pre-donation form includes questions to all donors about syphilis and gonorrhoea, chemsex, and anal sex with new or multiple partners; syphilis deferral was in place

pre-FAIR. This removed deferrals for some GBMSM and their female partners but introduced deferral for some who would not have been deferred previously, including women who have had anal sex with new or multiple partners.

Aims: To describe the impact of the FAIR policy on donor deferral in England by assessing on-session deferrals related to FAIR and return to donation during the 2-year post-implementation period.

Methods: Data were provided from the NHS Blood and Transplant donor management system for 5 new deferral codes implemented with FAIR: sexual activity—temporary and permanent deferral (for recent or continued sexual risk behaviour including FAIR), gonorrhoea, syphilis, and chemsex, along with the existing code for injecting drug use (to capture injecting chemsex) between 14 June 2021 and 13 June 2023, and donor type, sex, age, ethnicity, and return date. The rate of deferral was calculated per 100,000 donations made along with crude and adjusted incidence rate ratios and 95% confidence intervals by demographic.

Results: 286 donors were deferred on-session based on codes described (9.0 donors per 100,000 donations): 14 (0.4) permanently for sexual activity, 182 (5.7) temporarily for sexual activity, 25 (0.8) for chemsex, 13 (0.4) for gonorrhoea and 52 (1.6) for syphilis. No injecting chemsex was seen. 64 (3.7 per 100,000) female donors were deferred for sexual activity. Highest adjusted rate ratios of deferral were seen in first-time, male, 25- to 34-year-old, and Mixed and Other donors. Of 220 (77%) donors with temporary deferrals, we traced 140 and found 55% had returned to donate by 13 June 2023.

Summary / Conclusions: The impact of FAIR policy on donor loss in England appears low, with 286 deferrals among over 3 million

PA01-L05 Table 1: Number of donors deferred and rates by demographics

	Donors deferred (rate per 100,000 donations)	Incidence rate ratio (95% CI)	Adjusted incidence rate ratio (95% CI)
Total FAIR	286 (9.0)		
Donor type			
First-time	145 (57.1)	11.80 (9.30-14.99)	2.74 (2.43-3.09)
Regular	141 (4.8)	1.00	1.00
Sex			
Male	193 (13.4)	2.49 (1.94-3.23)	2.64 (2.06-3.38)
Female	93 (5.4)	1.00	1.00
Age group			
17 to 24	51 (23.5)	2.08 (1.42-3.02)	1.23 (0.85-1.79)
25 to 34	136 (20.6)	1.82 (1.36-2.46)	1.53 (1.15-2.04)
35 to 44	71 (11.3)	1.00	1.00
45 to 54	17 (2.5)	0.22 (0.12-0.38)	0.27 (0.16-0.45)
55+	11 (1.1)	0.10 (0.05-0.19)	0.13 (0.07-0.24)
Ethnicity			
Asian	23 (22.5)	2.94 (1.83-4.53)	1.33 (0.86-2.05)
Black	10 (25.4)	3.32 (1.57-6.22)	1.36 (0.72-2.57)
Mixed and Other	26 (35.3)	4.62(2.95-6.94)	2.12 (1.41-3.19)
Unknown	3 (11.8)	1.53 (0.41-4.55)	2.20 (0.70-6.89)
White	224 (7.7)	1.00	1.00

donations, or 9.0 per 100,000, up to 2 years after implementation. Most were temporary and where known, 55% returned to donate. This small impact was highest in young, male, and first-time donors. Women were impacted less, and deferral rates were lower than a pre-FAIR evaluation estimating 949 women per 100,000 donations had anal sex with new or multiple partners within 3 months. It is unclear if the low deferrals on session are due to successful messaging or lack of disclosure on session. A survey of blood donors in 2024 will explore donor understanding and compliance.

Parallel session— immunobiology

Haemolytic havoc— understanding blood cell destruction

PA02-L01 | Mechanism of post transfusion hyperhemolysis in sickle cell disease

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Hemolytic transfusion reactions are the most feared complications of blood transfusion in patients with sickle cell disease (SCD), especially when they developed accelerated clearance of transfused red blood cells (RBCs) with concomitant clearance of their own RBCs, and reticulopenia, developing the so called hyper hemolysis syndrom. This syndrom is frequently underdiagnosed because of the symptoms that are mainly related to the disease. This condition is life-threatening, accounting for 6% of all-cause mortality of adult patients with SCD, as demonstrated in a retrospective survey. (Habibi Am J Hematol 2016). Indeed, hyperhemolysis is characterized by intra vascular hemolysis, and the release of free hemoglobin (Hb) and heme can induce multi-organ failure and death, most likely due to damage to the underlying vasculature. Anti-erythrocyte alloimmunization, with restimulation of an evanescent antibody, favored by the inflammatory state of these patients at the time of transfusion, is the main cause of this syndrom. Patients undergoing occasional, isolated transfusions have been shown to have a higher risk of developing this condition, especially if they have history of previous hemolytic transfusion reaction and known allo antibodies in the past. However, the severity of these accidents is not comparable with the general population of transfused and allo immunized patients. What's more, the mechanism is sometimes enigmatic when no allo antibodies are detected after the reaction, or at a distance, despite sensitive immuno-hematological work up. Complement is a key factor in these accidents, both in triggering them and in potentiating the patient's inflammatory state and tissue

damage. Free heme and hemoglobin that are released can interact with complement, causing tissue damage and inducing a feedback loop that increases destruction of donor red blood cells, because of the effect of free heme on complement inhibitor. It can be activated through the classical pathway, however, in the absence of detectable antibody, beside classical pathway, it is likely that alternative pathway could be involved. Case reports provide evidence for a final activation of complement pathways, and in some case an increase in Bb levels, indicating complement activation via the alternative pathway. In these cases, efficient treatment with eculizumab (anti-C5 convertase antibody) as a salvage therapy, resulted in a demonstrated effect on hyperhemolysis, highlighting the involvement of complement. Then, many observations in patients encountering this condition support the conclusion that complement is key disease driver and a promising therapeutic target in the context of transfusion-related hemolysis and hyperhemolysis for patients with SCD. However, there is an urgent need to develop evidence-based approaches for preventing and treating this reaction.

PA02-L02 | Knops causes decreased RBC clearance and increased antigen-modulation of other blood group antigens during incompatible transfusion in mice

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Background: Even for “clinically significant” alloantigens some incompatible units can be safely transfused while others can cause potentially lethal delayed hemolytic transfusion reactions (DTRs). Because there are currently no means to predict which incompatible units will cause DTRs, transfusion of incompatible blood is strictly avoided. This can become seriously limiting for multiply alloimmunized patients in need of ongoing transfusion (e.g., patients with sickle cell disease) for whom insufficient blood is available. If it was possible to determine which units of blood would not cause DTRs despite being incompatible, it would greatly increase available units and/or guide the choice of “least incompatible” units, when required. The biology of incompatible transfusion involves multiple simultaneous events, including IgG binding, C3 deposition, antigen-modulation, and hemolysis. Many essential mechanistic studies are neither technically nor ethically feasible in humans, requiring animal models. However, one distinct difference between mice and humans is that murine RBCs do not express complement receptor 1 (CR1 a.k.a. Knops).

Aims: To study the effects of CR1 on incompatible transfusion by introducing human CR1 into a murine model of incompatibility to the KEL2 alloantigen.

Methods: Transgenic mice with RBC specific expression of human CR1 were crossed with mice expressing the human KEL2 blood group antigen. KEL2+CR1- or KEL2+CR1+ RBCs were transfused into wild-type recipients that were first passively immunized with anti-KEL IgG (incompatible) or PBS (compatible control). Thus, there was a total

of 4 groups ($n = 10$ mice per group). Longitudinal peripheral blood samples were collected at 5, 30, min, 1, and 24 h post-transfusion. RBC clearance, C3 deposition, IgG binding (i.e., direct antiglobulin test [DAT]), and antigen-modulation were measured by flow cytometry with quantitation reported as median fluorescent intensity (MFI).

Results: Both KEL2+CR1- and KEL2+CR1+ RBCs were rapidly bound with anti-KEL IgG and had C3 deposition. However, KEL2+CR1- but not KEL2+CR1+ RBCs had brisk clearance from circulation (30% vs. 0%, $p = 0.001$) at 24 h. Antigen modulation was faster in KEL2+CR1+ RBCs compared to KEL2+CR1- RBCs (85% decrease vs. 77% decrease, $p = 0.004$) at 30 min. Likewise, surface bound IgG decreased faster on KEL2+CR1+ RBCs compared to KEL2+CR1- RBCs (MFI (1033) vs. MFI (1466), $p = 0.0007$) at 30 min. Finally, KEL2+CR1+ RBCs had increased C3 deposition compared to KEL2+CR1- (MFI(506) vs. MFI(280), $p = 0.0002$) at 5 min.

Summary / Conclusions: These data demonstrate that human CR1 on RBCs decreases clearance of incompatible RBCs and also facilitates antigen-modulation and lowers levels of surface bound IgG. We speculate that CR1 prevents clearance by increasing antigen-modulation and thereby decreasing surface IgG to levels below that which cause phagocytosis. This interpretation is consistent with the known participation of CR1 in “the immune transfer reaction”. High and Low Density Knops alleles have been described in humans, with 10 fold different levels of expression. Thus, the current studies justify consideration of the hypothesis that variant CR1 expression levels on donor RBCs may affect the extent of hemolysis during incompatible transfusion and could serve as a practical metric, when required, to identify incompatible units less likely to hemolyze.

PA02-L03 | Clinical significance of anti-e in sickle cell disease (SCD) patients undergoing transfusion carrying the RHCE*ceVS.01 and RHCE*ceVS.02.01 variant alleles

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Background: Anti-e is the most frequently observed alloantibody in Sickle Cell Disease (SCD) patients with variant RHCE alleles. However, not all variants are implicated in hemolytic transfusion reactions, suggesting that the risk of alloimmunization and the clinical significance of Rh antibodies produced may vary based on the specific inherited variant.

Aims: Given that RHCE*ceVS.01 (RHCE*ce733G) and RHCE*ceVS.02.01 (RHCE*ce48C,733G) are associated with the partial e-antigen and are the most commonly found variant alleles in our SCD patient population, we aimed to analyze the risk of anti-e formation in transfused patients carrying these variants exposed to conventional e-antigen. Additionally, we aimed to assess the clinical significance of the produced anti-e antibodies.

Methods: SCD patients carrying the RHCE*ceVS.01 and RHCE*ceVS.02.01 variant alleles with a history of ≥ 15 transfusions were selected. RH genotyping was performed on all patients using the

RHCE BeadChip array (BioArray, Immucor). Antibody screening and identification with autologous control were conducted using the gel test. Direct antiglobulin test (DAT), adsorption with autologous RBCs, and crossmatching with allogeneic partial e-antigen from donors carrying the same alleles were also performed when possible. The clinical significance of anti-e was assessed by comparing hemoglobin levels recorded before and after transfusion at the time of antibody detection and by clinical suspicion of anemia and hemolysis.

Results: Sixty-one patients were enrolled in this study (38 with RHCE*ceVS.01 and 23 with RHCE*ceVS.02.01 variant alleles). Among SCD patients with RHCE*ceVS.01, 5 were homozygous, 28 were heterozygous, and 5 were compound heterozygous. Of these, 19 produced allo anti-e, 9 auto anti-e, and 5 did not develop anti-e. Among patients with RHCE*ceVS.02.01, 8 were homozygous, 7 were heterozygous, and 8 were compound heterozygous. Of these, 17 formed anti-e alloantibodies, 8 auto anti-e, and 3 did not develop anti-e. None of the allo anti-e developed by these patients were associated with a decline in hemoglobin levels or clinical suspicion of anemia after transfusion with conventional e-antigen.

Summary / Conclusions: Our findings demonstrate a high rate of anti-e alloimmunization (59%) in SCD patients carrying the RHCE*ceVS.01 and RHCE*ceVS.02.01 variant alleles. However, the anti-e antibodies produced were not clinically significant, as most patients who developed allo anti-e exhibited good survival of transfused RBCs when exposed to conventional e-antigen.

PA02-L04 | Evanescence of anti-Mia and associated risk of haemolytic transfusion reactions among thalassaemia patients

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Background: Anti-Mi^a is a common clinically significant red cell alloantibody in Southeast Asia. It is also the third most common clinically significant red cell alloantibody among our transfused thalassaemia patients. We previously followed the conventional recommendation of transfusing anti-human globulin (AHG) crossmatch-compatible red cells to patients with history of anti-Mi^a. However, this approach alone without selection of Mi^a-negative red cells, may not detect incompatibility with Mi^a-positive red cells in patients whose anti-Mi^a has evanesced and could lead to higher risks of haemolytic transfusion reactions in regions where blood donors have higher Mi^a prevalence. We recently encountered delayed haemolytic transfusion reaction (DHTR) in a transfusion dependent thalassaemia (TDT) patient with known anti-Mi^a, from inadvertent transfusion of incompatible Mi^a-positive red cells that was undetectable by AHG-crossmatching due to anti-Mi^a evanescence.

Aims: We evaluated the characteristics of anti-Mi^a evanescence and complications associated with anti-Mi^a among our transfused thalassaemia patients. This also prompted a review of our transfusion practices for thalassaemia patients with history of anti-Mi^a.

Methods: Transfused thalassaemia patients with history of anti-Mi^a was identified from databases of our other study on red cell alloimmunization among adult thalassaemia patients and our department's "Registry of Blood Conditions". Their data was de-identified and retrospectively analysed for characteristics of anti-Mi^a evanescence and haemolytic transfusion reactions associated with anti-Mi^a.

Results: 5 TDT and 8 non-TDT patients (3 males and 10 females) with history of anti-Mi^a were included. 62% of them ($N = 8$) had evanescence of anti-Mi^a after median (range) of 20 (1–299) months, and the remaining continued to have detectable anti-Mi^a after follow-up for median (range) of 44 (14–222) months. The overall median time to evanescence of anti-Mi^a by Kaplan-Meier survival analysis for all patients was 40 months. At the time of initial detection of anti-Mi^a, 1 NTDT patient had hyperhaemolytic transfusion reaction and 2 TDT patients had DHTR presenting as Hb decline detected during routine pre-transfusion monitoring—one of whom had a second DHTR after anti-Mi^a evanescence due to inability of AHG-crossmatching to detect incompatibility of the Mi^a-positive red cells.

Summary / Conclusions: There is a high rate of anti-Mi^a evanescence among our thalassaemia patients. Even though many had anti-Mi^a evanescence only after a few years, their need for repeated transfusions would pose them at higher risks of receiving incompatible red cells and hence DHTR once their anti-Mi^a evanesce, if only AHG-crossmatching is done as our blood donors have higher Mi^a prevalence (estimated to be at least 5%). We have therefore changed our practice to providing Mi^a-negative red cells to our Mi^a-alloimmunised thalassaemia patients whose anti-Mi^a has evanesced. The lack of Mi^a antisera licensed for automated blood donation phenotyping platforms has limited the availability of Mi^a-negative red cells to our patients but our blood service is working towards building up this inventory using antisera that is currently licensed only for the tube technique. When there is adequate Mi^a-negative red cells, it may be feasible to provide Mi^a-negative red cells to all Mi^a-alloimmunised patients and also pre-emptively to non-alloimmunised thalassaemia patients to further minimise their complications from Mi^a-alloimmunisation.

PA02-L05 | In vitro and in vivo analysis of platelet phagocytosis induced by anti-CD36 antibodies

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Background: CD36 is a membrane glycoprotein present on platelets, monocytes and endothelial cells and many other cells. Anti-CD36 iso-antibodies developed by immunized CD36 deficient individuals are responsible for the development of platelet transfusion refractoriness (PTR) and fetal neonatal alloimmune immune thrombocytopenia (FNAIT). However, little is currently known about the mechanism of platelet clearance mediated by anti-CD36 antibodies in these immune mediated disorders.

Aims: In this study, we analyzed the phagocytosis of anti-CD36 opsonized platelets both *in vitro* and *in vivo* using mouse monoclonal antibodies (mAbs) GZ1 (IgG2a) and GZ4 (IgG1).

Methods: *In vitro* phagocytosis analysis was performed with phRodo platelets as previously described (Takahashi et al., 2017). *In vivo* mAbs were administered at a dose of 0.8 mg/KG into C57BL/6J female mice via tail vein.

Results: *In vitro* phagocytosis analysis showed significant higher phagocytosis of mAb GZ1 opsonized platelets compared to mAb GZ4 (phagocytosis rate: 60% versus 40%). Incubation of platelets with mAb GZ4 caused platelet activation associated with significant up-regulation of CD62 and CD63, mainly through P38-MAPK and NF- κ B signaling pathways. Our inhibition studies with mAbs against Fc γ Rs showed that platelet phagocytosis mediated by *non-activator* mAb GZ1 was efficiently blocked by anti-Fc γ RI, whereas platelet engulfment triggered by *activator* mAb GZ4 were efficiently inhibited by anti-Fc γ RIIa antibodies. Similar result was observed with subclass-switched recombinant mAb GZ1 (IgG2a>IgG1). Nevertheless, Syk inhibitor (R406) inhibited platelet phagocytosis induced by both mAbs. When these mAbs were injected into mice, similar phenomenon was observed. MAb GZ1 showed significant higher platelet clearance in comparison to mAb GZ4. Although rapid decline was observed, the platelet count returned to normal after 60 minutes. In contrast, anti-allb3 antibody (mAb LeoF2) induced continuously strong platelet clearance. Again, R406 blocked platelet clearance in this animal model.

Summary / Conclusions: This study demonstrated that anti-CD36 antibodies could induce platelet clearance, both *in vitro* and *in vivo*. Our *in vivo* mice model, however, showed that anti-CD36 antibodies induced rapid recovery of platelet count; differed from platelet clearance caused by "platelet-specific" anti-allb3 antibodies. Furthermore, the degree of platelet clearance could be influenced by the presence of activated platelets/aggregates triggered by IgG1 antibody subclass. In respect to the important of platelet phagocytosis assay for the prediction of severity of thrombocytopenia, these hints should be further investigated in the near future.

Parallel session—clinical

When and what to transfuse?

PA03-L01 | ECMO and blood transfusion

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Blood transfusion can be lifesaving however in the past decades it has become clear it can also be life threatening. Several landmark trials have shown that a restrictive transfusion policy in the critically ill is safe and well tolerated and in some settings may even result in better outcome compared to a liberal strategy. This has resulted in

implementation of a restrictive transfusion policy in the majority of the critically ill patient populations. Patients on extra corporal membrane oxygenation (ECMO) are still transfused with a liberal strategy. In the current presentation an overview on literature and mechanisms in place which has resulted in this liberal strategy will be discussed. Also future research priorities will be discussed.

PA03-L02 | Effects of transfusion strategies in patients with acute compared to chronic anemia—a pre-specified subgroup analysis of the Myocardial Ischemia and Transfusion (MINT) trial

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Background: Acute and chronic anemia are both common in patients with acute myocardial infarction (MI). In acute anemia, insufficient time for physiological adaptation to sustain oxygen delivery might lead to additional clinical benefits from a liberal red blood cell (RBC) transfusion strategy.

Aims: To investigate a differential effect of liberal and restrictive RBC transfusion strategies on post-MI outcomes in acute versus chronic anemia.

Methods: The MINT trial randomized anemic patients (hemoglobin level (Hb) < 10 g/dL) hospitalized with acute MI to either a restrictive (Hb < 7-8 g/dL) or a liberal (Hb < 10g/dL) RBC transfusion strategy. In these subgroup analyses, we included patients with Hb measurement prior to any transfusion on admission day one or two. Acute anemia was defined as having a first Hb above 13 g/dL for men or 12 g/dL for women, or if there was a drop of Hb ≥ 2 g/dL between the first Hb and randomization. The primary outcome was 30-day all-

PA03-L02 Table 1

	Acute anemia (n = 1078)	Chronic anemia (n = 2066)
Age	72.0 (11.4)	72.4 (11.7)
Male	598 (55.5%)	1117 (54.1%)
MI Type 1	505 (46.8%)	802 (38.8%)
Critical Care	599 (55.6%)	871 (48.0%)
Mechanical ventilation	298 (27.6%)	128 (7.0%)
Number of antiplatelets or anticoagulants		
0	56 (5.2%)	159 (7.7%)
1	146 (13.6%)	336 (16.3%)
2	267 (24.8%)	666 (32.2%)
3	608 (56.5%)	905 (43.8%)

cause mortality or recurrent MI. Secondary outcomes were components of the primary outcome, cardiac death, heart failure, and pulmonary complications (transfusion-related acute lung injury, pneumonia, or acute respiratory failure). We tested the interaction between transfusion strategy and anemia acuteness, using mixed effect models adjusted for baseline risk factors. We also estimated the overall effect of anemia acuteness on the primary outcome using adjusted risk ratios (aRR).

Results: Of 3504 participants randomized in MINT, 3144 (89.7%) were included in these analyses; 1078 (34.3%) had acute anemia and 2066 (65.7%) had chronic anemia. Patients with acute anemia had more type 1 MI and a more severe clinical course prior to randomization (Table 1). Acute anemia, compared to chronic anemia, was associated with a higher risk of the primary outcome (aRR = 1.25; 95% confidence intervals (CI): 1.05 to 1.48). However, the effect of RBC transfusion strategy on all outcomes was similar for patients with acute and chronic anemia (no effect modification, see Table 2).

Summary / Conclusions: In patients with MI and anemia, transfusion strategies did not have different effects in acutely anemic compared to chronically anemic patients even though patients with acute anemia were sicker and had a higher risk of adverse outcomes. A liberal transfusion strategy may be prudent for MI patients, regardless if the anemia is acute or chronic.

PA03-L02 Table 2

Outcomes	Full Cohort	Acute Anemia	Chronic Anemia	Interaction p value
Death or MI	1.15 [0.98–1.35]	1.09 [0.85–1.40]	1.20 [0.97–1.48]	0.57
Death	1.15 [0.92–1.44]	1.14 [0.82–1.59]	1.16 [0.86–1.56]	0.97
MI	1.22 [0.96–1.55]	1.14 [0.77–1.69]	1.27 [0.94–1.72]	0.66
Cardiac death	1.73 [1.23–2.43]	1.70 [0.97–2.99]	1.74 [1.13–2.67]	0.95
Heart failure	0.89 [0.68–1.16]	0.71 [0.45–1.13]	1.00 [0.72–1.39]	0.23
Pulmonary complications	1.00 [0.83–1.21]	1.11 [0.84–1.47]	0.93 [0.72–1.19]	0.23

PA03-L03 | Transformative effects of patient blood management—a ten year review on red blood cell utilization in a community teaching hospital

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Background: Implementing Patient Blood Management (PBM) principles in a small hospital, amidst the absence of dedicated budgetary provisions, presents hurdles. Nonetheless, there exist various strategic measures to incorporate PBM practices without the reliance on supplementary funding. Using an organized, evidence-based multidisciplinary approach to revamp blood transfusion practices to enhance patient care for the patients who might need blood transfusion, a PBM program was initiated in 2013 and fully implemented in 2014 at the 350-bed community teaching hospital in urban New York City.

Aims: This investigation was to demonstrate the efficacy of the PBM program in reducing unnecessary red blood cell (RBC) transfusions at this hospital.

Methods: Employing a retrospective statistics analysis covering the period from 2013 to 2022, the study scrutinized RBC transfusion data over ten years.

Results: Significant enhancements became evident after the implementation of the PBM initiative. Notable achievements included a decrease in mean pretransfusion hemoglobin levels from 7.26 g/dL in 2013 to 6.62 g/dL in 2022, a remarkable 35.7% reduction in annual RBC unit transfusions, and a marked decline in units administered to patients with pre-Hgb levels ≥ 7 g/dL (from 1210 units in 2013 to 420 units in 2022). Additionally, the study observed a notable decrease in two-unit RBC orders for patients with Hgb levels ≥ 7 g/dL, dropping from 65 orders (3.4%) in 2013 to 36 orders (2.1%) in 2022. The total estimated cost-savings attributed to the nine-year duration of the PBM program, fully implemented in 2014, amounted to \$4.1 million US dollars. It is noteworthy that the study period encompassed the challenging times of the Covid-19 pandemic in 2020-2022. Despite the unprecedented circumstances, the PBM program demonstrated its sustained impact on RBC transfusions and maintained its effectiveness throughout the pandemic. The median length of stay (LOS) from 2013 to 2021 stayed flat at 3.0 days, suggesting no patient harm effect of the PBM program. Remarkably, the PBM program made significant strides throughout 9 years without the need for any supplementary budget allocation.

Summary / Conclusions: The implementation of PBM significantly reduced RBC transfusions and enhanced overall transfusion practices. This study underscores the resilience of successful PBM strategies, even in the face of a global health crisis like the Covid-19 pandemic. Furthermore, it highlights that PBM success is not contingent on

substantial resources or increased budgets but rather relies on the application of practical and intuitive methods, as exemplified by the outcomes of this research.

PA03-L04 | Evaluating retrieval-augmented generation (RAG) based Artificial Intelligence (AI) model against conventional large language models in transfusion medicine—a comparative study

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Background: Artificial Intelligence (AI) and Machine Learning (ML) have shown the potential to enhance clinical decision-making across various medical specialties. The major drawback with these tools is the lack of reliable results and the risk of producing inaccurate or fabricated information in the generated data, known as “hallucinations”. To address this issue, a unique AI model that utilizes the Retrieval-Augmented Generation (RAG) framework for clinical transfusion was developed by the collaborative efforts of transfusion medicine experts and data engineers.

Aims: To engineer and validate a computational framework powered by AI that leverages the RAG approach, to augment clinical decision-making processes in transfusion medicine and perform a comparative analysis with other existing large language models (LLM).

Methods: During the initial development phase, we adopted the RAG methodology, implemented on the OpenAI platform. This method integrates a meticulously annotated knowledge base, consisting of interconnected documents designed to provide referential context within targeted prompts. To bolster the reliability and accuracy of content generation, we embedded comprehensive guidelines and illustrative examples into the prompt architecture, thereby reducing the incidence of erroneous generative responses. Through iterative enhancements over the next four months, we refined the knowledge base, systematically improving the model's performance. We devised a questionnaire encompassing 24 diverse clinical transfusion scenarios, each detailing varying complexities of clinical conditions. This questionnaire was presented to our RAG model as well as to other publicly available LLMs, including ChatGPT 4, ChatGPT 3.5, and Google Bard. The responses from these models were anonymized and assessed by six subject matter experts across nine categories, including accuracy, personalization, adherence to evidence-based practices, patient safety and ethical considerations. Individual scores for each model were recorded and a comprehensive statistical analysis of the results was carried out.

Results: The RAG model demonstrated superior performance, achieving the highest mean score of 8.45 (out of 10) with 95% CI and a standard deviation of 0.37. In contrast, ChatGPT 4 attained a lower mean score of 6.65. One-Way ANOVA revealed significant differences in mean scores between models across all evaluated categories ($p < 0.001$), with an F -value of 37.90. Subsequent post hoc comparisons utilizing Tukey's

HSD test confirmed that the RAG model's performance was statistically superior to the other three models. ChatGPT 4 was found to outperform the remaining two models, while no significant difference was detected between the performances of ChatGPT 3.5 and Google Bard. Cohen's d effect size for the comparison between the RAG model and ChatGPT 4 was 3.18, which underscores a substantial difference in performance between these two models.

Summary / Conclusions: These results suggest that the RAG model's integration of retrieval-augmented processes may contribute to its enhanced decision-making capabilities in clinical transfusion scenarios. The model demonstrates the synergy of AI's analytical capabilities with expert human insight, enhancing decision support systems in efficiency and reliability. It offers a scalable, cost-effective solution compatible with current systems, serving as both a clinical decision aid and a tool for future blood usage audits.

PA03-L05 | Improving the diagnosis of paediatric iron deficiency—a lean Six Sigma success story

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Background: Iron deficiency is a common nutritional disorder in children and adolescents that is readily correctable, with significant implications for their growth and development in addition to iron deficiency anaemia. The Royal College of Pathologists Australasia recommends a cut-off of <20 ng/mL (<45 pmol/L) for diagnosing paediatric iron deficiency. The diagnosis and treatment of paediatric iron deficiency has been reviewed.

Aims: We aimed to evaluate the impact of a Lean Six Sigma intervention on the diagnosis of paediatric iron deficiency performed in our hospital.

Methods: A retrospective review was conducted on children and adolescents below 18 years who were tested to have a ferritin level <45 pmol/L in the New Territories West Cluster (including Tuen Mun Hospital, Pok Oi Hospital and Tin Shui Wai Hospital) between July 2022 and December 2023. Patients with known iron deficiency, on iron replacement or recent transfusion within 1 year were excluded. Same patient results after the first reading and patients defaulted follow-up/referrals after the initial ferritin result were also excluded. Data were collected on ferritin levels, serum iron profile, iron replacement prescription rates, and other relevant variables such as red cell parameters. Anaemia was defined by World Health Organization (WHO) definition of anaemia. A Lean Six Sigma approach was used to identify and address key factors contributing to the under-diagnosis and under-treatment of paediatric iron deficiency. A pre-intervention audit was performed to review the diagnosis and treatment of paediatric iron deficiency in the period July to December 2022. Root causes of the problem and potential interventions were identified. After intervention, a re-audit was performed to review the diagnosis and treatment of paediatric iron deficiency in the period November to December 2023.

Results: In the pre-intervention audit period, the rate of diagnosing paediatric iron deficiency was 54.7% (1.6 σ). The rate of iron replacement prescription was 50%. The rate of WHO-defined anaemia was 71.9%. In a follow-up period of 180 days, the patients with iron replacement have median best haemoglobin improvement of 2.3 g/dL compared with 0.1 g/dL in patients with no iron replacement given ($p < 0.00001$). The major problems identified in the process included: (1) only reference interval was available in the ferritin laboratory report but not the decision cut-off for diagnosing paediatric iron deficiency; and (2) lack of clear guidance for diagnosis and management of iron deficiency. Specific interventions included an addition of the decision cut-off of <45 pmol/L for diagnosis of paediatric iron deficiency in the ferritin laboratory reports and a teaching session organized by the haematology and oncology team of paediatrics department for pediatricians on the diagnosis and management of iron deficiency. A re-audit conducted after the implementation of the interventions showed significant improvement in the rate of diagnosing paediatric iron deficiency from 54.7% (1.6 σ) to 94.7% (3.1 σ) ($p = 0.0010$). The iron replacement prescription rate improved from 50% to 78.9% ($p = 0.0347$).

Summary / Conclusions: The Lean Six Sigma intervention was successful in identifying the problems in the process of diagnosis and treatment of paediatric iron deficiency and offering insights to the potential solutions. This audit highlights the potential of Lean Six Sigma as a tool for improving the quality of paediatric iron deficiency management.

Parallel session—cellular therapies

Sickle cell disease - are we near a cure?

PA04-L01 | Landscape of sickle cell in Africa—current treatment, challenges and what the future holds

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Sickle cell disease (SCD) is the most common genetic cause of morbidity and mortality in the world. Over 75% of the patients with SCD reside in Sub-Saharan Africa (SSA). It is estimated that, without proper care, up to 50% of patients with SCD will die in early childhood. Newborn screening and access to comprehensive care have been shown to reduce the SCD-related morbidity and mortality. However, due to resource constraints, most countries in SSA are yet to implement universal newborn screening programs for SCD. Moreover, care for patients with SCD in the region is hampered by several other challenges including shortage of qualified healthcare workers, lack of guidelines for the clinical management of SCD, limited infrastructure for inpatient and outpatient care, and limited access to blood, blood

products and disease modifying drugs such as Hydroxyurea which collectively contribute to the poor clinical outcomes. The curative options such as bone marrow transplant and gene therapy are not available in many countries in SSA and, when available, are generally expensive and inaccessible to many patients. In addressing these challenges, various initiatives are ongoing in SSA with the goal to enhance awareness on SCD, improve patient identification and retention to care, harmonize the standards of care for SCD, improve the skills of healthcare workers, enhance utilization of Hydroxyurea and conduct research on pertinent areas in SCD in the SSA context. Strengthening these measures is paramount to improving the outcomes of SCD in SSA.

PA04-L02 | Stem cell transplant in sickle cell disease

M Koh

Abstract not available

PA04-L03 | Building a diverse stem cell registry

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Background: Patients with certain hematological, immunological, or metabolic conditions are treated with hematopoietic stem cell transplantation, and stem cell transplants can treat over 80 diseases and disorders. In most cases, this transplant must come from an unrelated donor because there is no potential HLA matched donor in their family. Often patients cannot find an appropriate unrelated donor, because of disproportionate representation on individual registries, difference in genetic diversity across ethnicities and attenuation rates for donor registries. Stem cell registries struggle to recruit committed stem cell donors, particularly from targeted demographics such as underrepresented racialized communities.

Aims: To examine recruitment materials and outreach strategies to increase stem cell awareness and registration among underrepresented racialized Canadians, aged 17–35.

Methods: Focus groups were conducted with young adults ages 17–35 who self-identify as BIPOC/racialized to explore their views of the blood service, stem cell donation, promotional materials related to the stem cell registry, and community-led outreach strategies. Data analysis was conducted using thematic analysis (Braun & Clarke 2006) and informed by interpretive grounded theory (Charmaz 2003). NVivo 12, qualitative data analysis software was used in data analysis. All interview transcripts were de-identified and uploaded to NVivo. Canadian Blood Services' Research Ethics Board approved this study.

Results: Five focus groups were conducted with a total of 17 participants. Focus group participants' views on the blood operator were

related to the blood operator's overall lack of engagement with their community. While participants saw the blood operator as serving an important need, to collect and distribute blood, they were not sure about how donation fit into the healthcare system, the need for stem cell donations, or the importance of donations from racialized communities. Most had not donated because of a lack of awareness of the blood service, screening criteria, no centre in their neighborhood, and their community had not been engaged. Half registered concerns when asked about their opinion of the blood operator, stemming from 1, the belief that screening criteria were discriminatory toward racialized and sexual and gender minorities, and 2, the blood operator wasn't visible in their community. They felt materials promoting the stem cell registry in Canada needed to clarify what stem cell donation is and why racialized communities matter. Further, that these materials should be more sensitive to representation of racialized communities. Finally, outreach events should be tailored to the communities' needs and interests.

Summary / Conclusions: This study found that knowledge of stem cell donation, messaging to promote stem cell donation, and strategies to register donors from underrepresented racialized communities are inter-connected and require partnership with these communities. Building a diverse registry requires working with communities to offer them agency and control to co-develop materials that resonate with their members, and so that they can understand the relationship with the blood operator as a long-term effort to meet the needs of all Canadian patients. Through this relationship, the blood operator has the opportunity to learn what it means to accommodate, support and foster belonging for these communities.

PA04-L04 | Establishment of umbilical cord blood and cord tissue banking as somatic stem cell source for medical implementation

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Background: Human umbilical cord blood (CB) and umbilical cord tissue (UC) are attractive sources of somatic stem cells for gene and cell therapies. CB and UC can be obtained noninvasively from donors. Currently CB is not only for hematopoietic stem cells transplantation (HSCT) source but also the optimal source for immunotherapy. Human UC has been rapidly utilized as an abundant source of mesenchymal stromal cells (MSCs) worldwide due to its ease of collection, noninvasive collection procedure, and categorization as biological waste at birth. We established CB and UC bank, namely IMSUT CORD, to support the cell therapies.

Aims: Our aim is to establish a stable supply system of frozen CB and UC-derived cells by solving legal, ethical, and social problems to aim the medical implementation.

Methods: IMSUT CORD collected both CB and UC after obtaining informed consent from the guardian of the baby. The collected CB and UC are transported from the hospitals to the IMSUT CORD. CBs is processed into mononuclear cells and cryopreserved. UC tissues are cryopreserved until obtaining confirmation that the baby exhibits healthy, normal development and the mother remains free from infection within at least six months after delivery. Processing method of UC into UC-MSCs is described previously (Nagamura-Inoue, IJH, 2022). Briefly, frozen-thawed UC tissues were cultured by improved explant culture procedure. The migrating cells are harvested and denoted as master cells. Frozen master cells are thawed and expanded to have intermediate cell products, or final cell products, according to the physician's or company's requirements of cell modalities with quality and safety test.

Results: Informed consent explanation includes ownership right transition, intellectual property, proxy consent, the commercial use of CB and UC to save the life of patients, discard policy, and so on. CBs have been processed into MNCs and cryopreserved for further processing easily. UC-MSCs showed significantly faster and higher proliferation potencies than bone marrow and adipose tissue derived MSCs. UC-MSCs are plastically adherent, positive for CD105, CD73, and CD90 but negative for CD45, CD34, CD14, CD19, and HLA-DR surface molecules, and differentiate into adipocytes, chondrocytes, and osteoblasts *in vitro*. Even 3rd-party UC-MSCs suppress the proliferation of CD4 and CD8-positive cells activated by allogeneic dendritic cells. Indoleamine 2, 3-dioxygenase 1 and PGE2 are induced in UC-MSCs by IFN- γ . UC-MSCs constitutively express the PD-L2, while PD-L1 is induced in response to IFN- γ . UC-MSCs secrete large amount of HGF and BDNF, suggesting the tissue repair potent. These characteristics of UC-MSCs described above are expected to contribute to the development of treatments in the fields of immunotherapy and regenerative medicine. We have shipped UC-MSCs as final products for clinical trials including steroid-resistant aGVHD (P1), and COVID-19-related ARDS (P1), cerebral palsy (P1/2), and Non-infectious pulmonary complications after HSCT (P2). No severe adverse events related to the injection of our UC-MSCs products.

Summary / Conclusions: Both human umbilical CB and UC may serve as effective and sustainable sources for gene and cell therapies in immunotherapies and regenerative medicine. Promotion of social acceptance might be necessary for further stable supply.

Parallel session—clinical

Following guidelines—AABB / ISBT joint session

PA05-L01 | International RBC transfusion guidelines

S Stanworth

Abstract not available

PA05-L03 | RBC transfusion strategies: AABB guidelines, debate—which restrictive hemoglobin threshold? MINT trial results

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Three topics will be presented in this overview of RBC transfusion thresholds. (1) Simon Stanworth will present an overview the methods and recommendations from recently published AABB RBC transfusion guidelines. There is an expanding number of randomized trials of different red cell transfusion thresholds which have been appraised in multiple iterations of a Cochrane systematic review. This meta-analysis formed the basis for an international guideline and the development of recommendations to address several key 'PICO' questions. These were framed by a core Steering Committee and reviewed by a broad international panel, and included whether for hospitalized, hemodynamically stable adult patients, should clinicians transfuse with a restrictive strategy (typical hemoglobin level <7-8 g/dL) versus a liberal strategy (typical hemoglobin level <9-10 g/dL)? Other PICO questions addressed red cell transfusion strategies in patients with hematological malignancies and in children. (2) Debate: Jeff Carson vs Simon Stanworth, Moderator Monica Pagano. Given numerous randomized trials compared different hemoglobin concentrations to determine the optimal threshold to make transfusion decisions, one question was whether there was now sufficient trial data to agree if a single common restrictive hemoglobin threshold for red cell transfusion, such as hemoglobin concentration of 7 g/dL, could be generalized for multiple clinical settings. Consistently across studies and clinical settings, restrictive transfusion thresholds have demonstrated similar outcomes when compared to liberal thresholds. However, studies defined the restrictive arm using hemoglobin concentrations with ranges from 7 to 8 g/dL depending on the setting and clinical studies. It is unclear whether differences in hemoglobin concentrations (i.e., 7 vs. 7.5 vs. 8 g/dL) are of clinical relevance and can potentially impact patients' outcomes in all clinical settings. The two speakers will debate these two perspectives providing arguments for and against these options. These issues were extensively discussed within an international panel for an updated AABB sponsored guidelines on the use of red cell transfusions, and the final recommendations will be presented. (3). MINT Trial: The landmark and long-awaited MINT trial results will be summarized and interpretation discussed. 3504 patients with acute myocardial infarction with hemoglobin less than 10 g/dL were randomly allocated to a restrictive transfusion strategy of 7-8 g/dL or liberal strategy to maintain hemoglobin concentration ≥ 10 g/dL. The primary outcome of death or recurrent myocardial infarction occurred in 16.9% in the restrictive and 14.5% in the liberal strategy yielding a relative risk 1.15; 95% confidence interval, 0.99 to 1.34; $p = 0.07$.

PA05-L04 | 2023 National comparative re-audit of NICE quality standard QS138P Davies¹, J Grant-Casey²¹NHS Blood & Transplant, Newcastle, ²NHS Blood & Transplant, Oxford, United Kingdom

Background: The UK's National Institute for Clinical Excellence (NICE) supports the improvement of outcomes for people using NHS and other public health and social care services by producing evidence-based guidance and advice, developing quality standards and performance metrics and providing a range of information services. NICE has developed Quality Standard 138 (QS138), which covers the general principles of blood transfusion in adults, young people and children over 1 year old. It describes high-quality care in priority areas for improvement. This re-audit re-evaluates compliance with the four quality statements set out in QS138 to highlight where practice is deviating from guidelines and identify opportunities for improvement.

Aims: Provide the opportunity to evaluate local evidence of compliance with NICE QS138. Provide data to hospital teams to allow their understanding of what steps they can take to implement Patient Blood Management (PBM) and to measure its effectiveness in improving patient care. Allow the transfusion community to benchmark the progress of PBM and improvements in patient care

Methods: All UK National Health Service providers (Trusts) were invited to take part in the audit. Trusts were allowed to enrol one or more hospitals, so we use the term "sites" to describe those that contributed data. Each participating site was issued with a stationery pack that allowed them to audit up to 40 patients. The audit standards were derived from the statements in QS138, and the audit was divided into four sections, A, B, C & D, and a patient's record could be used for more than one section. Data were collected on transfusions that occurred during January to March 2023.

Results: 1269 sites contributed data, representing approximately 64% (107/167) of UK Trusts. Data from 3710 patients were analysed. 608/1020 (60%) of the patients who were known to have iron deficiency anaemia prior to being admitted for surgery were treated with iron before surgery. 893/1323 (67%) patients undergoing surgery with expected moderate blood loss received tranexamic acid. 756/1195 (63%) patients receiving elective red blood cell transfusions had both their Hb checked and a clinical re-assessment after a unit of red cells was transfused. 470/1346 (35%) of transfused patients had evidence of receiving written and verbal information about the risks, benefits and alternatives to transfusion.

Summary / Conclusions: The re-audit found little evidence of progress towards compliance with the four NICE Quality Statements for Blood Transfusion, and there is still some way to go to achieve uniformly good practice. There remain opportunities to improve patient care, with the potential to reduce length of stay and reduce costs, as well as protecting the blood supply by reducing unnecessary use of red blood cells. Four out of every ten patients known to be iron deficient are not being given iron to combat anaemia, and a third of patients undergoing surgery are not receiving tranexamic acid. Both

of these provide opportunities to reduce use of blood loss and the need for transfusion, a potential reduction in both cost and risk to patients. Work is currently underway to identify the barriers to implementation of QS138 and preliminary results of this investigation will be shared. We will also be able to share the findings from an organisational survey which allowed us to identify the features of service provision that are likely to lead to better compliance with standard.

PA05-L05 | Red blood cell transfusion in the European neonatal intensive care unit

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Background: Red blood cell (RBC) transfusions are often administered to preterm infants in the neonatal intensive care unit (NICU), though robust evidence supporting neonatal transfusion practice is scarce. The extent to which the evidence from two recent trials comparing hemoglobin thresholds for transfusion—the ETNNO trial (Franz,

JAMA, 2020) and the TOP trial (Kirpalani, NEJM, 2020)—has been integrated into clinical practice since publication in 2020 remains uncertain. A prospective study was timely to provide crucial detailed contemporary data on neonatal transfusion practice in Europe.

Aims: To describe prevalence rates, cumulative incidence, indications, volumes and infusion rates, pretransfusion hemoglobin levels, hemoglobin increment, and adverse effects of RBC transfusion in preterm infants admitted to the NICU in Europe.

Methods: We conducted an international, prospective, cohort study across 64 NICUs in 22 European countries. Data collection took place between September 2022 and August 2023, with a six week study period per center. Study outcome measures included prevalence rates, cumulative incidence, indications, volumes and infusion rates, pretransfusion hemoglobin levels, hemoglobin increment, and adverse effects of RBC transfusion.

Results: A total of 1143 infants were enrolled, with a median gestational age at birth of 28+2 weeks (IQR: 26+2-30+2) and a median birth weight of 1030 grams (IQR: 780-1350). During the study follow-up, 396 patients received one or more RBC transfusions, totaling 903 transfusions. Prevalence rates were 3.4 (95CI: 2.7-4.2) and 2.0 (95CI: 1.6-2.6) transfusion days per 100 admission days during day 1-28 and after day 28 of life, respectively. By day 28 of life, 36.5% (95% CI: 31.6- 41.5) of infants had received at least one transfusion. The majority of transfusions were given based on hemoglobin threshold (748/903, 82.8%). Among these, most transfusions had volumes of 15 mL/kg (470/738, 63.7%), durations of 3 h (388/738, 52.3%), and infusion rates of 5-9.9 mL/kg/h (428/738, 58.0%). Hemoglobin levels before transfusion were below the restrictive thresholds set by the ETTNO and TOP trial in 20.8% (152/729) and 42.8% (448/729) of these transfusions, respectively. The median Hb increment per transfusion was 32 g/L (IQR: 22-41). Adverse effects potentially linked to RBC transfusion were documented in 0.22% of transfusions (2/903).

Summary / Conclusions: RBC transfusions are commonly transfused to preterm infants admitted to the NICU, with varying rates, indications, hemoglobin triggers, and volumes across European countries. This study highlighted the existing variation between countries and underlines the need for international collaboration, further research to understand the causes of this variation, and consensus on strategies to optimize neonatal transfusion practices.

Parallel session—blood products

Platelets and other smaller things

PA06-L01 | The procoagulant action of platelet derived microparticles—a comparison between fresh and frozen platelets in a clot formation set-up

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Background: Transfusion of platelets is essential to stop acute bleeding. Cryopreserved platelets can be suitable for use in remote locations and as a backup for situations when the availability of fresh platelets is limited. Previous studies have demonstrated cryopreserved platelets to initiate coagulation faster which is suggested to be partly driven by an increased release of platelet-derived microparticles (PMPs). However, it remains uncertain to what extent these PMPs promote coagulation.

Aims: In this study, we aimed to investigate the influence of PMPs on clot formation in fresh compared to cryopreserved platelets using rotational thromboelastometry.

Methods: A total of 10 platelet units were assessed before and after cryopreservation. For each unit three samples were examined (1) the platelet unit, (2) platelets isolated by centrifugation and resuspended in plasma, and (3) PMP-rich supernatant after centrifugation. Additionally, the expression of phosphatidylserine (PS) on both platelets and PMPs was analyzed with flow cytometry.

Results: Results showed an increase of PMPs in cryopreserved platelets (25% ± 2%) compared to fresh (0.6% ± 0.2%) ($p < 0.001$). Additionally, the proportion of PS+ PMPs and platelets was larger in the cryopreserved units with an average of 55% PS+ PMPs and 44% PS+ platelets compared to 11% and 7% for fresh platelets ($p < 0.001$). The cryopreserved platelets showed delayed clot formation and weaker clot firmness compared to fresh platelets. Further, it was observed that neither the isolated platelets nor the supernatant of

PA06-L01 Table 1

	Clotting time (sec)	Clot formation time (sec)	Maximal clot firmness (mm)
Platelet unit: Fresh	34 ± 3	45 ± 11*	41 ± 11*
Platelet unit: Cryo	41 ± 11	85 ± 27*	32 ± 2*
Isolated platelets: Fresh	44 ± 6	51 ± 8*	44 ± 7***
Isolated platelets: Cryo	41 ± 10	>200*	19 ± 4***
Supernatant: Fresh	48 ± 4	>200	11 ± 4*
Supernatant: Cryo	39 ± 11	>200	20 ± 5*

cryopreserved platelets could reach the same clot firmness as a sample from the platelet unit. This was, however, not the case with fresh platelets where only the supernatant showed a reduced clot firmness (table 1).

Table 1. Rotational thromboelastometry. Comparison between fresh and cryopreserved platelets with samples ($n = 10$) analyzed from either the platelet unit, isolated platelets, or the supernatant. Data demonstrated as mean and SD, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

Summary / Conclusions: The study shows that the increased amount of PMPs in cryopreserved platelets affects clot formation as isolated cryopreserved platelets could not form an equally robust clot like the platelet unit could. This suggests that the platelets and PMPs in cryopreserved units cooperate to maintain the hemostatic action after freezing when the platelet function is likely to be reduced.

PA06-L02 | Intranasal administration of human platelet-derived extracellular vesicles promotes neurogenesis in adult mice

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Background: Platelet-derived-extracellular vesicles (PEVs), which are nanometer-size lipid bilayer vesicles released from platelets, have emerged as promising therapeutic candidates due to their potential benefits in regenerative medicine and as drug delivery vehicles. This functionality is attributed to their cargo of growth factors (neurotrophins, cytokines, antioxidants, miRNA) and the proteins on their membranes. However, translational research on PEVs is evolving, requiring more work to define their functional effects, especially for potential brain applications. In this study, we investigated whether PEVs can stimulate neurogenesis in an ex-vivo neurospheres assay and whether their intranasal administration can promote neurogenesis in young adult mice.

Aims: To (a) explore the ex vivo effects of PEVs on the proliferation and differentiation of neural stem cells (NSCs)/neural progenitor cells (NPCs) derived from the subventricular zone (SVZ) and dentate gyrus (DG) of 8-week-old female C57BL/6 mice and (b) assess the in vivo impact of intranasal administration on short-term proliferative effects and long-term maturation of NSCs/NPCs in SVZ and DG of 8-week-old C57BL/6 mice.

Methods: PEVs were obtained from serum-converted human platelet lysates (SCPL) via size exclusion chromatography using the Sepharose CL-2B column. The ex-vivo assay on neurospheres isolated from SVZ and DG tissues ($n = 5$ /per group) was treated with 1011 PEVs (or PBS control) for 7 or 10 days to assess NSCs/NPCs proliferation. Neurospheres were also treated by PEVs for 7 days to evaluate differentiation using NeuN, DCX, and s100B markers. For the in vivo neurogenesis study, C57BL/6 mice ($n = 12$ /group) received PEVs (or PBS control) intranasally for 3 consecutive days for the proliferation study, and for 28 days (3 times/week) for the maturation study, along with six IP injections of EdU (50 mg/kg). Neuronal proliferation and differentiation were assessed by EdU labeling and co-labeling of EdU + DCX, EdU + NeuN, and EdU + s100B. Imaging was conducted using a TissueFAXS slide scanner and confocal microscopy, with data analysis performed using TissueQuest software.

Results: In the ex-vivo neurosphere assay, PEVs exerted a stronger effect on the neural precursor cells isolated from the DG than from the SVZ, as assessed by the number and diameter of the neurospheres generated in the proliferation assay as well as the differentiation into neuronal cells. In vivo, we observed that PEVs had a more proliferative effect in the SVZ during the short term, following 3-consecutive days of intranasal (i.n) administration as compared to the DG. Moreover, PEVs treatment led to a higher number of co-labeled (EdU + NeuN), signing neuronal differentiation in both the DG area as well as in the olfactory bulb-granular cell layer (OB-GCL) after day 28 of (i.n) administration. Notably, co-expression of EdU + DCX and EdU + s100B showed fewer numbers in the same areas after long-term PEVs (i.n) administration.

Summary / Conclusions: These data indicated that EVs present in human platelet lysates have neurogenic capacities and might serve as a stand-alone biotherapy to support neurogenesis. Pending further studies, PEVs may potentially help to counter-balance cognitive defects in neurodegenerative pathologies, trauma, or during aging.

PA06-L03 | Cold-stored platelets retain the capacity to release soluble factors in response to haemostatic and immunologic stimuli for 21 days

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Background: Cold storage (refrigeration) of platelets offers key advantages over storage at room-temperature (RT), including a longer shelf life of at least 14 days, with better preservation of platelet haemostatic function. Notably, the supernatant of cold platelets contains a lower concentration of soluble factors throughout storage compared to RT platelets, suggesting retention within the platelet granules during cold storage. However, it is currently unclear if cold platelets maintain the capacity to release granular stores in response to stimulation following longer storage.

Aims: The aim of this study was to compare the release of soluble factors from RT and cold platelets in response to both haemostatic and immunological stimuli.

Methods: Two whole-blood derived platelet concentrates (30% plasma/70% SSP+) were pooled and split into equivalent components for storage at RT (20–24°C) with agitation or under refrigerated conditions (2–6°C without agitation; cold). Samples were taken on day 1, 7, 14 and 21 of storage, and the supernatant was collected from unstimulated samples or samples activated with TRAP-6 (10 µM), A23187 (10 µM), lipopolysaccharides (20 µg/mL) from *Escherichia coli* (O111:B4) or recombinant Histone-H4 (30 µg/mL) for up to 30 min at 37°C. Commercially available ELISAs were used to measure the concentration of soluble factors in the supernatant. The change in concentration was calculated by subtracting the soluble factor concentration in unstimulated samples from the stimulated samples at each time point throughout storage.

Results: As expected, unstimulated cold platelets contained lower supernatant concentrations of all the soluble factors examined at day 21, compared to RT. TRAP-6 stimulation caused cold platelets to release significantly higher amounts of RANTES and CD40L than RT platelets (Table 1). Cold platelets stimulated with A23187 released significantly higher concentrations of RANTES, CD40L and PF4 than RT platelets (Table 1). LPS stimulation resulted in the release of IL-27 and PF4, but not other soluble factors. LPS-induced IL-27 (220 ± 152 pg/mL, $p = 0.0142$) release was only observed during RT storage; and PF4 (949 ± 519 ng/mL, $p = 0.0002$) only during cold storage. Histone-H4 induced cold platelets to release RANTES, CD40L, IL-27 and PF4, but RT platelets were unresponsive by day 21 (Table 1). The concentration of TNF-α and OX40L was not measurably altered over 21 days regardless of storage temperature.

Summary / Conclusions: Cold platelets maintained the capacity to release soluble factors in response to haemostatic and immunological stimuli after 21 days of storage. Further, the concentration and composition of these soluble factors was dependent on the activating agent. Preserving the responsiveness of platelets to stimuli may better facilitate their haemostatic and immunological function post-transfusion.

PA06-L04 | Lyophilized human platelet lysate from platelet concentrates for cell biology research and manufacturing of cell therapy products

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Background: A significant number of platelet concentrates (PCs) is discarded daily in blood banks due to limited shelf life. With a decline in blood donations, there is an economic and ethical incentive to utilize these expired PCs. Human platelet lysate (HPL), derived from expired PCs, can serve as an ethical and sustainable alternative to fetal calf serum (FCS) in biomedical research and cell therapy production. HPL is commercially available as a cell culture substitute while still being subject to decisive disadvantages such as a high cost, batch differences, and a lack of storage stability. We developed a method to produce HPL under Good Manufacturing Practice (GMP) conditions for cell culture in basic research and the production of cell-based therapeutics. We follow the overall aim to enhance the applicability of HPL by lyophilization (L-HPL) leading to increased storage temperature and shelf life, while also minimizing batch variations for successful cell expansion and therapeutic applications. However, the influence of HPL lyophilization on key quality parameters remains elusive.

Aims: To investigating the influence of HPL lyophilization on parameters of quality control, growth factor concentrations, and the culture of human mesenchymal stromal cells (hMSCs).

Methods: Four pooled PCs, consisting of cell material from four donors each, were lysed by four successive freezing and thawing cycles. The cell debris was then separated by centrifugation ($4000 \times g$, 10 min) and the supernatant was frozen and lyophilized. We compared six batches of HPL and L-HPL regarding their quality parameters pH, total protein, osmolality, sodium, potassium and chloride concentration. Concentrations of 13 growth factors (e.g., TGF-β1, VEGF, bFGF) were compared between HPL and L-HPL. Additionally, we determined the cell yield and proliferation capacity of hMSCs

PA06-L03 Table 1. Change in soluble factor concentration following stimulation after 21 days of storage

Cytokine	Storage temperature	TRAP-6	A23187	Histone-H4
RANTES	RT	5 ± 9	1 ± 2	0
(ng/mL)	4°C	38 ± 10*	49 ± 26*	67 ± 2*
CD40L	RT	13 ± 33	18 ± 46	0
(pg/mL)	4°C	111 ± 41*	193 ± 207*	264 ± 268*
IL-27	RT	146 ± 106	64 ± 90	0
(pg/mL)	4°C	21 ± 30	188 ± 165	352 ± 253*
PF4	RT	1149 ± 730	190 ± 294	313 ± 626
(ng/mL)	4°C	1527 ± 1065	3508 ± 1893*	4851 ± 1068*

* $p < 0.05$ compared to RT platelets.

following expansion in cell culture media containing 10% FCS, 7% HPL and 7% reconstituted L-HPL as well as the potential of hMSCs for adipogenic, chondrogenic and osteogenic differentiation following expansion in cell culture media containing 7% HPL and 7% reconstituted L-HPL.

Results: The quality parameters pH, total protein, osmolality, sodium, potassium and chloride concentration and growth factor concentrations are comparable between HPL and reconstituted L-HPL (e.g., total protein: HPL 32.0 ± 1.7 g/L vs. L-HPL 28.0 ± 1.0 g/L; TGF- β 1: HPL 9.1 ± 6.6 vs. L-HPL 11.8 ± 6.4). Cell yield and proliferation capacity of hMSCs was significantly enhanced following culture in media containing HPL or reconstituted L-HPL if compared to cells cultured in FCS containing medium. Significant differences between hMSCs cultured in media containing HPL or reconstituted L-HPL in terms of proliferation capacity and differentiation potential were not detected.

Summary / Conclusions: We successfully developed a GMP-compliant method to produce well-applicable lyophilized HPL from expired PCs. The comparison of L-HPL with HPL revealed comparable quality parameters and growth factor concentrations, demonstrating the viability of L-HPL as a cell culture substitute. Notably, culture in media containing HPL or L-HPL proved increased cell yield, offering a more efficient and economical approach in comparison to FCS. These findings emphasize the potential of L-HPL for both basic research and therapeutic applications, providing a sustainable and ethical alternative in biomedical research and drug development.

PA06-L05 | Human platelet pellet lysates and extracellular vesicles—a potential biotherapy to protect mitochondrial dysfunction in neuronal cells

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Background: The utilization of human platelet pellet lysates (HPPL) and, more recently, platelet-derived extracellular vesicles (P-EVs) in cellular therapies and regenerative medicines is driven by their pleiotropic content of bioactive trophic factors, including antioxidants. Experimental evidence demonstrates the therapeutic potential of these platelet-derived preparations in the treatment of CNS disorders. Mitochondrial impairment is a key player in neurodegeneration, given that neurons, with their high energy demands, depend heavily on the efficient functioning of mitochondria. Platelet trophic factors may thus aid in cellular energy production involved in mitochondrial function. Future translational application needs to determine the functional role of HPPL and

P-EVs in maintaining mitochondrial health in neurodegenerative disorders.

Aims: The present study aims to explore the potential of HPPL and P-EVs in addressing mitochondrial dysfunction, with a primary focus on cellular bioenergetics and oxidative stress.

Methods: HPPL and P-EVs were isolated using differential centrifugation from clinical-grade apheresis platelet concentrates (PCs). They were characterized for their protein and growth factors and their physical properties were assessed using nanoparticle tracking analysis, dynamic light scattering, cryo- transmission electron microscopy respectively. Surface marker analysis was performed on P-EVs using western blot. N2A and SH-SY5Y neuroblastoma cells were incubated with or without 5% (v/v) HPPL and P-EVs labeled with Alexafluor-488 followed by mitotracker red fluorescent staining to visualize PEV co-localization in mitochondria. Cells were differentiated and pre-treated with 5% (v/v) HPPL and P-EVs followed by a 24-h exposure to 10 μ M Rotenone (an inhibitor of complex I in the mitochondrial respiratory chain). Cell viability was assessed by CCK8 assay, cellular metabolism by ATP and lactate, and oxidative stress using DCFDA and MitoSox dye for mitochondrial-specific superoxide.

Results: HPPL and P-EVs had a similar composition in trophic factors with high absolute anti-oxidative capacity. P-EVs showed expected nanometer size (100–200 nm), particle number (10^{11} – 10^{12} EVs/mL), and CD9, CD41, and CD63 surface membrane expressions. Laser confocal microscopy images showed a clear overlap of Alexafluor-488 and Mito-Tracker red suggesting the HPPL and PEVs co-localization within the mitochondria of cells. Cell viability assay confirmed a lack of cytotoxicity and significant neuroprotective effects of HPPL and P-EVs in rotenone-exposed N2A and SH-SY5Y cells. DCFDA and MitoSOX Green staining revealed increased cellular ROS and mitochondrial superoxide in the rotenone pre-treated cells. Both HPPL and P-EVs pre-treatment exposed to rotenone significantly reduced the fluorescence intensities suggesting substantial anti-oxidant ability. Moreover, pre-treatment with HPPL and P-EVs of rotenone cultured cells increased ATP and reduced lactate levels compared to rotenone-treated cells confirming the regulation of cellular bioenergetics of N2A and SH-SY5Y cells.

Summary / Conclusions: These findings provide valuable insights into the therapeutic effects of HPPL and P-EVs against mitochondrial dysfunction associated with neurodegeneration. This opens the door for further exploration of platelet-derived biotherapy on mitochondrial function in disease modeling.

PA06-L06 | Platelet factor 4 enhances resistance to Candida albicans infection by inhibiting macrophage PANoptosis

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Background: *Candida albicans* (*C. albicans*) is the most common human conditionally pathogenic fungus which could lead to severe inflammatory response and tissue damage, and it is the most lethal form with mortality rates exceeding 45%. Macrophages are important for host defenses against infection which can phagocytose pathogens and produce antimicrobial cytokines to amplify the immune response. It has been gradually recognized that platelets are involved in infection through the secretion of reactive antimicrobial proteins and chemokine cytokines, for example, platelet factor 4 (PF4). We performed a retrospective analysis of 337 patients with systemic fungal infections in our hospital and found that patients combined with thrombocytopenia had a longer mean hospital stay, a higher mean number of hospitalizations, and a worse prognosis compared with those patients with normal platelets.

Aims: To clarify the role of platelets in the immunization against *C. albicans* infection, to investigate the interactions between platelets and macrophages in the fight against *C. albicans* infection, and to explore how platelet-macrophage signaling interactions affect the *C. albicans* infection.

Methods: A systemic infection platelet-deleted mice model were developed by intravenous injection of *C. albicans* to explore the role of platelets in immunity against *C. albicans* infection. Western blotting was used to analyze key molecules of regulated cell death patterns to explore the effects of *C. albicans* infection on macrophage survival and specific modes of death. Flow cytometry was used to examine the effects of PF4 on macrophage survival and phagocytosis in mice model of *C. albicans* infection which were pre-treated with PF4. *Pf4* knockout mice were used to investigate the role of PF4 on *C. albicans* systemic infection.

Results: After *C. albicans* systemic infection, platelet-deleted mice were found to have a shorter survival time and significantly increased fungal loads; the percentage of macrophages in the peripheral blood of platelet-deleted mice were significantly reduced, and the percentage of macrophages phagocytizing *C. albicans* were also significantly reduced compared with wild type control mice. PF4 pre-treatment resulted in the simultaneous inhibition of the molecular hallmarks of apoptosis, pyroptosis, and necroptosis in *C. albicans*-induced generated macrophages, with a reduced level of cell death and IL-1 β secretion (suggesting the occurrence of PANoptosis). After PF4 treatment, the survival time of systemic infection mice model were found prolonged, and the fungal loads decreased. In addition, the phagocytosis level of *C. albicans* by macrophages in the peripheral blood of mice were increased after PF4 treatment. *Pf4* knockout mice infected with high doses of *C. albicans* via intravenous infection were found resistance to *C. albicans* infection diminished compared with wild type control mice.

Summary / Conclusions: Our data suggested that platelet-derived PF4 could inhibit macrophage PANoptosis to avoid excessive inflammatory response and may improve the prognosis of infection.

Parallel session—management and organisation

Thinking outside the blood box

PA07-L01 | Building a sustainable blood service

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In 2024, a sustainable blood service has two meanings; it relates to providing a secure service that ensures blood components and other deliverables are always available for the patients and, at the same time, operating in a manner that minimises environmental impact. It is not sufficient to consider “status as of today”—it is also critical to include both aspects in planning for the future. In this presentation, we will first go through the basic requirements for a sustainable blood service and then present a practical example of an environmental project: The Irish Blood Transfusion Service “Green Lab Initiative”. The key points for a sustainable blood service are: (1). Engage Executive Management Team and Board (2). Donor recruitment strategy directed to the diverse population (3). A sufficient donor pool—covering the needs for blood components without over-bleeding the donors (4). Donation facilities that are easily accessible for the donors (5). A production facility that allows streamlined procedures (6). A testing facility that is able to adjust to an epidemiological situation (7). Storage and distribution systems that ensure safe and prompt delivery to hospitals (8). Affinity with clinical users to ensure optimal use of the blood components—Patient Blood Management (9). Horizon scanning—how to engage in activities outside the traditional blood service such as cellular therapies (10). Develop strong research and innovation activities (11). Engage with international organisations (12). Engage in global health activities The “My Green Lab Certification” was founded in the USA to enable scientists to become active contributors in minimising the environmental impact of their laboratory operations. In 2021, an IBTS project started in one of the laboratories. Ambassadors were selected and trained, and staff was subsequently engaged. All activities, including working with suppliers, were included. A year later, the laboratory was “Green Lab certified”, and significant achievements were achieved. The process is now being rolled out in all IBTS laboratories.

PA07-L02 | Doing more with less—the impact of a contextualized hospital transfusion committee in clinical services

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Background: The ability to effectively coordinate the blood transfusion chain from donors to recipients depends on many factors among

which a hospital transfusion committee. This committee should be represented by all stakeholders of this chain such as transfusion medicine consultants, clinicians where blood is used, hospital administrator, and a quality manager among others. Baptist Hospital Mutengene (BHM), is a faith-based institution licensed by the Ministry of Public Health and supervised by the National Blood Transfusion Service (NBTS) to offer services in blood donor recruitment, collection, and hospital transfusion in Cameroon. In this light, a hospital transfusion committee was created in December 2021 comprising 6 people: a transfusion medicine specialist (1), the head of the department of the laboratory (1), the coordinators of the blood bank (2), a representative of the voluntary donors' association (1) and a religious leader of the community (1).

Aims: We sought to evaluate the impact of the activities of the hospital transfusion committee in blood transfusion services with regard to four groups: patients, voluntary blood donors, healthcare staff and the hospital.

Methods: We conducted a retrospective evaluation of the activities of the BHM, hospital transfusion committee between January 2022 and December 2023. The committee had between 1 and 3 monthly sessions lasting 45 to 90 min each. Monitoring and evaluation of each member's tasks and assigning new ones in line with the main objectives constituted the main activity of these meetings. Reports post each meeting, were submitted to the hierarchy. There were seven main activities of the committee which included: revamping the voluntary blood donors association, revising the documentation used in transfusion practice, developing local policy/guidelines in the transfusion service, organizing transfusion education sessions, prospective audit of blood use (hemovigilance inclusive), initiating patient blood management application and blood component preparation.

Results: In the patient category, specific conditions such as symptomatic thrombocytopenia and hemophilia patients were better managed and there was an estimated reduced out-patient healthcare expenditure of 11,720 USD. Concerning blood donation, the voluntary blood donation rate increased from 8.2% to 13.4% and a turn-around time for donors reduced from averagely 90 to 40 min between 2022 and 2023. Two major policies on donors' health were approved by the hospital administration on anemia and malaria prophylaxis. With regards the healthcare staff and hospital, there was an 18.9% reduction in blood collected, 24% reduction in type and crossmatch only orders, 22.2% reduction in the total number of transfusions and 44.3% decrease of inappropriate transfusions between 2022 and 2023. This led to over 2651 h (110 days) of work saved among healthcare staff. 54.4% of the gap between demand and supply of blood components was reduced, the safety margin of blood components increased by 5.2% and 0.8% of in-patient hospital admissions were referrals due to unavailability of blood in those facilities. Moreover, lack of traceability of blood components reduced from 1.5% to 0.12%.

Summary / Conclusions: Contextualizing the hospital transfusion committee to suit its blood transfusion system organization, would optimize its services from donor to recipient with available resources. This potentially benefits all stakeholders in the transfusion chain.

PA07-L03 | A comparative study of current blood matching guidelines and optimised algorithmic allocations to achieve a healthier future

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Background: Annually, NHS Blood and Transplant (NHSBT) collects 1.4 million units of donated blood to treat around 500,000 patients. Current transfusion guidelines emphasise the need for compatibility between the blood types of the patient and the transfused units for the ABO groups and the D type. They also recommend matching of C, c, E, e and K types in frequently transfused patients. Despite these recommendations, sensitisation against minor blood group antigens remains a significant problem, causing harm to nearly 18,000 regularly transfused patients in England each year. In anticipation of a policy of extended matching, NHSBT has begun genotyping large numbers of its donors using a new blood typing array. NHS England is also funding the genotyping of all haemoglobinopathy patients in 2024. Machine learning based matching is therefore essential to optimise blood allocation and bring the benefits of this dense genotyping data to the patients. However, matching algorithms, which aim to minimise immunisation at the population level, often deviate from current matching guidelines. These differences must be investigated before decision support algorithms can be safely integrated into routine matching.

Aims: We aim to investigate the differences between optimised algorithmic blood allocations and allocations following current matching guidelines.

Methods: We developed the Blood Matcher framework (a customisable and standardised software platform for algorithmic blood allocation) to consider three extended antigen matching strategies. These strategies took distinct approaches to the allocation of ABO and D types: first strategy fully optimised matching across all antigens by allocating compatible types; second strategy followed current guidelines for exact matching of the ABO and D antigens (i.e. D+ donor matched to D+ patient, rather than D- substitutions except in presence of antibodies); and third strategy matched exactly for ABO and D, then allocated compatible blood to meet residual demand. To assess their performances, we conducted a comparison using real-world data comprising 5708 requests and 2800 allocations of cDe/cDe (R0R0) blood units from 11 NHSBT stock holding sites to 911 patients.

Results: The first strategy was able to provide compatible R0R0 extended matched blood for 99.95% (5705/5708 units) of orders compared to the second strategy, which allocated blood for just 84.39% of orders (4434/5708 units). This was revealed to be mainly due to the overspecification of allocated units, defined by the fulfilment of requests with extensively typed or rare blood beyond order requirements, as we calculated an average overspecification score of

18.9 ± 2.7 for the fully optimised (first) strategy versus 20.4 ± 4.5 for the current guidelines (second) strategy. The third strategy fulfilled 95.26% of orders (5340/5708) with a 21.8 ± 3.4 overspecification score.

Summary / Conclusions: We show that machine learning based matching strategies can significantly improve blood allocation and simplify blood stock management. However, the current guidelines, which force exact ABO and D matching for all allocated units to prevent mixed-field reactions on subsequent transfusions, come at a cost with respect to blood provision. As an increasing number of regularly transfused patients have their extended genetically measured blood types available, the current guidelines should be re-evaluated to ensure algorithmic matching can benefit patients.

PA07-L04 | Unveiling intraday blood transfusion patterns—implications for resource optimization in Finland's largest hospital district

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Background: Efficient blood supply chain operation is crucial for patient care and resource management, but currently in Finland, there is very little visibility within and outside hospitals into the dynamics behind blood demand, complicating the management of the blood supply chain. Helsinki University Hospital HUS represents the largest hospital district in Finland, transfusing almost a third of all the blood in Finland. Understanding drivers of blood demand in HUS is the first step towards better demand forecasts and a more efficient blood supply chain. As a first example we have inspected intraday dynamics.

Aims: The purpose of this study was to analyze blood transfusion timings and their alignment with current understanding about intraday blood demand, scheduling of elective surgeries, and resource allocation within HUS. As a part of larger study modeling and predicting blood product demand in HUS, our goal was to describe daily patterns in blood use and identify strategies for optimization.

Methods: In this retrospective analysis of electronic health records, we obtained data from HUS data pool on all adult patients treated in HUS between 2021 and 2022, and focused on transfusion recipients. After careful data matching and filtering, we analyzed and summarized over 109,000 transfused units, with a special focus on the intraday timestamps of transfusions. The study categorized blood products into fresh frozen plasma (FFP), platelets (PLT), and red blood cells (RBC), analyzing their utilization throughout the day using hourly means and medians.

Results: A significant number of transfusions occurred outside of the office hours, expected to be the busiest for the hospital blood bank. This indicates substantial blood resource utilization occurs during periods not traditionally associated with elective surgery or hematological patient care. When comparing hourly medians, nighttime

demand for RBC products is approximately half of the daily maximum, and the demand is low in the morning office hours.

Summary / Conclusions: Our research provides rarely accessible timestamp precision for transfusion events in Finland's largest hospital district, uncovering patterns that question the established resource allocation at the HUS blood bank. This investigation reveals gaps in our understanding of intraday blood demand dynamics, suggesting a reevaluation of resources in blood product storage and scheduling. Ongoing exploration of other drivers of demand dynamics likely opens avenues for predictive modeling of blood demand, ultimately improving patient care and enhancing resource management.

PA07-L05 | Revolutionizing plasma quality control—a novel AI-enhanced approach for accurate plasma product assessment

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Background: In transfusion medicine, ensuring the quality of plasma products is critical. The traditional reliance on subjective assessments by medical technologist staffs can lead to variability in quality control (QC). This research introduces an automated image processing system to standardize and enhance the QC of plasma products.

Aims: The aim is to develop an automated system using image processing and machine learning to improve the accuracy and objectivity in QC of plasma products.

Methods: The methodology involves the following steps: (1). Plasma Separation: Conducting plasma separation using standard procedures. (2). Sample Selection by Senior Staff: Three experienced staff members select plasma bags with normal and abnormal colors or turbidity to train the AI. Their selections are based on a consensus agreement. (3). AI Training and Development: The AI is trained using the selected plasma bags. A machine learning model is developed to distinguish between normal and abnormal plasma. (4). Evaluation of Accuracy: The AI's accuracy is tested using additional plasma bags chosen and consensually classified as normal or abnormal by the three senior staff members.

Results: The initial training phase with a set of 200 plasma bags yielded an accuracy of 85%. Upon expanding the dataset with 486 images (287 normal and 199 abnormal), the system's accuracy improved to 94.60%. This performance indicates a high level of reliability in the AI's ability to correctly classify plasma bags, aligning closely with the consensus of the experienced medical staff.

Summary / Conclusions: The implementation of an automated image processing system in the quality control of plasma products marks a significant advancement in transfusion medicine. This technology promises to standardize quality assessments, reduce subjective variability, and enhance the overall safety and reliability of plasma products.

Parallel session— immunobiology

New horizons in blood group genomics and regulatory mechanisms

PA08-L01 | Dense HEA, HPA and HLA genotyping of 82,000 blood donors using a genotyping service embedded in a national blood supply organisation

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Background: Blood supply organisations must type a portion of their blood donors for a range of erythrocyte (HEA), platelet (HPA) and leucocyte (HLA) antigens to meet the need for compatible components for patients who have become immunised by transfusion or pregnancy. NHS Blood and Transplant (NHSBT), as part of the Blood transfusion Genomics Consortium (BGC), has set up a genotyping service in their accredited molecular laboratories to evaluate the performance of two DNA microarrays, the UKBBv2.2 and UBDT PC1 AxiomTM arrays, capable of extended antigen typing of both donors and patients. The UKBBv2.2 array, in addition to antigen typing, can type the genetic variants required for population genomics studies. This has allowed NHSBT to form a world-first partnership with the NIHR BioResource to provide genotyping services for the NIHR STRIDES study in which 82,000 NHSBT donors have consented to be genotyped.

Aims: To investigate the feasibility and benefits of at-scale genotyping for HEA, HPA and HLA as part of a routine genotyping service.

Methods: Samples from 82,000 blood donors were typed using the UKBBv2.2 array using a GeneTitan-MC instrument. The automated, integrated analysis package (IAP v1.0), consisting of the AxiomTM Best Practices, LiteTyper and HLA:IMP*02 algorithms, was used to analyse data from the first 21,882 samples. The ancestry of donors was inferred by principal component analysis and the use of a Gaussian Mixture Model. The array's HEA, HPA and HLA typing data was compared to electronic donor records to assess accuracy. Finally, several queries were designed to identify donors with in-demand phenotypes within the dataset.

Results: After performing genotype calling, contamination, and declared vs. genetic sex quality control analyses, 21,444 (98%) samples were suitable for downstream analysis. Genetic ancestry estimation identified samples were from 16,602 (82.2%), 1966 (9.1%), 346 (1.6%), 666 (3.1%), 575 (2.9%) donors of European (EUR), South Asian (SAS), East Asian (EAS), Admixed American (AMR) and African (AFR) ancestry respectively. 1,251,732 HEA (52 antigens), 300,442

HPA (4 antigens), and 128,664 HLA types (class I and II) were generated from the data—an 8.5 fold increase from 196,496 to 1,680,838 antigen types. HEA and HPA typing concordance with previous results was 99.9% in 186,672 comparisons across 44 antigens. 10,176 donors had a homozygous genotype at the *RHCE* locus, and 5.9%, 25.0%, and 71.6% of EUR, AMR and AFR donors were R0R0 (D+C-c+E-e+) with 537 being Fy(a-,b-) and 6 being U-. With respect to rare phenotypes, 161 donors were negative for one or more of the following Lu(b), k, Js(b), Yt(a), Co(a), McC(a), Kn(a) and Vel. 502 donors were homozygous across HLA class I, and 350 HPA-1b,1b; HPA-5a5a donors were identified.

Summary / Conclusions: We show that at-scale genotyping can be embedded in a national blood supply organisation by genotyping 82,000 DNA samples and analysing data from the first 21,882 samples. Concordance between array inferred antigen types and previous typing was excellent at 99.9% and dense typing data allowed for rapid identification of 11,189 donors with high-demand phenotypes suitable for patients with HEA, HPA or HLA alloantibodies. The observation that almost 1 in 5 donors were non-European shows that preferentially seeking consent for DNA-based typing from African, mixed American and Asian donors can be operationalised at scale. Genotype data from the remaining 60,118 donors is being analysed and will be available in March 2024.

PA08-L02 | The potential of adaptive sampling in blood group genomics—a new horizon?

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Background: Adaptive sampling (AS), a computational enrichment method from Oxford Nanopore Technologies (ONT), could mark a significant advancement in blood group molecular diagnostics. Leveraging the power of long-read sequencing, it shows considerable promise in accurately identifying complex structural variants (SVs) in the RH and MNS blood group systems—areas historically challenging for conventional diagnostic techniques. Moreover, AS provides a novel avenue for comprehensive analysis of the entire blood group genome, highly relevant for patients requiring chronic transfusions, such as those with thalassemia or sickle cell disease. Complementing method for high-throughput genotyping of donors, sequencing by AS aims at accurately matching patient and donor profiles, which could reduce risks associated with alloimmunization and delayed hemolytic transfusion reactions.

Aims: This study aims to evaluate the effectiveness of AS in resolving complex SVs in the RH and MNS systems while in parallel assessing its ability to provide complete and precise sequence information for all blood group genes.

Methods: We assessed AS using four distinct samples: a sample with a suspected deletion in the *RHCE* gene, one with a suspected *RHD*01N.06* hybrid allele, one with an unresolved hybrid allele in the MNS system, and finally a sickle cell patient with an unresolvable SV in the RH system. Sequencing was performed on ONT's PromethION P2 solo platform using the latest V14 chemistry. The reference FASTA file, conveying the genomic regions to enrich, contained all known red cell blood group genes ($n = 51$), 2 transcription factors, 7 platelet, and 4 neutrophil antigen genes. We included 50 kb flanking regions for each locus to increase chances of retrieving long on-target reads. In sum, we targeted ~ 8.6 Mb; corresponding to $\sim 0.27\%$ of the human genome. Reads were mapped to the novel human reference genome (T2T-CHM13v2.0), and variants called using Clair3 and Sniffles2. To assess variant calling accuracy, ONT sequencing results were compared to pre-typed genetic data for up to 17 blood groups systems.

Results: Raw sequencing output ranged from 40.1 to 50.9 Gb per PromethION flowcell. Mean coverage for the targeted genomic regions was between $37.4\times$ and $46.6\times$. The decision for on- or off-target during sequencing was taken on average after ~ 650 bp, corresponding to approximately 1.5 s. N50 values fluctuated between 18 and 22 kb for the 4 libraries, with the longest read being 684,247 bp long. In the first sequenced sample, a perfect concordance was observed on 73 pre-typed variants spread across 17 blood group systems. For this same sample, long reads across the *RHCE* gene ($45.3\times$ coverage; 29 reads > 25 kb; max length of 98 kb) facilitated the detection of a novel 8,636 bp deletion spanning from intron 8 to intron 9 of *RHCE*, representing to our knowledge the largest deletion ever reported in this gene. Detailed analyses for the three remaining cases will be presented at the congress.

Summary / Conclusions: AS represents a significant advancement in the genomic characterization of blood group systems, offering a powerful tool for resolving complex SVs as well as providing access to the whole blood group genome. Its application has proven to be exceptionally versatile and accurate, providing a new avenue for the comprehensive genetic analysis of blood group genes. This method holds promise for improving patient outcomes, particularly for those requiring chronic transfusions.

PA08-L03 | Characterization of erythroid transcription factor binding profiles across blood group genes to explain variations in antigen expression

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Background: Blood group expression varies qualitatively and quantitatively. Quantitative expression is affected by transcription factor (TF) binding to regulatory sites (RS). Loss of TF binding can result in weakened expression as in the Helgeson phenotype (Knops system), or abolished expression as in Fy_{null} . Most binding sites for relevant

TFs in blood group genes have not yet been characterized, and the relationships between TFs and their predicted binding sites not established. To date, the binding site thought responsible for KLF1 in the In(Lu) phenotype has not been located, and recently some variants in the coding sequence assumed to cause JK weak or null phenotypes were questioned and researchers are now seeking explanations in non-coding regions.

Aims: We aim to improve the prediction of RS in blood group genes by characterizing the TF binding profiles and study the relationship of TF binding and phenotype with functional assays.

Methods: Raw ChIP-seq data for TFs GATA1, KLF1, RUNX1 and NFE2 were obtained from experiments in primary erythroblast, analyzed with the nextflow 'chipseq' pipeline and annotated using the ChIPseeker package in Rstudio. The peaks were filtered to include binding 10 kbp up- or downstream of the 51 known blood group-associated genes. Characterization of TF binding was annotated by the ChIPpeakAnno package using the 'TxDb.Hsapiens.UCSC.hg38.knownGene' library. Known antigen variations were examined and TF binding sites were confirmed by electrophoretic mobility shift assay (EMSA).

Results: The TF peak profile showed that exons and introns contain the most binding sites regardless the TF (Tables 1 & 2). The 2nd most abundant region for KLF1 and RUNX1 was in the promoter, in line with their role in direct recruitment of RNA polymerase. GATA1 had its 2nd highest binding in the distal intergenic regions, suggesting GATA1 has higher potential to serve as both proximal and distal regulator. For the distribution across exon/intron/intergenic regions, all TFs had the most binding sites in the introns, except NFE2, indicating introns most likely to serve as RS. GATA1 showed more binding sites in intergenic regions than in exons. In exons, most sites are found in the 5' untranslated region (UTR) than other regions for all TFs,

PA08-L03 Table 1.

	GATA1 (%)	KLF1 (%)	RUNX1 (%)
Promoter	12.7	28.6	26.5
Downstream	2.3	4.1	2.9
Gene body	61.7	55.1	50
Distal intergenic	23.3	12.2	20.6

PA08-L03 Table 2.

	GATA1 (%)	KLF1 (%)	RUNX1 (%)	NFE2 (%)
Exon	16.1	29.6	29.4	38.1
5'UTR	7.3	23.5	17.6	21.4
3'UTR	3.5	2	8.8	7.1
coding	2.6	3.1	NA	4.8
other	2.7	1	2.9	4.8
Intron	56	48	47.1	35.7
Intergenic	28	22.4	23.5	26.2

denoting higher regulatory potential at the start of the gene than the end. To our surprise, there were no KLF1 binding sites observed within or close-by the *BCAM* (*LU*) gene, suggesting a more complex mechanism behind the *In*(*Lu*) phenotype than just partial loss of KLF1 binding to *BCAM*. GATA1, KLF1 and RUNX1 showed co-occupancy at a site ~1 kbp upstream of *SLC14A1* (*JK*). We validated GATA1 binding with EMSA, and loss of binding when the variant rs1375999702:A>T is introduced.

Summary / Conclusions: Our study shows how binding of selected TFs is distributed. This enable us to focus on the regions that are most likely to serve as RS for these genes. Findings provide insight into the blood group regulome in general, and may help explain how *JK* and *LU* expression is regulated in particular. *SLC14A1* promoter region with 3 TFs binding may affect the expression of *JK* antigens. However, no proximal KLF1 binding to *BCAM* was observed.

PA08-L04 | Functional analysis of KLF1 mutations within the non-DNA-binding regions in *In*(*Lu*) individuals

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Background: *In*(*Lu*) is a rare blood group primarily caused by heterozygous mutations in the KLF1 gene, which result in reduced or loss of function. The KLF1 missense mutations that cause the *In*(*Lu*) phenotype are predominantly found in the DNA-binding domains, specifically in the three zinc finger regions. However, there are a few exceptions. Although several functional studies have been conducted on missense mutations in the zinc fingers of KLF1, there is still a lack of functional evidence for missense mutations in the non-DNA-binding regions, such as zinc finger linkers.

Aims: In this study, we aimed to investigate the mechanism of three KLF1 missense mutations located in the zinc finger linkers, reported by our group previously from *In*(*Lu*) individuals (KLF1*916A, KLF1*1010A, and KLF1*1013T).

Methods: The full-length KLF1 coding sequences of wild type and three mutant types were cloned into the pcDNA3.1 vector, respectively. In addition, the known *BCAM* (Lutheran blood group glycoprotein coding gene) and HBB promoter regions, which represent KLF1 binding motifs, were cloned into the pGL3-BASIC vector, respectively. Different groups were established, including a blank control, empty vector controls, co-transfection of different recombinant KLF1 vectors and promoter motif vectors with the pRL-TK vector. The HEK293T cells were transfected and the dual-luciferase reporter assay was carried out 48 h after transfection using a commercial detection kit. The constructed wild-type and three mutant pcDNA3.1 recombinant vectors, as well as the wild-type and mutant pEGFP-C1 recombinant vectors constructed in a similar way, were separately transfected into two groups of HEK293T cells. After 48 h of transfection, cells in one group were treated with Hoechst stain and nuclear localization was observed under confocal fluorescence microscopy. Nuclear protein was extracted from another group of cells to detect

KLF1 protein expression using western blot with GAPDH as an internal reference.

Results: All recombinant vectors were successfully constructed. Three missense mutations in the KLF1 zinc finger linkers have been identified to inhibit the interaction between KLF1 transcription factor and the promoter of *BCAM*. However, these mutations do not affect the interaction between KLF1 and the promoter of HBB. Additionally, it has been observed that these three missense mutations do not modify the nuclear localization or expression level of KLF1.

Summary / Conclusions: The conducted functional studies on three KLF1 mutations indicate that these mutations could potentially be responsible for the *In*(*Lu*) phenotype. It is worth noting that these mutations only partially affect the function of the transcription factor KLF1 and have different effects on different target genes. Considering the diverse range of KLF1 mutation types and their varied effects, it is crucial to conduct functional characterization of different mutations.

PA08-L05 | Identification of novel and rare blood group variants leading to null phenotypes in red cell alloimmunized patients using targeted next generation sequencing—Indian experience

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Background: Red cell alloimmunization due to presence of pan-reacting or nonspecific blood group (BG) alloantibodies creates serologic incompatibility and makes selection of compatible blood units for transfusion therapy difficult. In Indian patients, about 14%–36% specificity of antibody is not identified. Currently, extensive serology followed by simple genotyping assays are used to characterize these antibodies. However, the utility of these genotyping assays is limited when blood phenotypes are result of novel variants. Alternatively, targeted Next Generation Sequencing (T-NGS) based approaches have been successfully applied for resolving complex cases.

Aims: To resolve complex cases in blood group serology using T-NGS and helped in predicting the antibody specificity once the full antigen profile of the patient is known.

Methods: Eighteen cases where the specificity of the antibody could not be identified were analysed using T-NGS assay for 51 genes associated with 41 blood group system antigens. Predicted antibodies were further confirmed by using glycerol preserved ABO compatible high frequency antigen negative rare red cells. Family studies were also carried out for identifying more rare donors.

PA08-L05 Table 1: Novel and rare variants identified in red cell alloimmunized patients using T-NGS

Case no.	Gene/ SNP change	Protein Change (#:Novel)	Phenotype/Alloantibody
1	KEL/ c.523_524ins, 11bpc.517_518del, c.T516C, c.A512C	#p.E175Gfs*18, p.I173*, p.V172V, p.Q171P	K null with anti-Ku
2	A4GALT / c.72dupC	#p.I25Hfs*30	P null with anti-PP1P ^K
3	A4GALT / c.218delG	#p.G73Afs*41	P null with anti-PP1P ^K
4	A4GALT/ c.592delC	#p.L198Sfs*	P null with anti-PP1P ^K
5	A4GALT/ c.547_548del	#p.M183Vfs*99	P null with anti-PP1P ^K
6	A4GALT/ c.C392G	#p.S131X	P null with anti-PP1P ^K
7	A4GALT/ 26bp del c.972_997	p.R325Afs*113	P null with anti-PP1P ^K
8	ACKR1/ c.-67T>C & c.125G>A	p.O & p.Gly42Asp	
9-12	GYPB/ Exon 4 deletion	GPB absent	S-s-U- with anti-U
13	ACHE/ c.1057C>A	p.His353Asn	Yt(a-b+) with anti Yta
14-15	C4B/ Exon 28 deletion	#C4B absent	Ch- with anti-Ch
16	CD44/ c.137G>C	p.Arg46Pro	In(a+b-) with anti-Inb
17-18	GYPC/ c.60_116del	p.Ala23_Met41del	Ge: -2,3,4 with anti-Ge2

Results: Novel and rare mutations identified by T-NGS which resulted in null phenotypes is summarised in Table. Overall ten novel mutations (resulting in seven novel alleles) were identified: Knull (1), Pnull (5) and Ch- (1). Most of these mutations were of frameshift type. In five cases, known rare mutations were identified: deletion of 26-bp c.972_997 in A4GALT (Pnull), c.-67T>C & c.125G>A in ACKR1 (Fynull), GYPB Exon 4 deletion (S-s-U-), c.1057C>A in ACHE (Yta-), c.137G>C in CD44 (Inb-) and c.60_116del in GYPC (Ge:-2,3,4). Family studies of proband identified five more rare blood donors namely: Pnull (3), Fynull (1), S-s-U- (1). Specificity of all the antibodies further confirmed using glycerol preserved rare red cells wherever available. All the novel mutations were validated using Sanger sequencing and in-silico analysis predicted the novel variants to be damaging.

Summary / Conclusions: This is the first study to report seven novel allele variants which are responsible for causing null phenotypes in Indian patients using NGS-based approach. High-through put genotyping is effective tools to resolve transfusion-related serologic problems and help in identifying rare donors. Rare donors identified will be registered in the Rare Donor Registry of India (RDRI) for provision of rare units nationally and internationally.

PA08-L06 | Identification of a frequent replacement of part of downstream Rhesus box with large fragment of part of TMEM50A inversion and RHCE by long-read whole-genome sequencing

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Background: Compared with the common *RHD* variants like missense mutations, in frame insertion or deletion, and *RHD-CE-D* hybrid alleles, large segment structural abnormality of *RHD* gene is rarely described.

Aims: In this study, long-read sequencing including whole genome sequencing was applied to clarify a large fragment structural abnormality of *RHD* gene distributed in the Chinese population.

Methods: Nine samples with reduced copy number of *RHD* exon 10, which were identified by multiplex ligation-dependent probe amplification in previous study, were selected for long-range sequencing of *RHD* gene combined with long-read whole genome sequencing. PCR with specific primers was developed for further screening of the variant allele in the Chinese Han donors.

Results: A novel *RHD* complex structural variant allele with short fragment deletion of downstream *Rhesus box* (688bp) coupled with a 21.8 kb large segment insertion involving part of *TMEM50A* inversion (from intron 2 to exon 7, 21 648bp) and *RHCE* gene (non-coding region of exon 10, 174bp), as well as five point mutations, was identified in two samples with reduced copy number of *RHD* exon 10 by long-read whole genome sequencing. The 5'- and 3'- breakpoints of this complex structural variant allele were validated by Sanger sequencing. Repetitive *AluSx* element sequences were detected at the breakpoint by bioinformatics analysis. Approximate 4.1% (40/982) of Chinese D+ donors carried this complex structural variant allele.

Summary / Conclusions: A common *RHD* variant allele with a complex structural variation was identified, demonstrating that long-read whole genome sequencing is a powerful technique to clarify the large segments of complex genetic variation. In addition, this common structural variant allele should be taken into account for developing *RHD* genotyping assay to avoid allele dropout.

Parallel session—educational session

Managing the message

PA09-L01 | Working with the media—how to effectively communicate with the public

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This interactive presentation will provide a background to the strategy of working with the media and provide practical tips for making your science accessible and relevant to your audience, while avoiding potential pitfalls. Scientists can play a significant role in advancing the cause of their organisation through engaging constructively with media, and conversely, media exposure can help raise the profile of your research, secure funding and spark new collaborations. We'll discuss the "why" of communicating your science with the media, and how to choose the right story and news outlet to achieve your goals. For example—is your goal to support a call out for blood from your organisation? or to inform the public that your research has reached a key point that's useful or interesting for members of the public? Or part of a broader piece on an aspect of life, lifestyle, science or technology? The audience is a key part of any communication, and media is no exception. To get your message across, it's important to understand the knowledge, attitudes and beliefs of the audience. They may be quite different to your own, or those of your fellow scientists or workplace colleagues. A story in the media is very different from a scientific presentation. People don't have to read or watch it, so you need to grab their attention. You may be surprised at how long (or how little) the average reader spends on a news article, or the length of time you have to get your message across in a TV or radio interview. We'll talk about how this environment means that as scientists, we need to "upend" our thinking about communicating our work and adapt those communication styles that have been taught to us over years of training. Your organisation's media team (if you have one) are your key partners in dealing with the media, and finding new opportunities. They're as well trained in media as you are in science, so it's useful to get to know them, and work with them as "critical friends". We'll demonstrate some of these aspects with case studies and provide examples of how media and science have worked together at Australian Red Cross Lifeblood. We've been able to capture the attention of mainstream journalists, provide growth for our social media platforms, increase the reach of our research communications, conduct strategically aligned research and attract new collaborators for our researchers.

Parallel session—clinical

Dealing with the risk of incompatible transfusions

PA10-L01 | Risk of ABO incompatible platelets and plasma

L Estcourt

Abstract not available

PA10-L02 | Two severe cases of haemolytic disease of the fetus and newborn—is it possible to do more on prevention?

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Background: The global failure to provide enough immunoprophylaxis and to prevent Rh disease, contributes to an enormous burden of hemolytic disease of the fetus and newborn (HDFN) and provides an important challenge to the international health care system, even 50 years since it has been at disposal.

Aims: To present two cases of severe HDFN caused by Antibodies (Abs) from Rh system.

Methods: Blood groups were performed by Diaclon ABO/Rh gel card for newborns DVI+ (Bio-Rad Laboratories Inc. Cressier FR, Switzerland). Abs screening and identification were performed by column agglutination method using Coombs Anti-IgG gel cards with ID-DiaCell I-II-III and ID-DiaPanel test cells (Bio-Rad Laboratories, Inc. Cressier FR, Switzerland). RBC phenotyping was done by standard tube agglutination method with commercial test reagents (CE-Immundiagnostic GmbH Neckargemuend Germany). Direct Antiglobulin Test (DAT) was performed by LISS/Coombs gel cards (Bio-Rad Laboratories, Inc. Cressier FR, Switzerland).

Results: The first case was a premature baby, born at 32 weeks of gestation (weight 1380 g), in severe condition with Apgar score 7/8 and clinical signs of anemia, hyperbilirubinemia and hemolysis. Per anamnesis, the mother's blood group was A RhD negative, she was in her fourth pregnancy, no immunoprophylaxis received. In the 28th gestation week of pregnancy titer anti-D Abs>1000 and two plasmapheresis were proceeded. In our findings, the newborn was A RhD positive phenotype ccEe, DAT 3+, Indirect Antiglobulin Test (IAT) 3+ with anti-D and anti-E Abs. Due to bilirubin level>450 mmol/L, two units of blood for exchange transfusion were prepared in first two days, urgently. In third day, he received one top up dose of erythrocytes and thrombocytes. On the fifth day, complications occurred, worsening of the general condition, bleeding and death. The other

case depicts a premature baby that was born in the 33rd gestation week with severe clinical presentation of HDFN due to anti-D. The newborn body weight was 2690 g, with Apgar score 4/5. The baby was intubated, with clinical signs of anemia and hyperbilirubinemia. Per anamnesis, mother's blood group was O RhD negative, she was in her second pregnancy, no immunoprophylaxis received. Two intra-uterine transfusions proceeded during pregnancy. In our findings, the newborn was O RhD negative blood group, DAT negative and IAT 3+. Due to severe condition the newborn died in 2nd day.

Summary / Conclusions: Despite regular controls during pregnancy and the applied therapy, clinical course of two HDFN babies caused by anti-D Abs, was severe and resulted with fatal outcome. Despite the existing recommendation on the use of immunoprophylaxis there are still cases where it is lacking. Therefore, it is necessary to provide a stronger prevention, that included mandatory protocols for screening all pregnant women, and the use of immunoprophylaxis after all immunological events.

PA10-L03 | Pregnancy, perinatology and transfusion—analysis of the transfusion serious adverse events reported to the French national hemovigilance database from 2013 to 2022

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Background: In France, mandatory blood typing (ABO, Rh, Kell) and anti-red blood cell (RBC) antibody (Ab) screening are implemented during pregnancy. In the event of the detection of RBC Ab, reinforced monitoring is performed to anticipate any transfusions that may be necessary. This monitoring aims at detecting and managing a potential RBC maternal-foetal incompatibility (MFI). Despite these preventive measures, serious adverse events (SAEs) potentially impacting MFI management occur.

Aims: To identify and understand failures which impacted the management of the safety of the foetus or neonate by analyzing SAEs reported to the French national hemovigilance database.

Methods: All reported SAEs (with or without adverse reaction) pertaining to neonatology and obstetrics between 01/01/2013 and 31/12/2022 were analyzed with regard to frequencies and causes and compared to SAEs in other clinical sectors (Chi² and Fisher exact tests were used). Clinical impacts in the neonates were assessed in the perinatal period.

Results: Over a 10 years period, 759 SAEs were collated and analyzed, including 171 in neonatology and 588 in obstetrics corresponding to 2.1% and 7.4% of all reported SAEs ($n = 7967$) respectively. SAEs are likely underreported. SAEs in the neonatology and obstetric sectors were overall similar to other clinical sectors with however some specificities: blood typing and antibody screening process were more frequently implicated in obstetrics with 24.5% (144/588) of

SAEs and in neonatology with 20% (34/171) versus 13% for other sectors (971/7208) ($p < 0.01$). In 6% of cases in obstetrics (35/588) the failure was related to a typing/screening performed in an off-site laboratory (vs 1.4% $n = 104/7208$ in other sectors) ($p < 0.01$). Blood component issuing was implicated in 14% of SAEs (24/171) in neonatology and 7% (43/588) in obstetrics (vs 13% $n = 925/7208$ in other sectors) ($p < 0.01$ vs obstetrics). Transfusion delays were reported in 4.7% of SAEs in neonatology (8/171) (1.8% $n = 127/7208$ in other sectors) ($p = 0.03$). Patient misidentification occurred in 33% of SAEs in neonatology (56/171) and 41% in obstetrics (243/588) (vs 24% (1748/7208) in other sectors) ($p < 0.01$). In neonatology, 8.7% (15/171) of SAEs resulted in potential clinical impacts: 4 non-compliance with neonates transfusion guidelines, 1 recipient error (mother/neonate), 1 refractoriness of transfusion, and 9 delayed MFI managements during pregnancy (5 RH4; 2 RH1; 1 RH3; 1 unclear). In these 9 cases, failure to detect and/or monitor the positive Ab screening during pregnancy was due to a miscommunication between laboratories and/or obstetric teams and/or blood issuing structures regarding blood typing and Ab screening results. In at least 6 of the 9 cases, Ab tests were performed by off-site laboratories. In 3 cases with anti- RH4 MFI, neonates were transfused in emergency settings with an incompatible RBC RH4 due to unawareness of positive Ab tests. This resulted in a serious adverse reaction grade 3 in 2013, one death in 2016, one case with an unknown clinical outcome in 2020.

Summary / Conclusions: Our study highlights the critical risk posed by the lack of structural link between off-site laboratories, blood component issuing structures and obstetricians/midwives teams, leading to failure in managing RBC MFI and possibly incompatible RBC neonate transfusion. Prospective studies are needed to further inform the current miscommunication issues and provide pathways for improved transfusion processes regarding neonates and their mothers in France.

PA10-L04 | Ethical and equity concerns regarding the use of RhD positive LTOWB in pediatric and adult females of childbearing potential

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Background: Recent data suggests a lifesaving benefit from transfusing low titer group O whole blood (LTOWB) in trauma.

Aims: The objective of this study was to combine results of two surveys describing current practices regarding the use of RhD positive LTOWB for life-threatening bleeding in children and adults. Monitoring/counseling policies for RhD negative females of childbearing potential (FCP) after transfusion with RhD positive LTOWB to reduce D alloimmunization and potential future hemolytic disease of the fetus and newborn (HDFN) were also studied.

Methods: Survey data was collected from blood bank directors at adult and pediatric trauma centers in the US regarding the use of RhD positive products for life-threatening bleeding as well as current D alloimmunization risk mitigation and counseling strategies for FCPs. The second survey was sent to 25 pediatric level 1 trauma centers. This survey asked specific questions regarding the RhD status of LTOWB use in children that were not asked in the first survey.

Results: The first survey yielded a 60% response rate (95/157) from adult and pediatric level 1 trauma centers in the US. At the 60% of sites using LTOWB (57/95), it was not used for adult FCPs at 12%, pediatric FCP at 21%, and boys at 17%. Centers reported RhD-positive LTOWB use exclusively or preferentially more commonly in men (94%) and boys (54%) than adult FCP (40%) or pediatric FCP (21%). Of the responding institutions, only 46% (44/95) require the clinical team and/or transfusion service to counsel the patient/family of an RhD negative pediatric or adult FCP after exposure to RhD positive LTOWB or RBC unit about the risk of D-alloimmunization and future HDFN. The second survey of 25 pediatric level 1 trauma centers in the US yielded a 100% response rate (25/25). Of the 25 pediatric trauma centers, 36% (9/25) are currently using LTOWB for life threatening hemorrhage, and 16% (4/25) exclusively use RhD positive LTOWB regardless of patient sex or RhD type. Of the 9 sites currently using LTOWB, 2 (22%) are using a non-leukoreduced product. There were 64% (16/25) of sites that by policy can use RhD positive LTOWB in a girl before her RhD type is known or if she is known to be RhD negative and the RhD negative LTOWB supply was exhausted (or would be willing to do so once LTOWB was adopted at their site). In total, 36% (9/25) responded they would not use RhD positive LTOWB in pediatric FCP and would use component therapy with RhD negative RBCs.

Summary / Conclusions: The lack of RhD negative LTOWB is leading to RhD positive LTOWB use in pediatric and adult FCP. Due to the scarcity of RhD negative LTOWB, the use of RhD positive LTOWB will likely continue to increase. The ethical and equity issues around many centers providing LTOWB only to men and not to girls or adult FCPs will be informed by clinical trials that are ongoing that will assess the efficacy and safety LTOWB. Advancements in the detection and treatment of HDFN may also lead to improved outcomes for alloimmunized pregnancies. Since only half of the centers surveyed have policies regarding counseling for FCPs who may become D-alloimmunized through transfusion with RhD positive blood products, it is imperative that centers using RhD positive LTOWB in RhD negative FCPs adopt post transfusion monitoring, follow up and counseling policies to help further reduce the future risks of HDFN.

PA10-L05 | Novel approach to treat autoimmune hemolytic anemia secondary to immune checkpoint inhibitors in a mouse model

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Background: Immune checkpoint inhibitors (ICPi) have transformed the landscape of cancer treatment, but can lead to severe hematological adverse events. Autoimmune hemolytic anemia (AIHA), due to pathogenic autoantibodies that destroy red blood cells (RBCs), is the most frequently-reported hematological complication of ICPi therapy. AIHA secondary to ICPi therapy is associated with severe hemolysis and high fatal rates, and current AIHA therapies have variable success, with high relapse rates; as such, there is an unmet need to develop more efficacious treatments.

In a preclinical mouse model of AIHA secondary to ICPi that reflects signs, symptoms, and clinical course observed in human patients, a distinct CD39⁺ CD4⁺ T cell population (referred to herein as “CD39SP”) emerged in the peripheral circulation and reliably predict AIHA development. As CD39 is known to contribute to immunosuppression, by converting proinflammatory extracellular ATP into ADP and AMP, we hypothesized that modulating purinergic signaling could mitigate AIHA.

Aims: To evaluate apyrase, an enzyme that degrades ATP, as a therapeutic in a preclinical model of AIHA secondary to ICPi.

Methods: HOD mice (expressing an RBC-specific HOD transgene) were bred with OTII mice (with CD4 T cells recognizing HOD) to generate autoreactive HOD+OTII+ mice. HOD+OTII+ mice were treated with 4-antibody ICPi cocktail against IL-10R (500 µg i.p. weekly) and CTLA-4, LAG3, and PD1 (200 µg i.p. each, every other day) for 14 days to induce AIHA. Apyrase therapy (5 units infused daily for 10 days) was evaluated at 2 timepoints after the initiation of ICPi: (i) starting after 14 days of ICPi (when robust autoantibody production was seen), or (ii) starting after 7 days of ICPi exposure (when CD39SP are detectable, but autoantibodies are not). RBC autoantibody levels were measured by flow cytometry with quantitation reported as mean fluorescence intensity (MFI); hematocrit (reported as percent) and survival rates were also evaluated.

Results: Compared to ICPi-treated control HOD+OTII+ mice, therapeutic apyrase treatment commencing at 14 days after exposure to ICPi did not effect RBC autoantibody levels or hematocrit, but did significantly improve survival rates (53.8% of ICPi-treated control vs. 89.6% of ICPi+apyrase-treated mice; $p < 0.05$). Leveraging CD39SP as a predictive biomarker of AIHA onset, apyrase was infused at the initial time of detecting circulating CD39SP. Compared to ICPi-treated control HOD+OTII+ mice, apyrase led to significant reductions in RBC autoantibody levels (MFI 1934 with 95% confidence interval of [1653, 2316] vs. 1247 [867, 1723], $p < 0.01$), anemia (evidenced by increased hematocrit (14.4 ± 1.7 vs 21.2 ± 1.8 , $p < 0.05$)), and mortality (i.e., 100% survival). These results reflect 4 independent experiments with 8 mice/group.

Summary / Conclusions: Apyrase treatment significantly improves clinical signs of AIHA secondary to ICPi. And, leveraging CD39SP as an easily-measurable predictive biomarker, apyrase administration at the time of CD39SP detection significantly attenuated AIHA progression, leading to 100% survival. Together, these results highlight the

translational potential of targeting purinergic signaling pathways to treat, or potentially prevent, human AIHA.

Parallel session—blood products

RBC products for intrauterine and neonatal transfusion: from the bench to the bedside

PA11-L01 | International approach to preparation and processing of red blood cell components for neonatal transfusion

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Red blood cell transfusions are commonly administered to neonates in the neonatal intensive care unit, yet they pose unique challenges as neonates are vulnerable recipients with different physiological characteristics compared to children or adults. These differences include a smaller blood volume and immature haemostatic system, which can put them at higher risk of transfusion associated adverse events. To address these challenges, Blood Establishments frequently take additional safety measures when processing blood components for neonatal patients compared to adult patients. However, there is a lack of evidence-based guidance on the optimal preparation and processing of blood components specific to neonates. Additionally, recommendations for storage age and optimal usage of blood components for neonates may differ from those for children and adults. This lack of standardization leads to variability in clinical practices, as previously highlighted in a nationwide survey of Blood Establishments in the United States (Reeves, Transfusion, 2021). Current variability of practice in Europe has only been partially described in a recent international forum involving four European countries (Arora, Vox Sanguinis, 2023). Therefore, the Neonatal Transfusion Network (<https://neonataltransfusionnetwork.com>) recently performed a large international survey ('Survey on blood COmponent specifications for Neonates in Europe (SCONE)') targeted at transfusion experts in European Blood Establishments, with responses received from 19 countries. The survey covered aspects of preparation and processing of neonatal red blood cell components including donor selection criteria, leukocyte reduction, use of CMV antibody-screened components, anticoagulant and storage medium used, storage age, irradiation, washing and the use of small-volume aliquots. The survey has provided insights into the current approaches and existing variation in neonatal component

specifications in European Blood Establishments and a context in which to consider which strategies may be most appropriate for the future.

PA11-L02 | Assessment of Red Blood Cell (RBC) hemodynamic functionality by membrane proteins—implications to blood transfusion

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Background: The primary role of red blood cells (RBC) is to supply oxygen to tissues. To accomplish this, RBC have unique flow-affecting properties, which define hemodynamic functionality, namely, their capacity to affect blood circulation. A major effector of the RBC hemodynamic functionality is the cell's **deformability**, expressing the cells ability to adapt their shape to the dynamically changing flow conditions, minimizing their resistance to flow. This is particularly important for their passage through the capillaries, which are narrower than the RBC. In previous studies, we have shown that the deformability of transfused RBC is a potent effector of the transfusion outcome, expressed by the transfusion-induced hemoglobin increment and skin

PA11-L02 Table 1: Correlations between the levels of RBC membrane proteins and the cell deformability.

No	Proteins	Significance, p	Pearson coefficient, r
1	Ezrin	0.00006	0.83
2	Long-chain-fatty-acid—CoA ligase 4	0.0002	0.81
3	Argonaute-2	0.0003	0.79
4	Protein band 4.1	0.0003	0.79
5	Glycophorin C	0.0005	0.77
6	GTP-binding proteins Ras	0.0007	0.76
7	Stomatin	0.0008	0.75
8	G-adducin	0.001	0.75
9	Flotellin-1	0.001	0.73
10	CD44	0.002	0.72
11	Band-3	0.002	0.72
12	Flotellin-2	0.002	0.71
13	Integrin-associated protein CD47	0.003	0.72
14	Glycophorins A	0.004	0.67

blood perfusion. This suggests that determining transfused RBC deformability is essential for assessing blood transfusion outcomes. RBC deformability is determined exclusively by physical methods using various techniques. However, the cell's deformability is determined primarily by its structure and composition, and its alteration should correlate with changes in biochemical measures.

Aims: The present study explored the correlation between RBC membrane protein composition and cell deformability.

Methods: RBC membrane protein composition was comprehensively analyzed by mass spectroscopy, and the proteins' levels were examined for correlation with the cell deformability, as determined by image analysis, expressed by the flow-induced cell elongation. Upon their consent, sixteen RBC samples were obtained from two sources: (1). Nine freshly collected blood samples from healthy volunteer donors. (2). Six samples from expired packed RBC (PRBC) units (stored for 42 days). All samples were taken under the approval of the Hadassah Hospital Ethical Committee, Jerusalem, Israel.

Results: 752 membrane proteins were identified. However, the deformability was positively correlated with the level of only fourteen proteins involved in membrane rafting and/or the membrane-cytoskeleton linkage (Table 1), as well as negatively correlated with the content of membrane-bound hemoglobin isoforms. Of particular interest, a highly significant **inter-correlation** was found between the levels of these deformability-expressing proteins.

Summary / Conclusions: The study's findings suggest that the reduction of deformability is a programmed (not arbitrary) process of remodeling and shedding of membrane fragments, possibly mirroring the formation of extracellular vesicles. The highly significant inter-correlation between the deformability-expressing proteins infers that the cell deformability can be assessed by determining a few, or possibly one, of these proteins. As the deformability of transfused RBC is a potent effector of blood transfusion outcome, these findings provide a facile method for assessing the hemodynamic functionality of transfused RBC before transfusion for improving blood transfusion therapy.

PA11-L03 | Effect of proteasome activity on red blood cell phenotypes linked to storage lesion

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Background: Red blood cells (RBCs), due to their inability to synthesize new proteins, rely on protein homeostasis mechanisms to maintain their proteome's integrity and functionality. The proteasome machinery is responsible for degrading defective proteins and has

been found to possess a central role in biological networks of stored RBCs. The proteasome's role during storage has been scarcely examined. Yet, the observation that stored RBCs from donors with increased proteasomal activity present fewer oxidative defects, suggests a protective role.

Aims: This study aims to further unravel the effects of proteasome activity on RBC phenotypes linked to storage lesion.

Methods: Twenty-four leukoreduced units of packed RBCs in CPD-SAGM were examined during storage for hemolysis (e.g., osmotic, oxidative), redox (e.g., reactive oxygen species; ROS), senescence, and proteasomal parameters. Proteasome activities were correlated with storage lesion phenotypes using Pearson's and Spearman's tests. The units were also categorized into subgroups of high and low proteasomal activity to assess their differences. Lastly, a pilot *in vitro* examination was conducted, by incubating RBCs ($n = 5$) with proteasome inhibitors and analyzing parameters of their physiology and biochemistry. Significance was accepted at $p < 0.05$. The study has been financed by the funding programme "MEDICUS" of the University of Patras.

Results: All three cytosolic proteasomal (cProteasome) activities presented inverse correlations with ROS accumulation upon exposure to oxidative stress (e.g., late-storage: caspase-like activity—phenylhydrazine-induced ROS, $R = -0.601$, $p < 0.01$). Regarding the membrane, all three activities (mProteasome) were inversely linked to fragility indices, especially during middle-storage (e.g., chymotrypsin-like activity—oxidative fragility, $R = -0.666$, $p < 0.01$). A *posteriori* stratification of the units based on their cProteasome or mProteasome activity revealed that the high activity subgroup was characterized by a lower propensity to spontaneous lysis (e.g., end-of-storage hemolysis: 94.35 ± 40.7 vs. $62.11.19 \pm 28.73$ mg/dL, low vs. high cProteasome activity, $p < 0.05$), as well as to stimulated lysis, especially in the middle of the storage period. The cProteasome high group was also characterized by decreased ROS (with and without stimulation) in late storage (e.g., intrinsic ROS: 1012 ± 587 vs. 608 ± 344 MFI, low vs. high, $p < 0.05$). These descriptive, yet promising, results led to targeted experiments of proteasome inhibition to examine its implication in maintaining integrity and redox equilibrium in RBCs. The use of Ada-Tyr-(Ahx)3-(Leu)3-vinyl sulfone inhibited all active subunits (35%-55% activity decrease, $p < 0.01$). The modified samples were prone to lysis with (e.g., approximately 25% increase of oxidative hemolysis, $p < 0.05$) or without (3-3.5 fold increase, $p < 0.01$) external stress and to ROS accumulation (approximately 25% increase, $p < 0.05$).

Summary / Conclusions: The proteasome machinery seems to be a crucial contributor to RBC integrity, shielding RBCs from lysis and excessive ROS generation. Our *ex vivo* results highlight the proteasome's potential to stratify blood units regarding their hemolytic propensity and oxidation profile while the pilot *in vitro* proteasome inhibition further supports its protective role. Considering proteasome's central role in biological networks of stored RBCs, the presented data hint toward its fine-tuning to counteract storage lesion.

PA11-L04 | Biological and genetic determinants of glycolysis—phosphofructokinase isoforms boost energy status of stored red blood cells and transfusion outcomes

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Background: Glycolysis is a central metabolic pathway in health and disease, an essential one in mature red blood cells (RBCs), which rely on the Embden-Meyerhof-Parnas pathway as the sole source of energy generation. During aging in vivo and in vitro under blood bank conditions, the adenosine triphosphate (ATP) generated via glycolysis in mitochondria-devoid mature RBCs is essential for the modulation of oxygen kinetics, deformability and, ultimately, intra- and extra-vascular hemolysis.

Aims: To determine the impact of donor biology, including age, sex and genetics in the regulation of stored RBC glycolysis.

Methods: Here we drew upon multi-omics data from end of storage blood units obtained from a cohort of 13,029 volunteers enrolled in the Recipient Epidemiology and Donor Evaluation Study (REDS). These data were then correlated to functional outcomes, like in bag hemolysis and recipient clinical data (vein-to-vein database). First, we sought to identify the biological and genetic traits associated with heterogeneity in glycolytic phenotypes. For the latter, we leveraged a panel of ~879,000 single nucleotide polymorphisms (SNPs) to determine metabolite Quantitative Trait Loci (mQTL) associated to glycolytic heterogeneity. Gene-metabolite associations were validated by testing glycolysis in fresh and stored RBCs from 525 Jackson lab Diversity Outbred (J:DO) mice, derived from extensive cross-breeding of eight genetically diverse founder strains. ⁵¹Cr post-transfusion recovery studies were performed in an independent cohort of 79 donors enrolled in the Donor Iron Deficiency Study (DIDS).

Results: Lower levels of glycolytic metabolites were detected in units from older donors. Sex dimorphism was observed, with higher levels of glucose and lower levels of glycolytic metabolites in females up to age 50, the average age of menopause in the United States. Associations were also observed to ancestry-specific genetic polymorphisms in regions coding for phosphofructokinase 1, platelet (PFKP—especially the rs2388595 SNP—FDR-corrected *p*-value = *e*-163)—hexokinase 1, and for the ADP-ribosyl cyclase 1 and 2 (CD38/BST1—rs4478185). Gene-metabolite associations in the J:DO population confirmed an association between glycolytic metabolites and

polymorphisms in the regions coding for PFKP and CD38/BST1. Through a combination of proteomics and systems biology-based metabolic reconstruction models of RBC metabolism, we showed that PFKP is detected in pure mature RBCs and promotes glycolytic flux when ATP consumption outpaces synthesis rates, such as in stored RBCs. As a result of storage-induced oxidant stress, ATP is consumed into lower energy pools and deaminated into hypoxanthine. Here we show that ATP and hypoxanthine are negatively and positively associated with hemolysis in vitro and in vivo, respectively. Specifically, through PTR studies in the DIDS cohort we describe an association between in vivo survival and the genetic factors/levels of these metabolites in healthy autologous recipients. By interrogating a vein-to-vein database, we also show that end of storage ATP and hypoxanthine levels are associated with in vivo hemoglobin and bilirubin increments in 4700 heterologous clinical patients receiving transfusions.

Summary / Conclusions: Genetic and non-genetic factors regulate glycolysis of fresh and stored human and murine RBCs, impacting both storage hemolysis in the unit and extravascular hemolysis upon transfusion in healthy autologous recipients and clinical patients receiving transfusion.

PA11-L05 | FlowScore, a flow-cytometric surrogate of the oxygen-unloading rate from red blood cells and a marker for assessing storage lesion in banked blood

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Background: Under storage, red blood cells (RBCs) undergo metabolic and morphological changes. To investigate how this storage lesion impacts O₂ handling, we applied single-cell oxygen saturation imaging, a method we developed to measure the rate of O₂ unloading from RBCs (Richardson et al, PNAS, 2020). We found that storage progressively slows O₂ unloading (longer time constant, τ) in a donor unit-dependent manner (Donovan et al, Haematologica, 2022). Using perfused human kidneys, we demonstrated how storage-related kinetic attrition reduces cortical oxygenation, indicating a clinically significant effect of τ (Dumbill et al, Blood, 2023). Thus, we propose that O₂-delivery depends not only on O₂-carrying capacity (monitored by

haemoglobin measurements), but also on O₂-handling kinetics that are labile in storage, yet not routinely measured. Although a potentially important quality metric of the performance of blood production and storage, single-cell oxygen saturation imaging is a resource-intensive method and not readily transferable to blood banking facilities. This limitation warrants a search for convenient surrogates of O₂ handling kinetics. Our analyses previously indicated that τ depends critically on cellular geometry, hence flow cytometric blood analysers were investigated.

Aims: To test metrics of RBC geometry widely available on haematology analysers as surrogates for τ , and to investigate their reproducibility when applied to stored blood measurements across a number of sites.

Methods: Measurements of τ were paired with counts performed on a Sysmex XN-series analyser at NHS Blood and Transplant in England. Recordings were performed on freshly drawn (reference) donor bloods ($N = 95$) and from samples of RBC components over storage (between days 2 and 49; $N = 165$). Multivariate regression analysis was used to identify candidate parameters from the analyser, which were then tested for consistency and stability using commercial QC standards; the same lot number was internationally distributed for the study to three additional sites: Lifeblood (Australia), Canadian Blood Service, and Banc de Sang i Teixits (Spain).

Results: The strongest correlation between τ was observed with RET-RBC-Z, the channel representing side-scatter. The relationship was strengthened further when its related parameter, RET-RBC-Y, representing forward-scatter, was included. By best-fit, we described an algorithm of these two parameters, which we call FlowScore. Using the 95th percentile of reference data as a cut-off, FlowScore showed good (>80%) sensitivity, specificity, and accuracy as a marker of storage lesion. Since RBC geometry and metabolic state are inter-related and jointly affect oxygen release, a variant of FlowScore described intracellular [ATP] and [2,3-diphosphoglycerate], biochemical markers of storage lesion. FlowScores of the QC calibration standards were comparable between the four blood services, with errors no greater than 5% compared to their global average.

Summary / Conclusions: Our study establishes FlowScore as a cost-effective and readily available surrogate of RBC oxygen-unloading kinetics: a parameter that is labile under storage and may change in disease. We propose FlowScore as a marker of RBC quality for blood-banking—an innovation that can be integrated seamlessly into existing workflows, potentially setting a new standard for RBC assessment in the field.

Parallel session—blood safety

Fireside chat - haemovigilance and undertransfusion

PA12-L01 | Haemovigilance fireside chat - under-transfusion

R Goel

Abstract not available

PA12-L02 | Tracking of under-transfusion in SHOT

S Narayan

Abstract not available

Parallel session—immunobiology

Anti-CD47 - taking the negative out of the positive

PA13-L01 | Transfusion management in the era of magrolimab (Hu5F9-G4)

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Magrolimab (Hu5F9-G4) is a novel anti-CD47 monoclonal IgG4 antibody under evaluation for the treatment of blood and solid-organ cancers. CD47 is a 'don't eat me signal' that is over-expressed in some cancers leading to evasion of removal of cancer cells by the immune system. Magrolimab (anti-CD47) blocks this signal, and allows phagocytosis of cancer cells by macrophages. CD47 is expressed on red blood cells (RBCs) and mediates their normal clearance as they age. Magrolimab binds to RBCs and interferes with pre-transfusion testing including blood group typing, and direct and indirect antiglobulin tests. Transient anaemia may occur after initial treatment with Magrolimab sometimes leading to an increase in transfusion requirements in patients already receiving transfusions or the need for transfusion in patients with pre-existing anaemia. This transient anaemia is likely to be due to splenic sequestration. There is little evidence of haemolysis due to Magrolimab. A phase I dose escalation study of Magrolimab in AML began in Oxford in 2015 which provided experience in these problems and how to overcome them. Since then, we have been working with others to provide guidance for other centres. We and others found that the interference with pre-transfusion testing was not observed with the Immucor Capture (Neo) system in contrast to other techniques. The Immucor Capture antiglobulin (*Gammaclone*) does not detect anti-IgG4. Key actions to mitigate the interference of pre-transfusion testing by Magrolimab begins with timely communication with the transfusion service before the patient starts treatment. Patients must have pre-transfusion testing in advance of the use of anti-CD47 to determine their ABO, D types and if RBC antibodies are present. RBC genotyping is recommended so that compatible blood can be provided by matching donor blood to the patient's genotype, if necessary. Magrolimab interference with pre-transfusion testing is not overcome by using dithiothreitol (DTT) or enzyme-treated RBCs as for patients taking anti-CD38 therapies. A monoclonal anti-human

IgG that does not recognise IgG4 should be used for antibody detection and crossmatching. A more laborious alternative is multiple alloadsorptions with RBCs or platelets to remove Magrolimab from the patient's plasma. Other anti-CD47 therapies are in development and some are not IgG4; their effect on pre-transfusion testing may be different to Magrolimab and will require consideration of effective mitigation strategies.

PA13-L02 | Evaluation of soluble recombinant CD47 on a French cohort of patients treated with anti-CD47 therapy

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Background: High expression of CD47 on the red blood cell (RBC) surface explains interferences with pre-transfusion tests in patients treated with anti-CD47 therapy. The Capture-R method (Immucor) uses an antiglobulin that does not recognize IgG4. Therefore, it can be used to overcome interferences from IgG4 anti-CD47 therapies such as Magrolimab (Hu5F9-G4) with, however, a non-negligible percentage of aspecific reactions. In order not to rely on a specific technique for IgG4-based anti-CD47 therapies, NHSBT has developed a soluble recombinant protein that contains the extra-cellular domain of CD47.

Aims: The purpose of this work is to evaluate the soluble recombinant CD47 (srCD47) manufactured by NHSBT for Indirect Antiglobulin Test (IAT) in French patients treated with anti-CD47 therapy (Magrolimab and Evorpacept) in various clinical trials, and to compare the results with the first line Capture-R technique.

Methods: 42 samples were tested, totaling 17 patients included in 5 different clinical trials. Sixteen patients were treated with Magrolimab (Hu5F9-G4) and one patient with Evorpacept (fusion protein with D1 SIRP α domain + IgG1). All samples were titrated on rr phenotype RBC (that contain the highest copy number of CD47 on its surface). The protocol is 60 μ L of patient plasma incubated with 30 μ L of srCD47 at room temperature for 15 min to sequester anti-CD47 therapy. An IAT is then performed with a dilution control in parallel. Plasmas with a known allo-antibody were spiked with plasma from patients treated with Magrolimab to ensure the absence of influence of srCD47 in the detection of allo-antibodies.

Results: 20 samples from Magrolimab treated patients with negative IAT using Capture-R technique were also negative when tested with srCD47. Out of 21 samples from patients on Magrolimab with aspecific IAT reactions using Capture-R technique, 11 were negative with srCD47. One sample became negative by doubling the amount of srCD47 (sample titre is 1/2²⁰, the highest in the dataset). The 9 samples that remained positive with srCD47 came from the same patient

with titres ranging from 1/2¹⁴ to 1/2¹⁹. Alloimmunization was excluded during antibody identification with srCD47. The Evorpacept sample showed a negative reaction with srCD47. Regarding plasma spiked with plasma from patients treated with Magrolimab, all allo-antibodies specificities (anti-D, anti-E, anti-Kell, anti-Fya) were detected after inhibition by srCD47.

Summary / Conclusions: srCD47 inhibition of plasmas from patients treated with anti-CD47 therapies (Magrolimab and Evorpacept) is a simple method to deploy in immunohematology laboratories. Inhibition works for samples with anti-CD47 titers up to 1/2²⁰ (by doubling the amount of srCD47). This method works regardless of the nature of the anti-CD47 therapy used (IgG4 or IgG1) and enables identification of underlying allo-antibodies.

PA13-L03 | CD47 regulates red blood cell sensitivity to antibody-induced clearance and antigen modulation following incompatible transfusion

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Background: Red blood cell (RBC) alloimmunization can make it difficult to find compatible RBCs and increases the risk of adverse events. However, not all incompatible RBC transfusions result in hemolytic transfusion reactions. Recent studies suggest that antibody engagement of RBCs can result in clearance and/or antigen modulation, a process whereby bound antibody selectively removes the target antigen. While antigen modulation can cause RBCs to become resistant to antibody-mediated clearance, the factors that dictate whether antibodies will induce RBC clearance, antigen modulation or both remain incompletely understood. As CD47 regulates RBC survival in general, we hypothesized that CD47 may also play a critical role in dictating the consequences of antibody engagement following transfusion.

Aims: To define the role of CD47 on RBC clearance and antigen modulation following an incompatible transfusion.

Methods: Mice expressing the HEL, OVA, and Duffy (HOD) antigen were crossed with CD47 knockout (KO) mice to generate donors that were HOD antigen positive but were CD47 wild type (WT), heterozygote (Het) or KO for CD47. HOD CD47 WT, Het, or KO RBCs were labeled with a fluorescent lipophilic dye, followed by transfusion into anti-HOD immunized or non-immunized recipients. HOD RBC survival, HOD antigen levels, bound antibody and complement were detected on RBC by flow cytometry. At least 10 recipients were analyzed per group with a one-way analysis of variance with a Tukey multiple comparison test used to compare groups.

Results: While no detectable differences in HOD antigen levels were observed on HOD CD47 WT, HOD CD47 Het and HOD CD47 KO RBCs, HOD CD47 WT RBCs possessed the same CD47 antigen levels as WT RBCs, while HOD CD47 Het RBCs expressed half the levels of those observed for HOD CD47 WT

RBCs ($p < 0.0001$). No detectable CD47 was present on HOD CD47 KO RBCs. Transfusion of HOD CD47 WT RBCs into anti-HOD immunized recipients failed to result in detectable RBC clearance, despite clear evidence of antibody engagement immediately post-transfusion. However, within 10 min of transfusion, bound antibody levels quickly declined, raising the possibility of antigen modulation. Consistent with previous results, HOD antigen levels also decreased and paralleled decreases in antibody levels. In contrast, while HOD CD47 Het RBCs failed to exhibit any difference in survival when compared to HOD CD47 WT RBCs following transfusion into non-immunized recipients, HOD CD47 Het RBCs did exhibit increases in clearance following antibody engagement ($p < 0.01$). While antibody and HOD antigen levels likewise decreased on HOD CD47 Het RBCs following transfusion into immunized recipients, the rate and magnitude of decrease was no different than that observed for HOD CD47 WT RBCs. In contrast, while HOD CD47 KO RBCs experienced accelerated clearance following transfusion into non-immunized recipients, the clearance rate was substantially accelerated following transfusion into immunized recipients ($p < 0.001$). The rate and magnitude of antigen loss following antibody engagement of HOD CD47 KO RBCs also exceeded that observed following transfusion of HOD CD47 WT or HOD CD47 Het RBCs when assessed in parallel ($p < 0.01$).

Summary / Conclusions: These results demonstrate that CD47 can regulate the rate and magnitude of HOD RBC clearance and antigen modulation following an incompatible transfusion. Future studies will be needed to define whether CD47 similarly regulates incompatible RBC transfusion outcomes clinically.

PA13-L04 | Therapeutic anti-CD47 monoclonal antibody induces significant phagocytosis in a monocyte monolayer assay—potential for extravascular hemolysis

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Background: Monoclonal antibody therapy for cancer treatment is increasing and some therapeutic monoclonal antibodies bind to red blood cells (RBCs) as well as the intended malignant cells. This off-target RBC binding, reported for anti-CD38 (daratumumab, isatuximab) and anti-CD47 (magrolimab/Hu5F9-G4), results in interference in laboratory pre-transfusion testing, and for magrolimab a transient anemia is seen requiring transfusion in ~35% of patients. The mechanism of anemia associated with this IgG4 anti-CD47 is poorly understood and occurs in the absence of laboratory markers of hemolysis

Aims: To examine the activity in the allogeneic monocyte monolayer assay (MMA) of plasma from 6 patients treated with monoclonal IgG4 anti-CD47 (magrolimab) to determine the potential to cause extravascular hemolysis and understand the variability in anemia.

Methods: Patient plasma samples showed direct agglutination (range 1+–4+) and 4+ reactivity in indirect antiglobulin testing (IAT) by standard hemagglutination tube method using total anti-IgG. MMA was performed using R2R2 RBCs opsonized with patient plasmas diluted in PBS to avoid direct agglutination. Dilutions varied from 1/500–1/5000, and flow cytometry (FACS) was used to semi-quantify the level of antibody on the RBCs by mean fluorescence intensity (MFI). CD47 expression on 55 random healthy donor RBCs was determined by FACS with commercial anti-CD47 conjugated to Alexa Fluor™ 647 (CD47-AF647; BDBioscience). Fcγ receptor blocking experiments used F(ab')₂ antibodies to FcγRI (CD64), FcγRII (CD32) and FcγRIII (CD16) (Ansell Immunology or Absolute Antibody).

Results: Five of six patients plasma samples showed phagocytic indices (PI) ranging from 20 to 60, with PI>5 considered a clinically significant antibody. All five showed MFI values >20,000. The sixth sample gave a PI<5; however, the antibody was weaker with an MFI of 2000 at a dilution of 1/100. There was a large range of CD47 expression levels on RBCs of random donors with MFI ranged from 35 to 534; mean 161 ± 12 . Blocking MMA revealed the primary Fcγ receptor involved in the phagocytosis was FcγRI. To a lesser extent, FcγRII also was involved in the phagocytosis and FcγRIII was not involved.

Summary / Conclusions: Plasma from patients in a clinical trial receiving anti-CD47 magrolimab showed significant phagocytosis in the MMA. The results support the observation that therapeutic administration of an IgG4 anti-CD47 has potential to be clinically significant and result in in-vivo extravascular hemolysis, providing an explanation for the anemia that is seen in a subset of patients. Whether an individual experiences anemia/hemolysis may be dependent on the expression level of CD47. We observed a range of CD47 expression levels on RBCs from healthy individuals, and it is known that CD47 expression levels on RBCs differ depending on Rh phenotype. Future studies correlating levels of anemia with CD47 expression and Rh may offer insight as to which patients may be at risk for anemia/hemolysis.

PA13-L05 | Evaluation of a soluble CD47 recombinant protein for a neutralization procedure in plasma of magrolimab treated patients prior immunohematological tests

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Background: CD47 is a transmembrane glycoprotein ubiquitously expressed on hematopoietic cells descendants, including red blood cells and on certain hematologic cancer and tumor cells. Interaction of CD47 with the inhibitory immunoreceptor SIRPα expressed on phagocytes prevents elimination of healthy or malignant cells ("don't eat

me" signal). The Anti-CD47 magrolimab (formerly Hu5F9-G4) from Gilead Sciences, Inc., is a human monoclonal IgG4 currently in clinical trials for the treatment of solid tumors. However, the antibody present in the plasma of treated patients is a source of analytical interference during pre-transfusion tests as it binds reagent red blood cells (RRBC) used in indirect antiglobulin tests (IAT) and ABO reverse grouping tests (Velliquette, Transfusion, 2019).

Aims: The aim of this study was to test the ability of a proof-of-concept soluble CD47 recombinant protein (sCD47) to neutralize Anti-CD47 in the patient's plasma/serum and, thus, remove its interference on immunohematological (IH) tests. This would provide laboratories with a standardized procedure that requires only a short incubation of the patient's samples with sCD47 prior routine IH tests.

Methods: Thirty-five leftover samples from ten magrolimab-treated patients, collected at different post-infusion times, were collected for this study. These plasmas were pre-treated with increasing quantities of sCD47 (between 2 and 12 µL per 25 µL of plasma), or PBS as control, for 15 min at 37°C and, then, tested in IAT. Additionally, as none of the patient samples contained allo-antibodies, three samples were spiked with an anti-Kell to verify the possibility of detecting a clinically relevant antibody in these sCD47 pre-treated plasma. Thirteen out of these thirty-five samples were also tested in reverse-typing. Pre-treatment in this case was done using between 4 µL and 12 µL of sCD47 per 50 µL of plasma for 15 min at room temperature. IAT and reverse typing were performed using gel technique (Diagnostic Grifols S.A.) in manual method.

Results: Magrolimab interference could be neutralized in all IAT by using a variable volume of sCD47 depending on the sample. For 48.5% of the samples, it was sufficient to use from 2 to 8 µL of sCD47 and for 51.5% 10 to 12 µL of sCD47. The spiked anti-Kell antibody was detected on all 3 samples tested in IAT after successful neutralization of magrolimab. For all 13 samples tested in reverse typing, sCD47 could neutralize magrolimab interference. For 76.9% of the samples, 4 µL of sCD47 was sufficient to achieve complete neutralization, 15.4% of the samples required 8 µL and 7.7% 12 µL. All plasma treated with PBS instead of sCD47 still caused panagglutination of the RRBC used for IAT or reverse typing.

Summary / Conclusions: This study has shown that sCD47 can efficiently neutralize magrolimab-related analytical interferences in both IAT and ABO reverse grouping tests. This suggests that it is possible to have a fast and standardized procedure for immunohematology laboratories to be able to bypass anti-CD47 induced panagglutination. urgently undergoing clinical trials (e.g., ALX148, TTI-621 and TTI-622).

Parallel session—cellular therapies

CAR-T - from collection to preparation and clinical use

PA14-L01 | Characteristics of apheresis products and CAR-T cell therapeutic outcomes

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The need for starting material for CAR-effector cells, thus far predominantly patient-derived T cells collected by unmobilized leukapheresis, has given transfusion medicine a spotlight position in the burgeoning field of immune effector cell therapy. Especially when CAR-T manufacturing fails, or CAR-T products conforming to specification fail to demonstrate the desired clinical effect, transfusionists are asked to explain what went wrong during apheresis to generate these outcomes. However, the ability of transfusion medicine to affect qualitative apheresis outcomes is highly limited, and even quantitative outcomes can only be achieved if targets are communicated by the CAR-T manufacturers, which often is not the case. Mathematically complex formulae such as the one proposed by O'Reilly et al. (Cytotherapy 2023) or more simple ones such as the one proposed by the EBMT CAR-T guidelines (Hayden, Ann Oncol, 2022 or Lehmann, Cytotherapy, 2023) support apheresis planning with respect to apheresis volumes needed to achieve certain target doses of mononuclear cells or T-cells. While in adults, typically required T-cell doses can normally be achieved with conventional apheresis technology in a reasonable time, the same targets can be challenging in smaller children (Jarisch, J Clin Apheresis, 2020). If more precise requests were made these could also be met. Thus certain minimal doses of CD4 and CD8 cells could be communicated, or even more granular requests such as a minimal doses of stem/memory, naive or central memory T-cells – the predominant phenotypes conducive to generation of “good” CAR-T cells – could be accommodated. With respect to qualitative outcomes, at this point in time, it is clear that age, disease and history of treatments for that disease have negative long-term effects on T-cell fitness for CAR-T manufacturing. Only the short-term effects on T-cell fitness due to transient toxicity of anti-cancer therapy can be at least partially accounted for. Minimal wash-out periods for many of the more common therapeutic agents have thus been recommended (Worel, Bone Marrow Transpl, 2023; Yakoub-Agha, Haematologica, 2020), but can be unacceptably long for certain drugs, such

as Bendamustine or some therapeutic antibodies. After allogeneic transplantation, donor-derived T-cells collected from the patient remain functionally deficient for extended periods of time. Thus T-cells collected from pediatric/adolescent ALL patients who relapsed and became CAR-T-eligible beyond six months after allo-HSCT have excellent outcomes (Bader, Blood Adv, 2023). Outcomes at earlier time points were disappointing, though, and some of that was attributed to poor T-cell quality, so that in these cases T-cells collected from the donor may even be preferable (del Bufalo, Blood, 2023). Analysis of T-cells for markers of exhaustion, senescence or anergy may in the future be able to discern who could be a good candidate for CAR-T manufacturing, but currently our understanding of how to recognize a “good” CAR-T product is insufficient to guide such decision making. In the absence of better parameters, the minimally and optimally required T-cell dose to be collected needs to be communicated by CAR-T manufacturers.

PA14-L02 | Affordable CAR-T manufacturing

S Lozano

Abstract not available

PA14-L03 | Predictive factors of transfusion needs after CAR-T cell therapy for B-cell malignancies—a monocentric experience

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Background: Chimeric antigen receptor T-cell (CAR-T) therapy is emerging as a highly effective treatment option for patients affected by B-cell malignancies, including large B-cell lymphoma (DLBCL), mantle cell lymphoma (MCL) and B cell precursors leukemia (B-ALL). While initial efforts were focused on the characterization of early complications, namely cytokine release syndrome (CRS) and immune effector cell-associated neurotoxicity syndrome (ICANS), immune effector cell-associated hemato-toxicity (ICAHT) has been recognized as the most frequent late adverse event in real-world studies. Cytopenic patients often necessitate transfusion support with packed red blood cells (RBC) or platelet concentrates to alleviate symptomatic anemia or avoid major bleeding.

Aims: This retrospective monocentric study characterizes the transfusion burden of patients receiving anti-CD19 CAR T-cell therapy outside clinical trials to identify predictive factors associated with transfusion needs.

PA14-L03 Table 1.

	All (n = 67)	Transfused (n = 41)	Not transfused (n = 26)	p
Disease				
DLBCL	53 (79.10)	32 (78.05)	21 (80.77)	0.75
MCL	10 (14.93)	7 (17.07)	3 (11.54)	
B-ALL	4 (5.97)	2 (4.88)	2 (7.69)	
Sex (M/F)	36/31 (53.7)	18/13 (43.9)	18/8 (69.2)	0.04
Age at infusion (Years)	58 (33.8-71.2)	56 (48-62)	59.5 (51-67)	0.56
ECOG PS at infusion	1 (0-3)	1 (1-2)	1 (0-1)	0.24
Prior lines > 3	14 (20.90)	12/29 (29.70)	2 (7.69)	0.03
Bridging therapy				
None	6 (9.09)	5 (12.2)	2 (7.69)	
A-SCT	9 (13.64)	9 (21.95)	0	0.04
Chemoimmunotherapy	20 (30.30)	12 (17.07)	8 (30.77)	
Local radiotherapy	18 (27.27)	7 (17.07)	11 (16.42)	
Others	13 (19.70)	8 (19.51)	5 (19.23)	
RBC units within 3 months before CAR T(n)	1.37 (0-4.2)	2.21 (1.17-3.26)	0 (0-0)	<0.001
Plts units within 3 months before CART(n)	1.11 (0-4.2)	1.78 (0.84-2.71)	0 (0-0)	0.007
CAR-HEMATOTOX high risk	41 (61.19)	32 (78.05)	11 (42.31)	0.002
CAR-T product				
Axi-cel	32 (47.66)	20 (48.78)	12 (46.15)	
Tisa-cel	21 (31.34%)	12 (29.27)	9 (34.62)	0.89
Brexu-cel	14 (20.90%)	9 (21.95)	5 (19.23)	
CRS grade > 2	39 (58.21)	25 (60.98)	14 (53.85)	0.56
ICANS grade> 2	9 (13.43)	7 (17.07)	2 (7.69)	0.27

Methods: Consecutive patients treated with anti-CD19 CAR-T cells between September 2019 and November 2023 at our department and presenting with at least a 3-month follow-up after infusion were included in the analysis. Data regarding patient baseline status at infusion, disease characteristics, hematological history (number and type of previous line of therapy, including bridge therapy), infused product, CAR-T cells associated early toxicity (CRS, ICANS), and transfusion support were collected. CAR-HEMATOTOX score (Rejesk, Blood, 2021) was calculated for all patients.

Results: 67 patients were included in the analysis. 61% received at least 1 transfusion post-infusion. Baseline characteristics for the entire cohort, and a comparison between those receiving at least one transfusion after CAR T-cell infusion versus no transfusion are summarized in TABLE I. Transfused patients were in prevalence female, received more prior lines of therapy and more frequently chemotherapy-based bridge therapies, faced a heavier pre-infusion transfusion burden and were characterized by a higher CAR-HEMATOTOX score. In the multivariate model, the number of RBC units transfused within 3 months before CAR T-cells therapy and a CAR-HEMATOTOX score >2 were significantly associated with post-infusion transfusion needs ($p = 0.03$ and $p = 0.04$ respectively).

Summary / Conclusions: Our results may help to optimize the management of CAR T-cell-treated patients and support strategies that reduce transfusion needs in this population.

PA14-L04 | Analysis of cytokine release syndrome related factors after chimeric antigen receptor T cell therapy

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Background: Anti-CD19 chimeric antigen receptor T (CAR-T) cell therapy is an innovative treatment in relapsed/refractory (r/r) CD19 positive B-cell malignant lymphoma. CAR-T cell therapy can cause several adverse events, including cytokine release syndrome (CRS), immune effector cell associated neurotoxicity syndrome, infection, cytopenia, and therapy induced coagulopathy. CRS is the most frequent and severe form of the disease, so it needs to be diagnosed and treated at an early stage. However, the mechanism of CRS onset is not fully understood, and although reports regarding CRS-related factors have been published, there are few reports regarding the in vivo dynamics.

Aims: To analyze the temporal changes of factors reported to be associated with the onset of CRS to aid in the early diagnosis and treatment of CRS.

Methods: A total of 24 patients who were treated at our hospital for cases of r/r diffuse large B-cell lymphoma with CAR-T cell therapy using Tisagenlecleucel (Tisa-Cel), were enrolled in the study between January 2021 and October 2023. Product lot data (total viable cell number, total number of Tisa-Cel, and interferon (IFN)- γ expression), patient data (age, sex, disease, tumor volume, platelet count, CRS onset, CRS grade, and treatment details), and CRS-related laboratory

data (white blood cell count, lymphocyte count, C-reactive protein (CRP), prothrombin time (PT), activated partial thromboplastin time (aPTT), fibrinogen (Fibg), inorganic phosphate (IP), and magnesium (Mg)) were collected. The day of Tisa-Cel infusion was defined as day0 (baseline) and changes in CRS-related factors were monitored.

Results: CRS developed in 19 patients, including 16 treated with tocilizumab (anti-IL-6 antibody). Patients with CRS had lower platelet counts on day 0 and higher Tisa-Cel total viable cell numbers and IFN- γ expression than those without CRS ($p = 0.011$, $p = 0.053$, $p = 0.044$). The lymphocyte count was decreased by lymphocyte depleting chemotherapy prior to Tisa-Cel infusion, but increased on day 3, suggesting an in vivo expansion of Tisa-Cel after infusion, especially in the CRS group, in which a higher count than in the non-CRS group was observed. CRP is a biomarker of inflammation that reflects IL-6 production and reached its highest level on day 4 then returned to baseline by day 8 in the CRS group. This suggested that CAR-T cells or macrophages were activated consistent with clinical symptoms. The lowest PT and aPTT values were observed on day 6 and recovered on day 10 in the CRS group. Fibg mildly increased after infusion, followed by a marked decrease on day 6, and was lower in the CRS group during the subsequent observation period. The percentage change from baseline of the IP and Mg levels on day 2 was significantly lower in the CRS group than in the non-CRS group ($p = 0.028$, $p = 0.048$) and recovered with CRS remission.

Summary / Conclusions: A low platelet count at the time of infusion, high total viable cell number of Tisa-Cel, and high IFN- γ expression may be risk factors for CRS. Changes in blood cell counts, CRP, and coagulation markers may indicate changes after the onset of CRS while IP and Mg levels significantly changed before day 3, which is the median CRS onset after Tisa-Cel therapy. This suggests that IP and Mg may be related to the mechanism of CRS onset and reflect slight changes prior to onset. Future studies with larger sample sizes are required to evaluate their clinical utility as predictors of CRS onset.

Parallel session—donors and donation

Driving blood donations—getting the word out

PA15-L01 | Confronting blood supply challenges with decreasing birthrates

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Managing blood supply is never an easy task because of a highly dynamic clinical, societal and other factors. While blood service has to be actively recruiting and retaining their blood donors to donate at right time, clinical demand is fluctuating every day rendering accurate prediction down to weekly or monthly basis almost mission impossible. More

importantly, as prospective blood donors tend to donate at their own schedule, the requirement in passing the blood donation eligibility criteria contributes further to the above difficulties. Globally, the WHO lately estimated about 118.5 million blood donations collected annually with 40% of these being collected in high-income countries which represented 16% of the world's population. Based on per 1000 population, the blood donation rate is 31.5 in high-income countries, 16.4 in upper-middle-income countries, 6.6 in lower-middle-income (LMIC) countries and 5.0 in low-income (LIC) countries. There exists a definite imbalance in the blood supply and demand and blood insufficiency for clinical transfusion in particular in LMIC and LIC countries. Indeed, up till now, the LMIC and LIC countries are still struggling for a global direction in achieving 100% VNRBD and also working hard to enhance both quality and safety of their blood supply. Therefore, sustainability of safe blood supply remains their major challenge. To tackle the blood supply issue, one obvious solution would be to ensure a healthy and young blood donor pool so that many could be mobilized regularly and at the time when there is surge or insufficiency. Every effort has to make to recruit younger donors into blood donation as they are generally healthier and easy to mobilize. However, young donors recruitment and retention have serious limitations in the past decade when their intention to participate was in fact lower than before. Moreover, a global decline in birthrate further makes the blood donors recruitment difficult. Nevertheless, efforts still need to pay to overcome the challenges in blood supply due to ageing population and hence the blood demand. While education, promotion and recruitment to the younger should maintain with innovative idea to address the needs of this group of donors, improvement in workflow, use of modern technologies of their interest might be useful to increase their participation and align to their behaviours. As the young generation may be interested at cross culture activities, collaboration of blood donation related activities with other blood services around the world may be an alternative to try out. Besides, there are definitely a need to enhance the participation of the existing blood donors so that it not only compensates the decrease in new donors recruited but also the proportion of blood supply contributed by the younger donors. In summary, with ageing and decline in birthrate globally, more challenges are found in managing a sustainable blood supply. Therefore, more efforts have to make to encourage participation of different age sectors in blood donation so as to meet the clinical transfusion demand. Experience sharing would certainly help blood services to develop better and innovative ideas and activities to tackle these.

PA15-L02 | Encouraging frequent blood donation through personalized post-donation mental reward text messages using nudging techniques

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Background: The utilization of digital innovations and mobile communication tools provides numerous opportunities in various public health issues, such as blood donation. The integration of personalized post-donation reward text messages to blood donors via SMS, with the goal of enhancing the sense of accomplishment and mental reward, has been reported to contribute to an increase in the frequency of voluntary blood donations.

Aims: Our study aimed to assess the influence of personalized post-donation SMS texts, conveying a brief message of appreciation, on donor mobilization. We applied insights from pertinent past nudge initiatives in Europe and America.

Methods: This was an A/B type experimental study conducted by the Blood Establishment (BE) of a General Public Oncology Hospital, in collaboration with an independent Nudge Unit specializing in behavioral science applications. All blood donors from the BE were randomly assigned to two groups, serving as treatment and control, for an experimental period of 18 months. Throughout the testing period, all blood donations made at BE facilities or during BE-organized donation events were randomly assigned to the two groups. Donors in the treatment group received a personalized SMS text of appreciation shortly after their donation, while donors in the control group received no SMS text. Upon completion of the experiment, the number of donations in the BE and other BEs was examined for a random 10% of first-semester donors, spanning 18 months before and after the test initiation. Data were extracted from the BE database and the National Blood Donor Registry, and a T-test for treatment and control groups concerning SMS receipt and blood donation was conducted using SPSS.

Results: During the testing period, 4494 donations were made by 3836 donors in the BE. Of these, 2273 personalized messages were sent to 1942 donors in the first group, while the 1894 donors in the control group did not receive any messages. Both groups were confirmed to be balanced in terms of age, sex, and donation frequency. The statistical analysis revealed that the difference in the number of donations before and after the experiment's initiation was significantly higher in the treatment group when analyzing donations within the BE ($p < 0.01$). Similar results were observed when analyzing total donations (within the BE and in other BEs) but with lower statistical significance ($p < 0.05$). Regarding donations within the BE, the treatment group showed a significantly greater difference in the number of donations for men, volunteer donors, and directed donors (family and friends of a specific patient) ($p < 0.1$). For total donations, the greater difference in the number of donations to the treatment group was confirmed only for men.

Summary / Conclusions: We examined whether personalized reward-ing post-blood donation SMS texts contribute to the establishment of a sustainable donating habit. The A/B experimental study, conducted over 18 months, focused on blood donors from a BE and yielded noteworthy positive outcomes for SMS texts, showing an average increase in donations both within the BE and in total donations. Donors in the treatment group demonstrated a higher frequency of returning to the BE for blood donation compared to other BEs. However, a weak inclination to return as volunteer donors was noted

among those who initially donated as family and friends of a specific patient and received an SMS.

PA15-L03 | Online campaigns—unstoppable drivers of donation

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Background: Raising awareness about the need for donation and recruiting new donors has traditionally been achieved through conventional communication channels: generating news in the media, organizing collections at strategic points and contacting current donors via email, SMS or calls.

Aims: Evaluate the outcomes of online marketing to enhance the impact among potential donors, increase the number of new donors and preferentially incorporate donors aged 18–30 to ensure generational continuity.

Methods: Between December 2023 and January 2024, investing €2,575, an online marketing campaign is launched with the following actions: (a) Google Ads campaigns displaying youth-targeted ads in the areas served by our Blood Establishment on Google channels: YouTube, Gmail, display and Discovery. (b) Social Ads campaigns showing ads on Instagram and Facebook to individuals with the desired profile. All ads were directed to our reservation website using different formats and texts for testing and optimizing those with the greatest impact. In Google Ads, remarketing campaigns were conducted, targeting individuals who clicked on the ad but did not make a reservation. All impacts and results were tracked: the number of times the ad was viewed, clicks, website visits, reservations from the ad, individuals who made a reservation, individuals who came to donate and those who ultimately donated. These figures were related to the investment, monitoring the cost per click, cost per reservation, cost per visit, and cost per donation. The cost per donation from the campaigns was compared with the cost per donation from donations obtained through a phone call to the donor.

Results:

The campaign achieved nearly 1.6 million ad views, over fifteen thousand website visits, and more than 600 donations (55 of these were

new donors). The **cost per donation was 4.3€**. When donations were arranged **through telemarketing campaigns, the cost per donation was 13€**.

Summary / Conclusions: Comparing the campaigns, Google Ads achieved a better cost per donation, conversion to clicks, and visit percentage. In contrast, Social Ads campaigns reached a larger audience and obtained a more committed reservation profile, as almost all reservation participants went on to donate. Online communication campaigns are an effective channel for spreading the need for donation. They allow the message to reach the public massively and simultaneously in a segmented manner (by target profile, geographical area, etc.). Online campaigns spark interest, generate website visits, and secure reservations and donations from both regular and new donors. The cost per donation is significantly lower than in campaigns where regular donors are called by phone. Based on these results, we will continue to test online channels to verify their potential as donation drivers.

PA15-L04 | Challenges of donor notification for transfusion-transmissible infections in sub-Saharan Africa

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Background: Systems for notification of blood donors reactive for transfusion-transmitted infections (TTIs) during donor testing in blood establishments in sub-Saharan Africa (SSA) remain weak. Although the care of certain TTIs such as HIV has greatly improved over the years, gaps exist about linkage into care for blood donors confirmed positive for TTIs. In Uganda, donor notification previously relied on the services of Uganda red cross society counsellors. Currently, due to limited resources, there is no national standardized system to deliver test results to TTI reactive donors, or refer them for care.

Aims: To describe a pilot project for donor notification and referral into care of TTI reactive donors in Uganda.

Methods: A quality improvement project on donor notification was conducted at the Uganda Blood Transfusion Services (UBTS)—Nakasero regional blood bank, Kampala, between October and December 2022, led by two graduate students during their elective placements. Data on blood donors tested during this interval were reviewed. Blood donors reactive for anyone of the four TTIs tested at UBTS (HIV, Hepatitis B, Hepatitis C and syphilis) were identified, their mobile phones contacts retrieved and affected donors were directly contacted by phone call to return for counseling, confirmatory testing and referral into care. We developed and adopted donor notification and referral letters. We summarize the findings of our pilot project.

Results: A total of 18,623 blood donations were tested (routinely), of whom 1.6% (302) were reactive for any TTIs. Only 50% (150) of these had a valid mobile phone contact in the UBTS donor registry. When contacted, only 79.3% (119/150) could be reached by phone and

PA15-L03 Table 1.

Results			
	Absoluts	Conversion	Unit cost
Investment	2.575€		
Impressions	1.634.479		0.002€
Clicks	14.889	1%	0.173€
Appointments	1.251	8%	2.059€
Visits	687	55%	3.749€
Donations	603	88%	4.271€
New donnors	55	9%	

were notified. However, only 20% (30/150) of the notified donors returned to UBTS for counseling, confirmatory testing and referral, including 14 with HIV, 12 with Hepatitis B, 2 with Hepatitis C, and 2 with syphilis. Some of the HIV-reactive donors already knew their status prior to donating, casting doubt on their motivation to donate and/or the efficiency of the permanent deferral codes applied in the donor database.

Summary / Conclusions: Gaps in knowledge and practice still remain in donor notification in SSA, while referral into care (and early diagnosis) such as for HIV is an opportunity, towards possible elimination of HIV by 2030. Alternative and innovative strategies are needed to notify TTI reactive donors about their status in SSA.

PA15-L05 | License to save lives—special agents wanted

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Background: Our Regional Blood Bank, serving 1.1 million inhabitants across four islands, struggles to ensure blood supplies to the 15 hospitals within the region. A decline in donations due to the COVID-19 pandemic, an aging population and a decrease in the society's commitment to donating blood makes the task even more difficult.

Aims: Our Regional Blood Bank launched the “License to Save Lives” campaign in 2023 to address the urging need for blood donations, particularly targeting O negative donors, a universal blood type crucial in emergency situations. The campaign aims to recruit new blood donors across all blood groups, emphasizing the importance of O negative donations, and to achieve stable reserves of all blood groups throughout the year.

Methods: The campaign, inspired by the narrative of secret agents, uses popular references from iconic films and cartoons to encourage people to become “special agents with a license to save lives”. The Blood Bank encourages to donate blood through “missions” (blood drives) and to uncover hidden heroes possessing specific qualities (healthiness, altruism, bravery, generosity, and commitment) who obtain their “license to save lives” and become a Blood Bank special agent (potential donors). Kicking off on February 2, 2023, with an inaugural event supported by health authorities, our campaign employs a dynamic and multifaceted approach. Our strategic actions include email campaigns, proactive telemarketing, and collaborations with stakeholders such as businesses, educational institutions, hospitals, and suppliers. To enhance our campaign's visibility, we created a video aired on regional television and local media platforms. Journalists played an active role in amplifying our message through coverage on social media channels. Additionally, we created radio spots to broaden the campaign's reach. In a bid to mobilize O negative donors, we encouraged them to involve their family members, identifying those who had inherited the same blood type and to serve as

advocates, promoting the importance of their blood group. We provided real-time updates on campaign progress through a new donor counter on our official website. Furthermore, we integrated the campaign into the corporate email signatures of personnel, to spread visibility. Our staff members wore informational labels about the campaign. Moreover, we rebranded the donation center to align it with the campaign theme, creating a cohesive experience for donors. A planned schedule of events and merchandise ensured an extensive reach and impact for our campaign, engaging and inspiring our community to join us in saving lives.

Results: The “License to Save Lives” campaign obtained an increase in the number of new donors. Particularly noteworthy was the increase of O negative donors, with 214 individuals joining the cause, 28.1% more than the previous year. Furthermore, both AB negative and AB positive blood groups experienced substantial growth, with increases of 133.3% and 45.3%, respectively. Overall, the campaign recruited 2467 new donors, representing a 20% increase compared to the preceding year.

Summary / Conclusions: The “License to Save Lives” campaign successfully raised awareness and boosted recruitment of new blood donors in the region, particularly the number of O negative donors, essential for emergency transfusions. It is important to continue and expand these initiatives to maintain this trend in the recruitment of new donors.

Parallel session—blood products

Plastic bags for blood components - what is new?

PA16-L01 | Scientific and regulatory overview of the non-DEHP transition

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Di-ethyl-hexyl-phthalate (DEHP) is the main plasticizer used for whole blood collection systems and red cell storage bags. DEHP has been used in blood bags since 1955 to make PVC blood bag systems flexible and allow for processing of the drawn donor blood in a closed system into the various blood components. Concerns about the health effects and endocrine disruptive consequences has resulted in European legislation to diminish or ban the use of phthalate plasticizers. The European Commission has however recently postponed the DEHP ban in medical devices, which was originally set to sunset on May 27th 2025, to July 1st 2030. As various non-DEHP whole blood and apheresis systems are still being developed, it is instrumental to cooperate in validation efforts in order to uncover the optimal combination of plasticizer and storage solution. As various blood

institutions are beginning their non-DEHP system validation efforts, more and more data is becoming available on the impact of various DEHP- and storage solution alternatives on blood component quality. To this end, efforts are undertaken to allow for sharing and international adoption of validation data. The European Blood Alliance (EBA) is drafting a validation database that allows for capturing and sharing of validation data. Also, recently, EBA has published a rationale paper on the subject of the extent of *in vitro* component validations, providing a baseline validation requirements. Together, these may prove helpful in furthering validation and implementation efforts. In parallel to the DEHP-ban, blood bag sets are up classified from class 2b medical devices to class 3, requiring clinical evaluation. Currently, it is unclear what such clinical evaluation should entail, be it a hemovigilance study, recovery study, increment assessment. In this overview, a summary of the scientific and regulatory state of the non-DEHP transition will be presented as well as the challenges and hurdles that are still to be overcome.

PA16-L02 | Bacterial proliferation is comparable in red blood cell concentrates stored in DEHT/PAGGSM and DEHP/SAGM containers

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Background: Di(2-ethylhexyl) phthalate (DEHP) has been used as the plasticizer of choice in polyvinyl chloride (PVC) plastic storage containers for red blood cell (RBC) concentrates (RBCC), in part because DEHP was found to stabilize RBC membranes. However, since phthalates have been identified as endocrine disruptors and human carcinogens, European regulators have mandated that blood storage containers be manufactured with DEHP-free plasticizers from 2030 onwards. Canadian Blood Services is investigating containers plasticized with di (2-ethylhexyl) terephthalate (DEHT), an alternative non-phthalate plasticizer. To preserve RBC quality, the change from DEHP to DEHT has been coupled with a change in the additive solution from saline-adenine-glucose-mannitol (SAGM) to phosphate-adenine-glucose-guanosine-saline-mannitol (PAGGSM). While bacterial proliferation in RBCC is limited due to refrigerated storage, it is assumed that changing the plasticizer of the storage containers and the additive solution will not affect bacterial proliferation.

Aims: Evaluate bacterial survival and growth in RBCC stored in either DEHT/PAGGSM or DEHP/SAGM containers.

Methods: Paired ABO-matched whole blood units were collected into DEHT/PAGGSM whole blood collection sets (Macopharma

PRORQT4-A 500 mL), pooled, and split evenly into one DEHT/PAGGSM and one DEHP/SAGM whole blood collection set (Macopharma LQT710X 500 mL). RBCC were produced from each of the two whole blood units using a top/bottom buffy coat process with the same centrifugation and extraction programs. The resulting RBCC were tested for baseline sterility and then the pair was inoculated with one of four transfusion relevant bacteria. *Yersinia enterocolitica*, *Serratia liquefaciens*, and *Listeria monocytogenes* were inoculated at a target concentration of 10^2 CFU/mL while *Cutibacterium acnes* was inoculated at $\sim 10^3$ CFU/mL. Spiked units were stored at $1-6^\circ\text{C}$ for 43 days and sampled at days 0, 7, 14, 21, 28, 35, and 43 for bacterial enumeration. Bacterial identification of selected samples was performed at the end of storage to confirm identity of the inoculated organisms. The study was performed in triplicate for each bacterial species.

Results: No differences in survival/growth between DEHP/SAGM and DEHT/PAGGSM RBCC were observed for *Y. enterocolitica*, *S. liquefaciens* and *C. acnes*. The first two species grew to 10^7 CFU/mL by day 7 of storage, and then to 10^8 - 10^9 CFU/mL by day 14, with no further changes until the end of storage. As expected, *C. acnes* concentration remained unchanged at 10^3 CFU/mL until day 43. Interestingly, a decline in the growth rate of *L. monocytogenes* was observed in DEHT/PAGGSM units compared to DEHP/SAGM units between days 0 and 7 of storage and then the bacterium increased its growth rate in the DEHT/PAGGSM units, reaching the same concentration ($\sim 10^7$ CFU/ml) in both types of RBCC on day 43 of storage.

Summary / Conclusions: Comparable bacterial survival and growth was observed between RBCC stored in DEHP/SAGM and DEHT/PAGGSM. The slower growth rate of *L. monocytogenes* observed at the beginning of RBCC storage in the DEHT/PAGGSM units may be due to differences in the plasticizer of the storage container, additive solution and/or quality parameters between the two types of units, which merits further investigation. Overall, this study shows that bacterial safety risk of RBCC is not increased with the implementation of DEHT/PAGGSM storage containers.

PA16-L03 | Setting the stage for comparison with the new generation of non-DEHP materials—quality and storage parameters of leukodepleted Red Blood Cell Concentrates (RBCs)

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Background: The upcoming restrictions on phthalates and the shift towards non-DEHP systems will significantly impact qualification activities in the Blood Establishments (BE). According to the “Recommendations for *in vitro* evaluation of blood components collected, prepared and stored in non-DEHP medical devices” by the European Blood Alliance (EBA), qualifying non-DEHP systems for Whole Blood (WB) collection requires a comprehensive study on the processing

PA16-L03 Table 1

	Day 1	Day 14	Day 42
MCV	86.6 ± 3.7	91.9 ± 4.6	96.1 ± 4.4
Hemolysis (%)	0.14 ± 0.02	0.18 ± 0.08	0.27 ± 0.1
Glucose (mmol/l)	31.4 ± 0.6	23.4 ± 1.1	16.1 ± 2.0
Lactate (mmol/l)	3.8 ± 0.8	19.7 ± 1.9	34.7 ± 4.8
Sodium (mmol/l)	142 ± 1.6	126 ± 2.4	109 ± 2.9
Potassium (mmol/l)	3.7 ± 0.4	28.8 ± 2.9	44.8 ± 4.8
Osm. mOsm/l	323 ± 2.9	330 ± 2.0	334 ± 1.9
pCO ₂ mmHg	38.8 ± 4.6	62.4 ± 10.7	57.5 ± 10.3
pO ₂ mmHg	57.3 ± 16.9	97.9 ± 52.3	150.3 ± 55.5

and storage conditions of blood components, to ensure the maintenance of RBCs quality and the safety and efficacy of transfused blood.

Aims: To define the reference parameters for qualifying the new non-DEHP collection systems, in accordance with EBA recommendations, based on data obtained from RBCs prepared using a conventional DEHP collection system. This framework should serve as a starting point for currently qualification activities of non-DEHP collection systems.

Methods: Ten units of leukodepleted RBCs in SAG-M, obtained from WB collection in a DEHP quadruple bag system, CQ32255 (Fresenius Kabi), and separated using CompoMat G5 (Fresenius Kabi), were evaluated. During storage, the units were analyzed on days 1, 14, and 42 for hematological parameters, residual WBCs, hemolysis, glucose and lactate levels, sodium and potassium levels, osmolarity, pO₂, and pCO₂.

Results: All units met all the regulatory quality requirements: wbc × 10⁶/unit (0.02 ± 0.01), Hb g./unit (50.1 ± 7.7), Hct% (57.4 ± 2.9). Storage parameters were aligned with literature standards (Table 1, Average ± DS).

Summary / Conclusions: This study represents the starting point for the qualification activities of non-DEHP collection systems in our BE, providing insights into maintaining quality standards during the 42-day storage of RBCs. With the transition to non-DEHP systems, this framework could serve as a basis for evaluating compliance with regulatory and quality requirements of new collection systems. Further analyses, as recommended by EBA, will be crucial under different processing and storage conditions, such as post-irradiation, to confirm the safety and applicability of these systems in real-world usage conditions.

PA16-L04 | Evaluation of the in-vitro quality of plasma and red cell concentrates prepared from leukoreduced whole blood collected in non-DEHP collection system

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Background: Because of its potential toxicity, the European Commission is sunsetting the use of DEHP in Medical Devices in Europe per

the REACH Annex XIV process. Therefore, alternative plasticizers are investigated for use in blood collection systems. Fresenius Kabi (Germany) has developed hybrid non-DEHP whole blood collection systems of which the RBC storage bag is made of BTHC-PVC while all other bags are DINCH-PVC. Promising results were obtained with this collection system equipped with an in-line RBC leukoreduction filter. In the present study, a non-DEHP collection system containing a soft, square filter for removal of platelets and leukocytes from whole blood (WB) units was investigated.

Aims: To evaluate the *in vitro* quality of plasma and red cell concentrates (RCC) prepared from leukoreduced whole blood collected in a non-DEHP collection system.

Methods: 32 WB units (500 ± 50 mL) were collected in DINCH/BTHC systems (Fresenius Kabi GQ41575). After overnight hold at ambient temperature, WB was leukoreduced and processed into a plasma and RBC unit. Component volume and cellular composition were analysed. Plasma was analysed for PT, APTT, FVIII and fibrinogen. After adding the PAGGSM additive solution, RCCs were stored in the BTHC bag at 2–6°C and sampled at day 35 and 42 for quality evaluation. As a control, 20 WB units were collected and processed in the current DEHP/SAGM WB filtration system.

Results: The absence of DEHP in the whole blood collection system had no detrimental effect on the haematological characteristics of whole blood, the volumes of the components or the cellular contamination of plasma prepared from leukoreduced whole blood. Plasma coagulation factors were not affected by filtration. During storage of RCCs, a slight swelling of the cells was observed as can be seen from the increase in MCV. Swelling was more pronounced during storage in DEHP/SAGM as compared to BTHC/PAGGSM. Haemolysis increased during storage, but at day 42 all units complied to the European requirement (<0.8%). Haemolysis at day 42 was comparable for both conditions, with mean values of 0.40 ± 0.18% (BTHC/PAGGSM) and 0.34 ± 0.11% (DEHP/SAGM). ATP levels were better maintained during storage in BTHC/PAGGSM (4.0 ± 0.7 µmol/g Hb) as compared to DEHP/SAGM (3.0 ± 0.4 µmol/g Hb).

Summary / Conclusions: The absence of DEHP in the new WB collection systems has no negative effect on the volume and composition of components prepared from leukoreduced WB. Plasma coagulation factors were not affected by filtration. RCCs prepared in non-DEHP collection system, stored in PAGGSM in BTHC-PVC storage bag for 42 days showed comparable levels of haemolysis and higher ATP levels as compared to RCCs stored in DEHP/SAGM.

PA16-L05 | Evaluation of three platelet storage bags for the storage of pooled granulocyte components

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Background: Granulocyte transfusions are given in the UK as supportive therapy in patients with life-threatening neutropenia, particularly

in those with severe infections. In NHSBT, granulocyte pools are manufactured from whole blood derived buffy coats and have a shelf life to the end of day 1 post-donation. They contain neutrophils, which are thought to be the most functionally active cellular component, as well as a double adult therapeutic dose (ATD) of platelets. In this study we used updated assays to assess granulocyte viability and function, whilst incorporating investigations of platelet quality.

Aims: To compare platelet storage bags from three manufacturers: Haemonetics, Fresenius Kabi (FK) and Terumo, for granulocyte component quality over an extended storage period.

Methods: Granulocytes were manufactured from 10 buffy coats the morning after donation and irradiated before testing. Six granulocyte components were manufactured into each pack from Haemonetics (NFP1EE), FK (T2210) and Terumo (70030). Granulocyte count, viability and functionality (through the ability to phagocytose and to induce an oxidative burst) were assessed within 4 hours of manufacture and periodically up to 48 hours post manufacture. As each granulocyte contains a double ATD of platelets, the platelet count and level of platelet activation markers were also evaluated over storage.

Results: All 18 components met NHSBT specifications for volume (175–250 mL), granulocyte count ($>5 \times 10^9$ granulocytes/Unit) and platelet count ($>480 \times 10^9$ platelets/unit). The mean pH of the samples was approximately 7.0 on day of manufacture but 24 h later (10–14 h post expiry) had decreased to below 6.4 (NHSBT lower specification limit for platelets at expiry). Four flow cytometric assays were used to assess the viability and functionality of granulocyte components over the 48 h study period: Granulocyte viability was measured using a fluorescent dead cell stain. In all bags, mean viability of granulocytes was $>90\%$ 3–4 h post manufacture. Twenty-four hours post manufacture (or 10–14h after current expiry time-point) viability had decreased to between 65% and 94%. The phagocytic capacity of granulocytes was assessed by their uptake of fluorescent bacteria. The mean levels of phagocytosis were $>89\%$ across storage, although increasing measurement variability was observed from day 2. Their respiratory burst function was assessed by pre-labelling granulocytes with a dye that became fluorescent upon drug stimulation of NADPH oxidase activity. Levels of oxidative activity were stable throughout the components shelf-life before dropping after 24 h from manufacture. Granulocyte components contain a double ATD of platelets on which levels of activation were assessed over 48 hours. In all bag types, mean levels of platelets expressing activation marker CD62P were below 15% 3–4 h post manufacture, rising to $>30\%$ after 24 h and to around 50% by the end of the study. Assessment of granulocyte and platelet quality demonstrated that the Haemonetics and FK platelet bags performed as well as the current Terumo bags and are suitable for the storage of granulocyte components.

Summary / Conclusions: The granulocyte and platelet quality parameters applied in this study demonstrate that all three of the platelet bag types tested are suitable for the storage of granulocytes, as manufactured by NHSBT. Further analyses with increased sample numbers would be needed to provide evidence for a change to the current shelf life.

Parallel session— immunobiology

Blood groups evolving to adapt

PA17-L01 | The DARC side of vivax malaria in Africa—unveiling invasion pathways into Duffy-negative erythroblasts

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Vivax malaria has long been thought to be absent from sub-Saharan Africa owing to the high proportion of individuals lacking the Duffy antigen receptor for chemokines (DARC) in their erythrocytes. The interaction between *P. vivax* Duffy-binding protein (PvDBP) and DARC is assumed to be the main pathway used by merozoites to invade reticulocytes. However, the increasing number of reports of vivax malaria cases in genotypically Duffy-negative (DN) individuals has raised questions regarding the *P. vivax* invasion pathway(s). Here, we show that a subset of DN erythroblasts transiently express DARC during terminal erythroid differentiation and that *P. vivax* merozoites, irrespective of their origin, can invade DARC+ DN erythroblasts. These findings reveal that a large number of DN individuals may represent a silent reservoir of deep *P. vivax* infections at the sites of active erythropoiesis with low or no parasitemia, and it may represent an underestimated biological problem with potential clinical consequences in sub-Saharan Africa.

PA17-L02 | Evaluating potential GATA1-dependent regulatory regions in the *RHD* gene—applications in resolving samples with normal *RHD* exons despite weak D, Del or D phenotypes

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Background: The RhD antigen is clinically important as anti-D can result in transfusion reactions and hemolytic disease of the fetus and newborn. Currently, >300 *RHD* alleles are classified into weak D, Del and D-negative phenotypes in the ISBT *RHD* allele tables, with most gene variants located in the exons. We have identified a cohort of samples with normal *RHD* exonic sequences despite a weak D, Del or D-negative phenotype. A hypothesis is that a gene variant has disrupted a transcription factor binding motif, such as GATA1, in the promoter or enhancer, in these cases. It was only in 2017 that GATA1 motif disruption in the *RHD* proximal promoter for a weak D sample, *RHD* c.1-115A>C, was reported. Even if no enhancers in the *RHD* gene have been identified yet, a bioinformatic pipeline recently predicted potential additional GATA1 motifs in *RHD* intron 1 and intron 2 (Wu *et al.*, Nat Commun, 2023).

Aims: This study aimed to evaluate potential GATA1-binding regulatory motifs in the *RHD* gene for samples presenting with normal *RHD* exons, despite weak D, Del and D- phenotypes.

Methods: GATA1 motif candidates in *RHD* were overlaid with publicly available open chromatin region data (GSE128266). Genomic DNA from weak D, Del or D- samples with normal *RHD* exons ($n = 13$) were obtained from two reference laboratories in Australia and Sweden. *RHD* zygosity by quantitative PCR was performed and D +/D- chimeras were confirmed using flow cytometry. Then, the *RHD* promoter, intron 1 and intron 2 regions were amplified by gene-specific PCR for Sanger sequencing to identify potential disruptions in the GATA1 motif candidates. Electrophoretic mobility shift assay (EMSA) was performed using nuclear extracts from human erythroleukemia (HEL) cells to assess GATA1-binding when the motif is intact and disrupted by *RHD* c.1-115A>C and *RHD* c.1-110A>C alleles. Dual luciferase assays were performed as per manufacturer's instructions using HEL cells with *Renilla* control.

Results: Bioinformatic analysis identified five of six GATA1 motif candidates overlaid with open chromatin regions in the *RHD* promoter, intron 1 and intron 2. Luciferase assays showed GATA1 motifs in both intron 1 (forward 180%, reverse 246%) and intron 2 containing the rs675072: G>A variant linked with the R² haplotype (forward 242%, reverse 146%) function as enhancers. The D phenotypes of the 13 samples with normal *RHD* exons comprised of nine weak Ds, one query D variant, two routinely typed D-negative (r⁻r) and one Del. The qPCR showed nine were hemizygous for *RHD*, two were homozygous and two were D+/D- chimeras. RBCs available from one D+/D- chimera confirmed there were

two populations (78.4% D- and 20.7% D+) by flow cytometry. Sequencing showed GATA1 motifs were intact in 12 of 13 samples. Coincidentally, the *RHD**DEL5 allele was detected in one presumed r⁻r sample. In a sample with a Del phenotype, a novel *RHD* c.1-110A>C allele had disrupted the GATA1 motif in the promoter. The *RHD* c.1-110A>C allele was found to disrupt GATA1 binding more severely than the *RHD* c.1-115A>C allele in the EMSA. Reduced transcription for *RHD* c.1-110A>C (73%) was observed compared to the wild-type (100%) and *RHD* c.1-115A>C (96%) constructs in the luciferase assay.

Summary / Conclusions: Our study resolved a sample with a Del phenotype arising from a novel *RHD* c.1-110A>C allele, which disrupted the GATA1 motif in the promoter. Although most of the samples investigated remain unexplained, we provide GATA1 target regions which may be useful to future *RHD* regulatory investigations.

PA17-L03 | Gene-modified PBDEP-4 cell line enables the production of artificial panel cells for antibody screening and identification

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Background: The identification of alloantibodies is an important examination to prevent hemolytic transfusion reactions. In the currently used protocol, antibody identification panels are prepared using peripheral red blood cells (RBCs) with a specific combination of antigens. However, antibodies to high-frequency antigens (HFAs) and mixed alloantibodies are difficult to identify unless rare RBCs lacking each of the HFAs are being used. Therefore, efforts to produce artificial panel RBCs have progressed for the easy identification of alloantibodies in blood donors and recipients. We previously established peripheral blood-derived erythroid progenitor (PBDEP)-4 cells that were immortalized by tetracycline-inducible HPV E6/E7 genes. PBDEP-4 cells can undergo gene modification and possess erythroid differentiation potential. After a 7-day differentiation, PBDEP-4 cells differentiate into late erythroblasts, displaying a deep red color. By using PBDEP-4 cells, which can proliferate indefinitely and can be gene-modified, it is possible to stably supply blood cell reagents of the same blood type, including rare blood types.

Aims: The objective of this research was to produce antibody identification panels that express only few or a single antigen using PBDEP-4 cells. The PBDEP-4 cell line was established from an O-type volunteer donor with RhD/Ce, P1, Fy^a, Jr^a, Jk^a, Jk^b, Di^b, and MNs antigens (Table 1). The method using PBDEP-4 cells and the CRISPR/Cas9 system may enable the artificial production of blood cell reagents that are reproducible and convenient and facilitate easier identification of alloantibodies than ever before.

Methods: We attempted to delete major HFAs (Rh, P1, Fy, Jr, Jk, and MNS) expressed in PBDEP-4 cells by the CRISPR/Cas9 system. Genome editing of these HFAs was applied to PBDEP-4 cells by

PA17-L03 Table 1

Blood type	PBDEP-4	PBDEP-4/del
Rh	DCe	KO (antigen deleted)
P	P1	KO
Duffy	Fy (a+b-)	KO
JR	Jr (a+)	KO
Kidd	Jk (a+b+)	KO
MNSs	M+N+S-s+	on going
Diego	Di (a-b+)	Di(a-b+)

electroporation using CRISPR/Cas9 plasmids corresponding to target antigens. The genome-edited cells were cloned to a single cell using a cell sorter. Subsequently, flow cytometry analysis using monoclonal antibodies and Sanger sequencing were performed on each proliferated single clone cell.

Results: Target antigens (RhD/Ce, P1, Fy^a, Fy^b, Jr^a, Jk^a, and Jk^b) were not detected by flow cytometry analysis using post-genome-edited and differentiated PBDEP-4 cells (PBDEP-4/del). Residual major antigens were only MNs and Di^b in PBDEP-4/del (refer to Table 1). Sanger sequencing revealed that the target genes (listed above) had undergone frameshift mutations, suggesting that genome-edited cells express truncated proteins that do not react with antibodies.

Summary / Conclusions: We successfully produced a cell line that minimally expresses the major HFAs. This panel cell line was generated using an immortalized erythroid cell line and is amenable to gene modifications, including forced expression or antigen deletion. These findings strongly indicate the feasibility of artificially producing a panel cell that expresses only a limited antigens. We are now trying to produce a Di^b single-antigen-expressing cell that lacks even MNS antigens. We expect that this method can be applied to any major antigen to produce artificial panel cells for simply identifying alloantibodies against each of the major antigens in blood donors and recipients.

PA17-L04 | Comprehensive RHD and RHCE sequence analysis for the screening and characterization of RH variants in African origin donors

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Background: Due to migration movements from Sub-Saharan countries, the number of African origin patients with chronic transfusion requirements, for example, sickle cell disease (SCD), has increased exponentially over the last decade in our country. The high prevalence of molecular variants within African populations, particularly in the RH

system, coupled with an elevated incidence of red cell alloimmunization, complicates the transfusion support for these patients. Over the last five years, a big effort has been made to recruit African descent donors through a targeted campaign, with the objective to increase the likelihood of finding compatible red blood cell (RBC) units for alloimmunized SCD patients.

Aims: This study aims to establish a database of extensively typed African origin donors using a previously developed nonspecific quantitative next-generation RH sequencing (NGS) method.

Methods: The recruited donors of Sub-Saharan origin ($n = 374$) were extensively phenotyped for red cell antigens and tested for platelet GPIV deficiency. Blood group genotyping was also performed with ID CORE XT (Progenika-Grifols) for further characterization. Donors meeting specific RH genotype criteria (ce-733G/ce-733G; ce/ce; Ce/Ce; ce-733G/cE genotypes) were subjected to nonspecific quantitative RH sequence analysis ($n = 162$). The assay targets RH exons 1 to 10 and portions of intron 2 using M13-tagged primers that bind to consensus RH sequences, making amplification proportional to the copy number in genomic DNA. Amplicons were generated in two separate multiplex PCRs using either Set A or Set B primers to minimize the likelihood of data misinterpretation due to allele drop-out (Stef 2020). Multiplex PCR reactions were performed with QIAGEN Multiplex PCR Kit and treated with ExoSAP-IT. Indexes and adaptors were incorporated in a second round of amplification. The pooled library was sequenced using a paired end (2×250) sequencing protocol on the MiSeq system (Illumina) with a standard v2 Reagent Kit. Sequencing reads were analyzed with a custom-developed data analysis software coded in Python language (Stef 2020).

Results: Sequencing analysis of 162 donors revealed 35 having clinically relevant RH genotypes encoding partial Rh antigens and/or lack of high prevalence RhCE antigens. The following alleles were detected in homozygous state: CeRN (RH:-46, partial C, $n = 2$), ceAG (RH:-59, partial e, RH:-31, $n = 1$) and ce.VS (RH:-31, partial e, partial c, $n = 9$). We detected the ceAR allele in trans to common cE (partial e and probably RH:-19, $n = 1$) and the ce.VS in trans to cE (partial e, $n = 8$) or in trans to Ce (partial c, $n = 1$). We found RHD variant alleles, other than DAU0, in hemizyosity or compound heterozygosity in 9 donors (DAU3, DAU5, DIVa, DIIIc, DAU6, DFV) leading to partial D phenotype, and the DIIIa-CE(4-7) genotype encoding partial C in 5 donors. Furthermore, we detected alleles involving copy number variations and a novel DAU0+635-1G>A allele.

Summary / Conclusions: Non-specific quantitative NGS emerges as a valuable tool for RH genotyping, particularly in populations with complex genetic profiles like those of Sub-Saharan Africa. Its application to the screening and characterization of RH variants in our cohort of African ancestry blood donors has allowed us to identify clinically relevant rare genotypes in up to 9% of these donors. These findings will help us to improve the availability of compatible RBCs for RH-alloimmunized patients of African descent.

PA17-L05 | Interplay between red blood cell glycolipids and enzymes targeting the A and B antigens

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Background: Blood group A, B and AB blood requires careful matching before transfusion to avoid potentially life-threatening haemolytic reactions, while group O blood is considered safe for transfusion to recipients of all ABO types in most cases. Enzymatic conversion of ABO antigens on red blood cells (RBCs) to the universal group O (the ECO-blood concept) remains a long-sought goal to simplify blood management and eliminate the risk of erroneous mismatching of blood types. The discovery of several efficient bacterial exoglycosidases has resulted in apparent removal of A and B antigens. Despite this, persistent positive crossmatches remain between ECO-RBCs and recipient plasma, raising safety concerns and hindering clinical implementation. The cause of these positive crossmatch reactions remains a conundrum. Recently we discovered efficient enzyme cocktails in *Akkermansia muciniphila*, capable of removing not only the canonical A and B antigens, but also all known extensions thereof. This provides, for the first time, a unique opportunity to evaluate the full-removal of all known ABO antigens.

Aims: The aim of our work is to perform a glycome-scale analysis of RBC surface glycoconjugates to map the diversity and abundance of different ABO epitopes across different blood group phenotypes and to dissect the precise modifications of the glycomes by enzymes targeting the A and B antigens.

Methods: Enzymes for glycoconjugate digestion have been expressed in bacterial and yeast expression systems, to enable the subsequent glycan analysis. The enzymatic conversion of RBC surface glycans will be analysed using liquid chromatography-electrospray ionization-tandem mass spectrometry (LC-ESI-MS/MS).

Results: Preliminary data demonstrate endoglycosidase II activity on RBCs. Six glycolipid-derived glycan species, including the blood group A antigen have been detected using our methods, releasing glycans from glycolipids on A1 erythrocyte surfaces. Released glycans were analysed by LC-ESI-MS/MS. The latest results from the glycome generation and analysis will be presented at the congress.

Summary / Conclusions: The interactions of RBC glycomes, especially from glycolipids, with enzymes has not been investigated to date. This study will provide new insight into the enzymatic modification of RBC glycomes and potentially contribute to a molecular understanding of the glyco-signatures that underpin incompatibility issues of ECO-blood with recipient plasma.

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Parallel session—blood safety

Fine tuning donor selection and screening

PA18-L01 | Impact of removing variant Creutzfeldt-Jakob Disease (vCJD) deferral on donor recruitment in Australia

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Background: Variant Creutzfeldt-Jakob disease (vCJD) emerged in the UK, first reported in 1996 secondary to consumption of beef during 1980–1996 from cattle infected with bovine spongiform encephalopathy. Due to concerns about blood transfusion transmission, precautionary donor deferrals were introduced worldwide including in Australia. Subsequently three reported cases and one potential sub-clinical case of vCJD transmission by transfusion in the UK were documented. In Australia this deferral resulted in the loss of approximately 5% of the donor panel and multiple and ongoing complaints. With changes in agricultural practices, the epidemic in cattle and the crossover to humans was terminated. By 2017 it was documented the original vCJD human epidemic was petering out, and the feared second wave had not materialised, so the Australian Red Cross Lifeblood with the Kirby Institute, University of New South Wales planned a modelling study, which showed removal of the geographic deferral would result in an infinitesimally small risk of vCJD transmission 1:389 million with a clinical case risk being 1 in 1.45 billion [McManus, Vox Sanguinis 2022]. The Australian regulator approved removal of this deferral. Subsequently Lifeblood also applied to have the deferral for having a transfusion in the UK removed demonstrating even with conservative assumptions where transfusion cases were modelled to be higher than dietary cases that the risk was negligible, which was approved.

Aims: To describe initial and ongoing effects of removal of the geographic and transfusion vCJD deferrals.

Methods: Lifeblood databases were examined for donation changes resulting from removal of these deferrals. This was facilitated by retention of the donor questionnaire question about UK residency from lifting of the geographic deferral on 25/7/2022 and 12/2/2023.

Results: The initial modelling predicted an uplift of 17,000 donors and 58,000 donations per annum with removal of the geographic deferral. In the first six months 67,914 donations excluding discards occurred, more than double predicted. This occurred because a higher number of predicted donors donated at a higher frequency than predicted, facilitated by the fact plasma donors can donate up to fortnightly. Transfusion in the UK was the top reason previously deferred donors

remained deferred and was a unique deferral in 0.19% of donors who attended prior to removal. The vCJD transfusion deferral was lifted on 13/11/2023. Monitoring of vasovagal reactions and other donor adverse events in the 6-month period July to December 2022 showed an unadjusted decrease of -3.3%.

Summary/conclusions: Removal of the vCJD geographic and transfusion deferrals resulted in a significant sufficiency boost, and a cessation of complaints about these deferrals, and the return of many older donors who had been deferred for decades. Any residual risk impact is unlikely ever to materialize given the calculated residual risk. Other blood services such as New Zealand have used the Lifeblood/Kirby modelling to present successful argument to their regulators to have their deferral removed. Lifeblood also removed the vCJD deferral for milk donors.

PA18-L02 | Epidemiological features of hepatitis E virus infection among blood donors in Japan revealed by universal NAT screening

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Background: A total of 45 cases of transfusion-transmission of hepatitis E virus infection (TT-HEV) have been reported until 2020 in Japan, including two cases with low HEV load that may be undetectable by individual-donation nucleic acid amplification testing (ID-NAT). In 2020, an ID-NAT (Procleix[®] UltrioPlex E) that detects hepatitis B virus (HBV), hepatitis C virus (HCV), human immunodeficiency virus (HIV)-1/2, and HEV was implemented, and no cases of TT-HEV have been reported in Japan since then.

Aims: To document the geographical and chronological incidence of new HEV infections in Japan, we conducted a molecular epidemiological analysis of the nationwide data obtained by HEV NAT.

Methods: The HEV NAT positivity rate (PR) from Aug 2020 to Dec 2023 was analyzed by sex, age group, and area. The nucleotide sequences of the HEV ORF2 region of positive blood donation (BD) samples were determined, and molecular phylogenetic analysis was conducted along with previously reported strains.

Results: Overall, 17,166,516 ID-NAT were conducted. The PRs per 100,000 BD were 6.7, 1.9, 0.6, and 65 for HBV, HCV, HIV-1/2, and HEV, respectively. The HEV PR increased significantly from 50 to 80 during 2020–2023. The HEV PRs in every 10-year age group from 10 to 60 years of age were 40, 82, 81, 78, 70, and 51 in males and 31, 60, 59, 57, 44, and 34 in females. Regional HEV PRs, from north to south, were 65 in Hokkaido, 72 in Tohoku, 104 in Kanto-Koshin'etsu, 47 in Tokai-Hokuriku, 35 in Kinki, 30 in Chugoku-Shikoku, and 30 in Kyushu. The monthly HEV PR varied from 42 to 116, with the highest in Jan of each year; 68, 73, and 116 in 2021, 2022, and 2023, respectively. The HEV-PR was highest, at 176, in Jan 2023 in the

Tokyo area. Of the 1,113 HEV genotypes determined between Aug 2020 and Jul 2021, 3a and 3b accounted for 92%, and HEV-4 for 1.2%. While multiple clusters of indigenous strains were identified in each region, there were also independent strains with over 99% homology with strains isolated in foreign countries. Since travel restrictions due to the COVID-19 pandemic were imposed during the period, imported food could be a source of the infection. A rabbit HEV strain was first identified in Japan that had a low homology of 89% with previously reported strains, suggesting unknown sources of HEV infection. Of the 250 HEV strains isolated from Jan to Apr 2023 in the Tokyo area, 80% belonged to the same cluster of BD-derived strains detected in the same area in 2020–2021. Among these, there were five large clusters identified, with each including 10 or more strains (pairwise distance: 0.000–0.003).

Summary / Conclusions: The incidence of new HEV infections was nearly 10 times higher than that of HBV in Japan, with multiple indigenous HEV strains existing throughout Japan. The HEV epidemic was seasonal and regional, with particularly high incidence rates in the Tokyo area during the winter. The fact that 80% of the strains found in the Tokyo epidemic belonged to the same cluster suggests that common sources of infection, such as HEV-contaminated food, have spread widely. Some strains have also been detected over several years, indicating that the source of infection has been maintained in some areas. While TT-HEV has been prevented due to the implementation of blood screening with high sensitivity ID-NAT, the high incidence of new HEV infections among the general population raises a significant public health concern. Fundamental measures to reduce HEV infections throughout the nation should be implemented.

PA18-L03 | HEV Infection in blood donors in France—first results from NAT testing

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Background: In France, Nucleic acid Testing (NAT) for HEV was introduced in 2012 in pools of 96 apheresis plasma donations for plasma intended for SD- plasma production. From the end of 2014 onwards, HEV-NAT was continued in minipools (MP) of 6 donations to display HEV-free plasma components targeted towards at-risk patients. Since March 2023 (except in apheresis plasma for fractionation), all donations are systematically tested for HEV-RNA.

Aims: To analyse data obtained over the first 9 months of systematic HEV-NAT testing.

Methods: Approximately 90% of donations are tested in MP6 with the Cobas HEV assay Roche (Lod95%:18IU/ml) and 10% in Individual (ID)-NAT with UltrioPlex E, Grifols (Lod95%:3,6 IU/ml). Positive donations are discarded and analysed for viral load (VL) (AltoStar HEV 1.5

Altona, Lod: 10 IU/ml), serology (HEV IgM and IgG Liaison, DiaSorin) and genotype at the national reference center (NCR). Positive donors are deferred for 4 months. The archived plasma sample of positive donors who donated in the previous 4 months are retrospectively tested for HEV-RNA at the NCR. If this previous donation (d.n-1) is positive a lookback study is performed to identify an infection in the recipient.

Results: From March to December 2023, 1634 in 1,800,945 tested donations were HEV-RNA positive accounting to an overall incidence at 0.91/1000 donations. The incidence rates varied from 0.53 in the North West to 1.94/1000 donations in the South West of the country. Of the 1,630 HEV positive donations further investigated at the NCR, 1487(91.2%) had a quantifiable VL (mean 127,114 UI/ml, median 441 (67-4212) UI/ml). All the 472 samples that could be genotyped were of genotype 3 (75% 3c, 17% 3f, 3% 3m). Among the 1,630 positive donations 32.7% were seropositive (5.5% IgM+/IgG-, 5.5% IgM-/IgG+, 21.7% IgM+/IgG+).

Of the 1,634 HEV-RNA positive donors, 1490 (91.2%) were repeat blood donors and 554 (37.2%) of them donated in the previous 4 months. To date, 25 d.n-1 were retrospectively tested positive: 14 had not been screened by NAT (11 plasma for fractionation and 3 collected before NAT implementation) and 11 were negative in MP-NAT. The 14 donors for whom d.n-1 resulted to labile blood components had VLs between <10 to 138 IU/mL and a mean delay between d.n-1 and index donation at 63 days (29-98). Lookback results from these 14 positive d.n-1 were available for 6 patients and none of them had been infected. The d.n-1 with the highest VL (138 IU/ml) was an apheresis platelet concentrate (APC) collected 29 days before the index donation. This APC was divided into 4 components each containing an estimated amount of virus between 2277 and 7071 IU. These components were transfused to 3 patients. One recipient died from his pathology, the other two were HEV negative.

Summary / Conclusions: In France, in 2023, the HEV-RNA incidence in blood donors was 0.91/1000 but significant regional variations were observed. Retrospective analysis of previous donations from HEV-RNA-positive donors showed that the viremia could reach almost 100 days but with very low levels. Lookback investigations and haemovigilance data did not identified any transmission of the virus by transfusion since the systematic implementation of HEV-NAT.

PA18-L04 | Evidence of non-disclosed malaria history in regular donors after implementation of malaria screening

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Background: In non-endemic countries, selective malaria screening in blood donations is eventually implemented in order to increase blood safety and reduce the number of travel or residence deferrals. Contrary to other transfusion-transmitted infectious diseases, the lack of

confirmatory assays limits the capability of Blood Banks to accurately inform the donor either of true positive or false positive result.

Aims: Assess the usefulness of post-donation epidemiological survey in order to better characterize the probability of exposure in malaria positive donors.

Methods: In November 2022, selective malaria screening in blood donations was implemented for the first time in the Blood Bank of Catalonia (Spain). Anti-Plasmodium sp. antibodies were determined by ELISA (CAPTIA™ Malaria EIA, Trinity Biotech). Donors that had a repeat reactive result were contacted in order to be informed and were asked complete an epidemiological survey.

Results: From November 2022 to December 2023, 15,825 out of 321,874 blood donations (4.9%) were tested for malaria. Regarding the origin of the donors, 30.7% were born in malaria endemic countries of Latin America, 3.5% in Asia, 0.7% in Africa, and 65.1% were born in non-endemic countries. Two hundred donors (1.3%) were positive for malaria antibodies, of them, 46% of positive donors were born in Latin America, 18% in Asia, 13% in Africa and 23% in non-endemic countries. Epidemiological data could be prospectively obtained from 96 malaria positive donors (48%). Malaria history or compatible symptoms were completely ruled out by 49 donors (malaria S/Co 1.90 ± 1.35), of them 32 donors (65%) were from non-endemic areas. Conversely, 47 donors reported high probability of having been exposed to malaria (malaria S/Co 8.08 ± 5.85), of them 38 (81%) were born in endemic areas. Interestingly, 15 donors acknowledged a malaria episode, undisclosed in the present donation or, for 13 regular donors, in the previous donations. These donors were born in Honduras (N = 4), Colombia (N = 2), Ecuador (N = 2), India, Brazil, Senegal, Peru, Tanzania and Spain (N = 2).

Summary / Conclusions: Epidemiological questionnaire was useful in order to provide the donor with a more accurate information when found positive for malaria antibodies. As much as 15% of malaria positive donors were fully aware of, but did not disclose, their history of malaria despite a specific question in the blood donation questionnaire.

PA18-L05 | Bimodal distributions of anti-T.cruzi antibody levels in blood donors are associated with parasite detection and antibody waning in peripheral blood

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Background: Discordant *T. cruzi* serological results are common in Chagas disease screening, making the confirmation of infection and counseling difficult. In our previous study of blood donors in the

Argentinian Chaco Province, where Chagas disease is highly prevalent, we documented bimodal distributions of anti-*T. cruzi* antibody (Ab) levels, suggesting potential self-cure in donors with low-reactive samples.

Aims: This study aimed to correlate antibody levels with PCR results, to identify possible Ab thresholds indicative of persistent parasitemia; and to investigate whether individuals with low Ab levels and negative PCR exhibited Ab decline over time.

Methods: Blood donors who screened reactive for *T. cruzi* Ab at the *Servicio Especializado en Hemoterapia* in Chaco Province, Argentina from 2009 to 2018, were invited to attend an initial study visit and a follow-up two years later. Blood samples from both visits were tested using four serology assays: a parasite lysate Enzyme ImmunoAssay (EIA) (ELISA lisado, Wiener Lab.), a recombinant EIA (ELISA Recombinant, Wiener Lab.), Chemiluminescent (Chagas) ImmunoAssay (CMIA) (Architect, Abbott), and VITROS Immunodiagnostic Products Anti-*T. cruzi* (Chagas) assay (Ortho Clinical Diagnostics, Raritan NJ). PCR was performed on visit 1 samples. The VITROS Ab testing and *T. cruzi* target capture PCR on lysed whole blood were performed at Vitalant Research Institute, San Francisco, California; while the remaining assays were performed by the Chaco blood center.

Results: 452 donor samples were tested on all four assays at visit 1, with 68 (15%), 28 (6.2%), 29 (6.4%), 38 (8.4%), and 289 (64%) testing positive on 0, 1, 2, 3, and 4 assays, respectively. Non-reactive samples were assumed to be false positives on the initial donation screening and excluded from the analysis. Ab titers were clearly bimodal on all four assays and fit finite mixture models of two normal distributions. Ab titers were classified as “high” or “low” at a threshold chosen for each assay to optimally separate the two assumed latent distributions. 159 and 178 samples were fully concordant with four low and four high Ab results, respectively. The remaining 48 were

discordant. In the follow-up visit relative Ab decline was significantly higher among those classified as low Ab levels, considering assays with better precision as chemiluminescent EIAs Abbott median change −24% vs. −11% ($p < 0.001$) and Vitros −23% vs −2% ($p < 0.001$) for low and high Ab levels, respectively. PCR results according to Ab status for each test are shown in the table below. Low-react samples in all assays were PCR-negative.

Summary / Conclusions: Low Ab reactivity is associated with the absence of parasitemia. Greater Ab declines were detected among donors with low and/or discordant Ab reactivity and PCR-negative results, strongly suggesting parasite eradication in these donors.

Parallel session—clinical

Patient safety—from the lab to the clinic

PA19-L01 | Use of blood products in ex vivo perfusion of organs

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Organ transplantation is the only “curative” option for end-stage organ failure. As a potential strategy to enlarge the donor pool in order to reduce the pressing donor shortages, the transplantation field is now exploring ex-vivo normothermic machine perfusion (NMP) as an assessment tool for high-risk kidney grafts and as a means of achieving more physiological organ preservation and possibly ex-situ resuscitation of more marginal kidney grafts. NMP critically relies on the inclusion of an oxygen carrier in the perfusion protocols. In order to ensure clinical compatibility, most NMP protocols opt for stored red blood cells (RBCs) as oxygen carrier. While NMP has been successfully implemented for liver, progressive per-perfusion hemolysis has been reported during renal NMP. In an in-depth analysis, we confirm the occurrence of progressive hemolysis during 6-hour kidney NMP. Observed progressive perfusion-specific erythrocytosis in the glomeruli and in peri-glomerular vascular networks during NMP pointed to an interaction between the RBCs and the graft. Continuous hemolysis resulted in prooxidative changes in the perfusate, which could be quenched by addition of fresh frozen plasma. A cell-based reporter system showed that the NMP-associated hemolysis induced redox stress and exhibited toxic effects at higher concentrations. Further development and implementation of renal NMP critically relies on a better understanding of the physiologic interactions between the kidney graft and RBC under the conditions of NMP.

PA18-L05 Table 1

Serological test	Positive PCR N = 127*	Negative PCR N = 255*
CMIA (9.7%), n (%)		
High	124 (68)	60 (32)
Low	3 (2)	195 (98)
VITROS (5.2%), n (%)		
High	127 (67)	63 (33)
low	0 (0)	192 (100)
Lysate EIA (4.7%), n (%)		
High	126 (62)	77 (37)
Low	1 (0)	178 (100)
Recombinant EIA (4.6%), n (%)		
High	126 (58)	92 (42)
Low	1 (0)	126 (100)

* Total of 382 donors with at least one positive serology result at visit one and a valid PCR (two missing). % thresholds used to divide bimodal antibody distributions into high and low values. These values were defined by fitting mixture models (assuming bimodal normal) and using expectation maximization to select the optimum cutoff.

PA19-L02 | Serious transfusion related adverse events associated with a restrictive versus liberal blood transfusion strategy in patients with myocardial infarction and anemia—MINT randomized controlled trial

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Background: In the MINT trial, we randomized 3504 patients who presented with myocardial infarction (MI) and anemia to either a restrictive or liberal red blood cell (pRBC) transfusion strategy. Rates of recurrent MI or death at 30 days tended to be higher in the restrictive group (relative risk = 1.15; 95% confidence interval, 0.99 to 1.34). Given the results favored administering more pRBCs, we believe it is important to quantify the risks of transfusion related adverse events.

Aims: We hypothesized that patients with MI and anemia randomized to a liberal transfusion strategy would be at increased risk of serious transfusion related adverse events compared to patients randomized to a restrictive transfusion strategy.

Methods: We focused our analysis on serious transfusion related adverse events including transfusion associated circulatory overload (TACO), transfusion related acute lung injury (TRALI), anaphylactic transfusion reaction, and transfusion associated sepsis. TACO is of particular concern in patients with MI. It was defined as presence of signs and symptoms of heart failure that necessitated treatment if there was a transfusion within 6 hours of the onset of heart failure. Overall rates

PA19-L02 Table 1. Serious Transfusion Related Adverse Events by Arm - A

Per Participant			
	Restrictive N = 1749 n (%)	Liberal N = 1755 n (%)	Rate Ratio R vs. L* (95% CI)
TACO	8 (0.46)	23 (1.31)	0.35 (0.16, 0.78)
TRALI	0 (0.00)	5 (0.29)	-
TACO or TRALI	8 (0.46)	28 (1.60)	0.29 (0.13, 0.63)
Anaphylactic transfusion reaction	2 (0.11)	2 (0.11)	1.00 (0.14, 7.12)
Transfusion Associated Sepsis	0 (0.00)	1 (0.06)	-
Any of the above	10 (0.57)	31 (1.77)	0.32 (0.16, 0.66)

PA19-L02 Table 2 - B

Per 100 Units of pRBC ^{\$}		
Restrictive pRBC = 1237	Liberal pRBC = 4325	Rate Ratio R vs. L* (95% CI)
0.65	0.53	1.06 (0.46, 2.48)
0.00	0.12	-
0.65	0.65	0.87 (0.38, 2.00)
0.16	0.05	3.50 (0.49, 24.82)
0.00	0.02	-
0.81	0.72	1.02 (0.28, 2.13)

* R vs L = Restrictive vs Liberal.

^{\$} Rxn as in 1A.

per group were calculated as well as rates per patient and per 100 units of pRBC transfused. All events were reported by investigators at each participating site and were not centrally adjudicated.

Results: Of patients in the restrictive transfusion group, 33.7% received pRBC transfusion compared to 94.8% in liberal transfusion group. In the restrictive transfusion group 1237 units of pRBC were administered, compared to 4325 units in the liberal transfusion group. The rate of serious transfusion related adverse events was higher in the liberal arm per participant (absolute difference 1.2%; Rate Ratio for Restrictive vs. Liberal [RR] = 0.32, 95% CI [0.16, 0.66]), however, the difference was not statistically different per 100 units of pRBC (RR 1.02, 95% CI [0.48, 2.13]) (Table 1A and B). Similarly, higher rates of TACO or TRALI events were reported in the liberal arm per participant, (RR 0.29, 95% CI [0.13, 0.63]) however, the difference was not statistically significant per 100 units of pRBC transfused (RR 0.87, 95% CI [0.38, 2.00]) (Table 1A,B). No significant differences in anaphylactic transfusion reactions were found either per participant or per units transfused. Only one instance of transfusion associated sepsis was observed, which occurred in the liberal arm.

Summary / Conclusions: More than three times as many transfusions were given to patients in the liberal group, and with it a proportionate increase in transfusion-related adverse events. Rates were not sufficiently frequent and serious in the liberal transfusion strategy to offset the increase rates of death or MI observed in patients treated using a restrictive transfusion strategy.

PA19-L03 | Soluble CD40 ligand mediates endothelial cell cytotoxicity via neutrophil extracellular trap induction in a two-hit neutrophil-dependent pathway of transfusion-related acute lung injury (TRALI)

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PA19-L03 Table 1. percentage of neutrophils forming NETs

1st hit	2nd hit (sCD40L)	% cells forming NETs/field (Mean \pm SEM)	Multiple comparison
None	0 ng/mL (control)	0.000 \pm 0.000	-
None	10 ng/mL	0.131 \pm 0.131	$p = 0.8570$ versus no treatment
None	100 ng/mL	0.060 \pm 0.060	$p = 0.9042$ versus no treatment
TNF- α	0 ng/mL (control)	0.339 \pm 0.192	$p = 0.5367$ versus no treatment
TNF- α	10 ng/mL	4.507 \pm 0.447	$p = 0.0061$ versus sCD40L (10 ng/mL) $p = 0.0005$ versus TNF- α alone
TNF- α	100 ng/mL	8.958 \pm 0.675	$p < 0.0001$ versus sCD40L (100 ng/mL) $p < 0.0001$ versus TNF- α alone

PA19-L03 Table 2. HLMVEC cytotoxicity

1st hit	2nd hit (sCD40L)	Intervention	cells forming NETs/field (Mean \pm SEM)	Multiple comparison (vs TNF- α only)
TNF- α	0 ng/mL (control)	None	18.04 \pm 2.65%	-
TNF- α	10 ng/mL	None	47.32 \pm 6.3%	$p = 0.005$
TNF- α	100 ng/mL	None	54.86 \pm 10.66%	$p = 0.0001$
TNF- α	10 ng/mL	DNase (1U/mL)	26.89 \pm 5.6%	$p = 0.0001$ versus LPS + 10 ng/mL sCD40L

Background: Transfusion-related acute lung injury (TRALI) can be caused by the transfusion of biological response modifiers (BRMs) which accumulate in blood products during storage. Soluble CD40 ligand (sCD40L) is a BRM, released from platelets, that caused TRALI in an in vitro model via a two-hit neutrophil activation pathway. DNase protected or rescued mice from TRALI by disrupting neutrophil extracellular trap (NET) formation.

Aims: To investigate whether sCD40L induces NET formation in vitro. To use an in vitro model of human pulmonary microvascular endothelial cell (HLMVEC) cytotoxicity to test whether DNase prevented sCD40L-mediated HLMVEC cytotoxicity.

Methods: *NET assay:* Isolated neutrophils were seeded on poly-L-lysine coated glass coverslips in 24 well plates (2×10^5 cells/well). Neutrophils were primed (5 ng/mL tumour necrosis factor alpha (TNF- α); 30 minutes) and treated with sCD40L (10 ng/ μ L or 100 μ g/mL; 180 min). Neutrophils were fixed (2% paraformaldehyde), permeabilised (0.5% Triton X-100), blocked and stained (anti-DNA Histone-1 and anti-human neutrophil elastase), counterstained (Hoechst 33342), mounted in ProLong Gold, and visualized by fluorescence microscopy. Percentage of cells making NETs was quantified from 6 to 8 non-overlapping fields in 3 wells for each treatment. *HLMVEC cytotoxicity assay:* Confluent HLMVECs were treated with 2 μ g/mL lipopolysaccharide (LPS) and cultured for 6 h (37°C, 5% CO₂). Freshly isolated neutrophils were added (1:10 neutrophil:HLMVEC ratio). Cells were either left untreated or were treated with sCD40L (10 ng/mL or 100 ng/mL) for 30 minutes. DNase (1U/mL) was added simultaneously to some wells. After trypan blue staining, 3-5 fields per well were acquired with Olympus microscope with 10X objective and viable HLMVECs were identified by ImageJ analysis. *Statistical analyses:* Data were analysed by repeated measures

one-way analysis of variance (ANOVA) followed by Tukey's post hoc test. $p < 0.05$ was considered significant.

Results: Soluble CD40L treatment induced NET formation in freshly isolated neutrophils pre-treated with TNF- α in a dose-dependent way (Table 1). Both LPS and neutrophils were required for sCD40L-mediated HLMVEC cytotoxicity (Table 2), confirming the two-hit neutrophil activation pathway previously described. Addition of DNase to the TRALI model mitigated sCD40L-mediated HLMVEC cytotoxicity.

Summary / Conclusions: Concentrations of sCD40L comparable to those found in platelet concentrates induced in vitro NET formation. Also, in an in vitro TRALI model, these sCD40L concentrations mediated HLMVEC cytotoxicity. The addition of DNase mitigated sCD40L-mediated HLMVEC cytotoxicity.

PA19-L04 | Infusion of allogeneic extracellular vesicles of tolerogenic cells might still carry the risk of exacerbating TRALI mortality

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Background: Transfusion-related acute lung injury (TRALI), the main symptom of acute respiratory distress syndrome (ARDS), is the main post-transfusion complication leading to transfusion-related mortality in recent years. It has been reported that the presence of dendritic cells (DC) plays an important role in protecting the body from lung injury mortality. Our earlier studies found that rapamycin-treated DCs have a more stable immune tolerance function and have the potential

to control TRALI. Extracellular vesicles (EVs) have stronger application prospects than cell therapy because of their low immunogenicity, but they also have many unknown risks.

Aims: To explore the feasibility and risks of using EVs secreted by tolerant DCs to intervene in transfusion-related acute lung injury (TRALI).

Methods: DCs derived in vitro were induced into tolerant dendritic cells after 24 h of rapamycin treatment, and could maintain a tolerant state when stimulated by TLR-receptor ligands. By combining LPS with anti-H2Kd antibody, a two-hit animal model was established to induce the onset of TRALI in Balb/c mice. DC cells derived from mouse bone marrow were induced in vitro and treated with rapamycin for 24 h before pre-intervention in the mice, and then the mice were induced to develop TRALI. Observe and record the death of the mice, the wet-to-dry ratio of the lungs, rectal temperature, pleural effusion weight, and lung tissue pathological sections, and so forth, and compare and analyze the incidence of TRALI in the control group and the intervention group. Subsequently, we additionally collected the culture supernatant of C57BL/6 mouse tolerant DC cells (rapamycin treated for 24 h) and harvested EVs by gradient centrifugation. They also tried to use the allogeneic EVs to intervene in TRALI mice, and observed and recorded a series of data on the diseased mice.

Results: Compared with the TRALI onset group, after using rapamycin-treated tolerant DCs to intervene in TRALI onset mice, the mortality rate of the mice was significantly reduced and the weight of pleural effusion in the intervention group was significantly reduced ($p < 0.05$), although there was no statistical difference between lung wet-to-dry ratio and body temperature. Unexpectedly, intervention with EVs generated from allogeneic tolerant DC (after rapamycin treatment) aggravated the mortality of TRALI. Despite EVs intervention, lung wet-to-dry ratio, pleural effusion and body temperature were not significantly different from those in mice treated with LPS and H2Kd antibodies alone. However, the mortality rate after EVs intervention was positively correlated with the injection concentration, even though the allogeneic vesicles were derived from tolerant cells and did not induce TRALI in mice when combined with LPS.

Summary / Conclusions: Previous knowledge was that EVs can well follow the functions of source cells and have low immunogenicity, so they have the safety of allogeneic infusion and broad clinical application prospects. However, our study found that allogeneic extracellular vesicle infusion combined with anti-leukocyte antibodies will increase the risk of death in TRALI, even though syngeneic DC has a protective effect on TRALI after infection and the allogeneic extracellular vesicles combined with LPS does not cause respiratory distress. The data from this study suggest that we need to pay more attention on the risk of TRALI aggravating by the application of allogeneic EVs therapy.

PA19-L05 | A novel animal model of platelet transfusion induced transfusion-associated circulatory overload

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Background: Transfusion-associated circulatory overload (TACO) is a major complication, accounting for 23% of all transfusion-related fatalities. Understanding of potential risk factors remains limited. While previous pre-clinical studies have explored the impact of plasma and red blood cell transfusion in a rat heart failure model, the effect of platelet transfusion remains unexplored. Platelet transfusions, among all blood components, result in the highest overall number of adverse reactions per unit transfused. However, their specific influence on the development of TACO have not been investigated.

Aims: We aim to determine whether platelet transfusion induces circulatory overload compared to crystalloids and to evaluate if it leads to a more severe phenotype than plasma transfusion in a rat heart failure model.

Methods: We utilized a validated TACO model in male anaemic Wistar rats with acute myocardial infarction. Animals were randomized into three groups: platelets ($n = 8$), plasma ($n = 7$), or Ringer's lactate ($n = 7$). We evaluated the development of circulatory overload by measuring pulmonary capillary pressure via left ventricular end-diastolic pressure (LVEDP) with a left ventricular catheter. The primary outcome was the difference between pre- and post-transfusion LVEDP (Δ LVEDP). Secondary outcomes included pulmonary wet/dry weight ratio and hemodynamic measurements. Data are expressed as medians and interquartile ranges.

Results: In our study, pre-transfusion characteristics were comparable across all groups, including: weight, infarct size, LVEDP, mean arterial pressure (MAP), central venous pressure (CVP) and heart rate. The Δ LVEDP following platelet transfusion (10.54 mmHg; 7.30–15.24) was significantly larger than the Δ LVEDP following Ringer's lactate infusion (1.57 mmHg; 0.56–3.07), $p < 0.05$. Δ LVEDP following plasma transfusion (13.19 mmHg; 7.18–17.50) did not significantly differ from Δ LVEDP following platelet transfusion, $p > 0.98$. Platelet transfusion resulted in an increase in MAP and CVP compared to Ringer's lactate ($p < 0.001$ and $p < 0.05$, respectively). Pulmonary wet/dry ratio did not differ among groups ($p > 0.9$). However, the change in P/F ratio was significantly different between platelet transfusion and Ringer's Lactate compared to baseline (resp. -10.3 ± 6.24 and $+12.76 \pm 14.53$; $p < 0.005$).

Summary / Conclusions: Platelet transfusion induces circulatory overload, measured as Δ LVEDP, comparable to plasma transfusion in rats with myocardial infarction, compared to Ringer's lactate. This suggests that platelet transfusion can contribute to TACO development in a manner comparable to plasma transfusion. Whether the same underlying pathophysiology and mechanisms are involved requires further investigation.

Parallel session—donors and donation

Donor health—staying magnetic!

PA20-L01 | Ferritin-guided iron supplementation of blood donors

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Whole blood donations are associated with hemoglobin-bound iron loss and subsequent iron store depletion. Insufficient iron stores may reduce the recovery of hemoglobin levels after donation. Repeat donors therefore risk developing iron deficiency and iron deficiency anemia. Ferritin measurements for iron store monitoring are increasingly applied by blood establishments in order to identify donors that should be deferred from donating or supplemented with iron. Our research over the past years aimed to assess the effectiveness and acceptance of ferritin-guided iron supplementation for whole blood donors. In the randomized controlled FORTE trial, we assessed the effectiveness of different dosages and intake frequencies of iron supplements for donors with ferritin levels ≤ 30 $\mu\text{g/L}$. Acceptance was assessed by investigating knowledge and perceptions of whole blood donors and blood bank personnel on donor iron management in focus groups. The results of these focus groups were then used to set up an international donor survey on donor perceptions regarding iron management, which was distributed through the BEST Collaborative and the European Blood Alliance's working group Donor Studies. Our trial findings suggest that both doses (30 and 60 mg) of iron supplementation, whether taken daily or on alternate days, were effective in increasing ferritin levels and promoting iron store recovery compared to placebo intake. Donors and donor physicians seem generally positive towards iron supplementation, while blood collection staff appear to exhibit more reservations. This presentation will discuss the results of the FORTE trial and associated studies, ultimately guiding donor iron management policy recommendations.

PA20-L02 | Change in hemoglobin to identify a novel threshold for insufficient iron levels—a study in blood donors

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Background: Whole blood donors are at risk of iron deficiency and anemia, therefore blood establishments use a hemoglobin (Hb) test to confirm a donors eligibility for donation. Hb monitoring only does not necessarily prevent donors from developing iron deficiency. An increasing number of blood establishments have therefore started to monitor serum ferritin in addition to Hb levels, to detect low iron stores in blood donors. WHO guidelines indicate that healthy individuals with ferritin levels below 15 ng/mL are considered iron deficient. Nonetheless, clear evidence to support this cut-off is lacking and sufficient levels required for post-donation Hb recovery are currently unknown.

Aims: To define functional decision limits for ferritin based on a quantitative estimation of the association between change in Hb from baseline and ferritin levels in whole blood donors.

Methods: In data from The Netherlands (Sanquin), the U.K. (INTERVAL), South Africa (SANBS), Finland (FinDonor) and the U.S. (Vitalant), we selected donors for whom a baseline Hb was available (aiming to identify Hb levels not affected by prior donation). We then selected donations at which both serum ferritin and Hb were measured, and analysed the association between log base 10 ferritin level and change in Hb from baseline (dHb). As we observed a distinct change in the linear association between the dHb and log10 ferritin levels, we defined a two-segmented line where we assumed both segments to have different slopes, and used maximum likelihood estimation to determine the changepoint. We generated confidence intervals calculating various statistics on 1000 bootstrap samples for subsets containing either male or female donors.

Results: There is a linear association between the drop change in Hb since pre-donation screening and log10 ferritin levels, which changes or disappears when a donor recovers to the pre-donation Hb level. Similar patterns were observed in the level of the ferritin changepoint differs per country and donor subgroup (Table 1).

Summary / Conclusions: The changepoint we observe in the association between ferritin and dHb suggests that in healthy blood donors there is a certain ferritin level below which iron storage levels limit Hb production. Although this association is similar across settings, the ferritin level at this changepoint is not. This might result from differences in donor populations but is more likely due to a lack of a validated commutable international reference material for ferritin. This underlines the need for development and implementation of such a reference material to enable uniform ferritin cutoffs and allow the comparison of ferritin outcomes in different populations. Finally, since a drop in Hb levels is associated with a reduced ferritin level, donor management strategies should be aiming for maintaining the donor's homeostatic Hb level.

PA20-L02 Table 1. Ferritin changepoint at which pre-donation Hb drops per country and population subgroup.

Study population	Ferritin changepoint in ng/mL (95% CI)		
	Men	Premenopausal women	Postmenopausal women
The Netherlands	30.7 (30.1–31.7)	26.3 (24.4–27.5)	24.2 (23.1–25.3)
United Kingdom	41.2 (39.0–45.7)	34.4 (30.1–40.4)	27.0 (25.0–35.9)
South Africa	21.6 (20.7–22.7)	19.3 (18.1–20.2)	18.6 (17.1–20.3)
Finland	24.9 (22.4–33.1)	17.0 (14.1–45.0)	36.9 (30.0–68.1)
United States	29.6 (28.5–30.7)	25.1 (22.4–27.5)	30.6 (28.5–33.3)

PA20-L03 | Iron deficiency and risk of infection in Danish blood donors

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Background: Iron deficiency is the most common mineral deficiency worldwide affecting more than 2.5 billion people. It is especially prevalent among frequent blood donors due to loss of iron through blood donation. Iron stores can be determined by measuring the iron storage protein ferritin and the World Health Organization defines iron deficiency as ferritin < 15 mg/L. Symptoms of iron deficiency include fatigue, difficulty concentrating and restless legs. Furthermore, iron occupies a significant role in adaptive and innate immunity but the association between iron deficiency and infection risk has primarily been investigated in the presence of anemia or other comorbidities in developing countries.

Aims: In this study we aimed to investigate if iron deficiency was associated with infection risk in otherwise healthy blood donors.

Methods: The Danish Blood Donor Study (DBDS) is a national ongoing prospective study of blood donors, initiated in 2010. In four out of five Danish regions, ferritin is measured routinely in female first-time donors and donors with declining hemoglobin levels. Additionally, ferritin is measured in Central Denmark Region and Zealand Region upon DBDS inclusion and in the Capital Region at every 10th donation. From DBDS, we included 86,466 donors with 316,812 ferritin measurements in the period March 2010 to October 2022. In the main analyses, we performed multivariate cox regression with robust standard errors to estimate the risk of infection for iron deficient donors compared with iron replete donors in a three-month follow-up period. Age was used as the underlying timescale. Infection was defined as a filled prescription of antimicrobials registered in the Danish National Prescription Registry or a hospital contact with an ICD-10 code for infection registered in the Danish National Patient Registry. All analyses were stratified by gender and adjusted for donation intensity, health region, BMI, and smoking.

Results: During 72,282 person-years at risk, 16,329 prescriptions for antimicrobials were filled. Iron deficiency (ferritin < 15mg/L) was associated with an increased overall risk of infection for women (HR 1.08, 95% CI 1.01-1.15) in the adjusted model. The results were similar for six- and twelve-months follow-up time. In sub analyses for women, iron deficiency was associated with an increased risk of respiratory tract infections (HR 1.14, 95% CI 1.03-1.26) and urinary tract infections (HR 1.16, 95% CI 1.04-1.29). Iron deficiency was not associated with overall infection risk for men (HR 0.99, 95% CI 0.79-1.24). During 74,039 person-years at risk 2,008 hospital contacts for infection were registered. For both men and women, we found no association between iron deficiency and hospital contacts for infection in the adjusted models.

Summary / Conclusions: In female blood donors, iron deficiency was associated with an increased risk of infection measured as filled prescriptions of antimicrobials. However, effect sizes were small and there was no association between iron deficiency and hospital contacts for infection. For male blood donors, there was no association between iron deficiency and infection risk which could be owing to less statistical power or to sex-specific differences in iron metabolism. Consequently, risk of infection should not be considered an apprehension with regard to blood donation. However, these findings support the role of iron as a contributor to immune function and support monitoring of iron stores in female blood donors.

PA20-L04 | Retention of female blood donors with low haemoglobin or iron deficiency for plateletpheresis

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Background: According to European Standards, minimal required haemoglobin (Hb) levels for platelet (PLT) apheresis are 125 g/L for females and 135 g/L for males, but individual donations may be performed with Hb below these levels (EDQM, 21st Edition). The Swiss regulations set the lower acceptable Hb limit for PLT apheresis at ≥ 110 g/L and ≥ 120 g/L for females and males, respectively. Hereby we present longitudinal data of female donors who were not eligible for whole blood (WB) donation due to low Hb or low iron stores but could be accepted for PLT apheresis.

PA20-L04 Table 1

	No.	Hb / p-value	No.	ferritin / p-value
First WB donation	85	134 g/l	54	22.9 ng/mL
Switch	85	123 g/L / 0.0000	54	8.5 ng/mL / 0.0000
Last PLT Apheresis	85	130 g/L / 0.0000	49	13.7ng/mL / 0.0000

Aims: To analyse the course of Hb and ferritin over repetitive PLT apheresis in female donors not qualifying for WB donation because of low Hb or iron deficiency.

Methods: We retrospectively analysed data of women with Hb < 125 g/L and/ or ferritin levels \leq 15 ng/mL who switched from WB donation to PLT apheresis and underwent at least two PLT donations and one PLT apheresis/year. The following data were evaluated: donor age; Hb and ferritin at the first WB donation and at first and last PLT donation; number of PLT apheresis performed; reasons for temporary and definitive deferrals. Hb was measured from a venous sample at each PLT donation with the Sysmex K-4500 or the Sysmex 500 (Sysmex Digitana AG, Horgen, Switzerland). Ferritin was measured at least twice/year with a CMLA method with the Architect ci8200 or with the Alinity (Abbott Laboratories, IL, USA). Data were analysed with descriptive statistics. For paired samples the Wilcoxon signed-rank test and two-sided sign test were used. Comparisons were made for the first WB donation versus switch to PLT apheresis and switch versus last PLT apheresis.

Results: We collected data of 85 females who changed from WB donation to PLT apheresis between January 1997 and December 2023. The median number of WB donation before the switch was 3 (1-33). The reason for changing to PLT apheresis was Hb < 125 g/L and ferritin \leq 15 ng/mL in 54 (64 %) and low ferritin alone in 31 (36 %). Median age (range) was 30 years (18-53 years) at the first WB donation, 33 years (18-57 years) at switch, and 45 years (19-68 years) at the last PLT donation. Median Hb and ferritin values at the three time points are reported in the Table.

In the evaluated period, the total number of PLT apheresis performed was 2362 with a median of 10 (range 1-155) and a mean of 3.3 / year per donor. As to December 2023, 65 (76%) women of the cohort were still donating actively. Twelve (14%) were temporary deferred for PLT apheresis because of anaemia ($n = 3$), pregnancy ($n = 3$), donor wish ($n = 4$), low PLT value ($n = 1$) and repeated unspecific HCV screening ($n = 1$). Eight women were permanently excluded from blood donation because of donor decision ($n = 4$), arrhythmia ($n = 1$), anaemia ($n = 1$), cardiac surgery ($n = 1$) and severe adverse event ($n = 1$).

Summary / Conclusions: Female blood donors with iron deficiency with or without low Hb who do not qualify for WB donation can be retained for PLT apheresis in compliance with the Swiss regulations. Regular PLT donation in otherwise healthy women with low Hb and ferritin is not associated with a risk for anaemia or a further decrease

iron stores and contributes to ensuring a sufficient PLT supply in the context of increasing demands for PLT components and the shrinking of the blood donor population

PA20-L05 | Results of the FORTE study; a randomized controlled trial on the optimal iron supplementation protocol for iron deficient whole blood donors

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Background: Frequent whole blood donors have an increased risk of developing iron deficiency, and subsequently anaemia, causing detrimental health effects when left untreated. Donation intervals are often too short for iron store recovery and extending intervals reduces donor availability. Oral iron supplementation is known to shorten iron store recovery time and could serve as an alternative to extended donation intervals. However, there is no consensus on the optimal protocol in terms of dosage and intake frequency.

Aims: We investigated the effectiveness of iron supplementation on post-donation iron store (i.e., ferritin) recovery by evaluating the impact of four different iron supplementation protocols varying in dosage and intake frequency, compared to placebo supplementation.

Methods: Whole blood donors whose ferritin levels were measured during their upcoming donation were invited to participate. Donors with ferritin levels ≤ 30 μ g/L were included in a double-blind, randomized controlled trial. Participants were randomly allocated to one of six arms, with an equal distribution of sex and age groups (i.e. stratified according to 50 years of age). Depending on the study arm, donors were asked to adhere to a supplementation protocol for 56 days, taking capsules containing 0 mg (i.e. placebo), 30 mg or 60 mg of iron, either on alternate days or daily. Logistic regression was used to estimate the odds of recovery (i.e. ferritin levels > 30 μ g/L) after 56 days in all arms, compared to the daily placebo supplementation arm.

Results: A total of 834 donors, of whom 369 were male and 465 female, with a mean age of 47.0 ± 15.7 and 38.7 ± 14.2 years, respectively, participated in the trial. Baseline characteristics in terms of age, BMI, Hb-, and ferritin levels were similar across all supplementation groups. After 56 days, median ferritin levels in the 30 mg iron supplementation groups, for both daily (26 $\mu\text{g/L}$ [11.5, 40.5]) and alternate day intake (34 $\mu\text{g/L}$ [15.0, 53.0]), and the 60 mg iron supplementation groups (29 $\mu\text{g/L}$ [16.2, 41.8], 35.5 $\mu\text{g/L}$ [18.0, 53.0], respectively) were higher compared to both placebo groups (15 $\mu\text{g/L}$ [5, 25], 14 $\mu\text{g/L}$ [4.8, 23.3], respectively). The daily and alternate day 30 mg iron supplementation groups ((10.6, CI: 5.4 – 22.5), (3.2, CI: 1.6 – 6.8)), and 60 mg iron supplementation groups ((24.6, CI: 12.2 – 53.4), (6.6, CI: 3.4 – 14.1)) had a significantly higher odds of iron store recovery compared to the daily placebo intake group. Similar effects were observed for continuous ferritin and haemoglobin levels.

Summary / Conclusions: This study shows that iron supplementation effectively improves iron store recovery in iron depleted whole blood donors. The effects are dose and frequency dependent, with a higher dose and frequency leading to increased odds of recovery. Further analyses are needed to evaluate effects on health outcomes and gastrointestinal side effects.

Parallel session—blood safety

Transfusion associated agents—new findings to bug you

PA21-L01 | Review of evidence for transfusion transmitted cerebral amyloid angiopathy

G Edgren

Abstract not available

PA21-L02 | The efflux pump *norB* gene is involved in increased resistance to quinolones and enhanced virulence of *Staphylococcus aureus* grown in platelet concentrates

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Background: *Staphylococcus aureus* is a common contaminant of platelet concentrates (PCs) that poses a safety threat to transfusion patients. Transcriptome data obtained in the Ramirez laboratory have shown that the PC storage environment heightens the expression of antibiotic resistance genes in transfusion relevant *S. aureus* isolates.

PA21-L02 Table 1. MBC ($\mu\text{g/mL}$)

Strain	Ciprofloxacin		Norfloxacin	
	TSB	PCs	TSB	PCs
RN6390	0.5-2.0	16	1.0-2.0	16-64
RN6390 Δ <i>norB</i>	0.5	2	0.5-1.0	2-16
RN6390 Δ <i>mgrA</i>	0.5-1.0	16	1.0-4.0	16-64

One of these genes is *norB*, which encodes for an efflux pump implicated in quinolone resistance and is negatively regulated by *MgrA*. *NorB* has also been shown to enable *S. aureus* survival and quinolone resistance in a mouse subcutaneous abscess model. We were therefore intrigued to investigate if *NorB* has a role in antibiotic resistance and virulence in *S. aureus* grown in the immune challenging PC storage milieu.

Aims: To elucidate the role of the efflux pump *norB* gene in antibiotic resistance and virulence of *S. aureus* grown in PCs.

Methods: Wild type *S. aureus* RN6390 and two derivative deletion mutants, RN6390 Δ *norB* and RN6390 Δ *mgrA*, were used in this study. Differential expression analysis of *norB* in RN6390 and RN6390 Δ *mgrA* grown in trypticase soy broth (TSB) and PCs were performed using RT-qPCR. Minimal Bactericidal Concentration (MBC) assays were performed in TSB (37°C/static/24 h) and PCs (22°C/agitation/24 h) to compare the resistance of the three isolates to the quinolones ciprofloxacin and norfloxacin ($n = 6$). Furthermore, the virulence of these strains was tested in a *Bombyx mori* (silkworm) model. The lethal dose 50% (LD_{50}) was determined using 10-fold serial dilutions of bacteria in insect saline (0.6% NaCl) ($n = 3$).

Results: RT-qPCR confirmed upregulation of *norB* in the *mgrA* mutant (2.12-fold). Furthermore, RT-qPCR data demonstrated that *norB* in RN6390 was upregulated in PCs compared to TSB (8.43-fold). MBCs for ciprofloxacin and norfloxacin in wild type RN6390 and the *norB* and *mgrA* mutants grown in TSB and PCs are shown in Table 1. Interestingly, antibiotic resistance was unchanged between wild type RN6390 and RN6390 Δ *norB* grown in TSB but decreased 8-fold in the mutant compared to the wild type strain when they were cultured in PCs. No differences in resistance between RN6390 and RN6390 Δ *mgrA* were observed in TSB or PCs. Furthermore, virulence assays in silkworms showed LD_{50} values of $\sim 1.02 \times 10^4$ ($\pm 1.01 \times 10^4$) CFU/larvae for RN6390, $\sim 3.29 \times 10^6$ ($\pm 2.04 \times 10^6$) CFU/larvae for RN6390 Δ *norB*, and $\sim 2.85 \times 10^5$ ($\pm 1.90 \times 10^5$) CFU/larvae for RN6390 Δ *mgrA* indicating loss of virulence (higher LD_{50}) in RN6390 Δ *norB* compared to wild type RN6390.

Summary / Conclusions: The PC storage environment triggers upregulation of *S. aureus* *norB*, which is negatively regulated by *MgrA*. Functional assays demonstrated that *NorB* is linked to increased resistance to ciprofloxacin and norfloxacin when *S. aureus* is grown in PCs compared to TSB. Interestingly, our data also showed a role of *NorB* in enhanced virulence in a silkworm model. Unexpectedly, increased expression of *norB* in the *mgrA* mutant did not result in increased quinolone resistance or enhanced virulence in silkworms compared to

the wildtype strain. These results indicate that NorB is part of a complex regulatory machinery involved in quinolone resistance and virulence modulation of *S. aureus* in the PC storage milieu, which merits further investigation and should be considered for medical treatment of patients receiving PCs contaminated with this bacterium.

PA21-L03 | Transfusion-transmitted bacterial infection with *Bacillus mobilis* in a pathogen-reduced platelet concentrate in France—a case-report

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Background: Transfusion-transmitted bacterial infections (TTBI) related to platelet concentrates (PC) have become exceptional in France since the generalized implementation of pathogen reduction treatment (PRT) with amotosalen/UVA in 2017. We report here a TTBI with a PRT-PC contaminated with *Bacillus mobilis*. This species, belonging to the *Bacillus cereus sensu lato* group, is able of sporulation and filamentous surface motility under a large range of environmental conditions. TTBI or transfusion near-misses with *B. cereus sensu lato* group in PRT-PC have been reported before, but this is the first case specifically reported with *B. mobilis*.

Aims: To share with the scientific community that *B. mobilis* escapes the efficacy spectrum of amotosalen/ UVA PRT.

Methods: The PC was produced in November 2023 by Etablissement Français du Sang in Nancy, France. On Day 1 after whole blood donation, 8 buffy-coats were pooled and pathogen-reduced, using an amotosalen/UVA dual-storage kit. After overnight adsorption of residual amotosalen, the pool was divided in 2 twin storage bags on Day 2. One sub-units issuing was prevented at Day 4 because of a negative swirling. Quality control later identified its contamination with *B. mobilis*. Meanwhile, the other sub-unit had been issued on Day 3 with satisfactory swirling, and subsequently transfused. The recipient presented with isolated fever 24 h after transfusion, leading to identification of *B. mobilis* in an haemoculture. With adapted antibiotherapy, the patient fully recovered from sepsis. Whole genome sequencing performed on both *B. mobilis* strains at Institut Pasteur, Paris, showed perfect sequence identity. We therefore conclude to TTBI in this case.

Results: Donors were free of infectious symptoms at least 14 days before and after donation. Donors' skin was disinfected in 2 steps with alcoholic iodine povidone, and free of *B. mobilis* (negative elbow crease swabs within 2 weeks of the TTBI event). No *B. mobilis* were isolated in cultures performed on qualification samples, red blood cells and frozen plasma units originating from the 8 donations. No malfunction was observed concerning *in-process* sterile connections and weldings. Quality control data showed no failure in PRT process' completion. The adsorption bag was contaminated with a *B. mobilis* strain genetically

distinct from the one isolated in the patient and the non-swirling PC subunit. Association of a short interval between buffy-coat pooling and PRT (<3 h), and the presence of bacteria in the adsorption bag, is in favour of 1/ an upstream contamination, before PRT and 2/ *B. mobilis*' resistance to PRT. As the pooling bag and collection bags were destroyed, we cannot ascertain whether contamination occurred during donation, or during buffy-coat separation/pooling.

Summary / Conclusions: This case report emphasizes the paramount necessity to maintain, despite PRT, disinfection protocols at phlebotomy, *in-process* integrity controls, and swirling control before PC issuing—the latter having prevented here a second TTBI. Blood banks should also perform research on the *B. cereus sensu lato* bacteria, as gathered evidence grows about their resistance to available PRT.

PA21-L04 | Can next generation sequencing help to assess the residual risk of HBV transfusion transmission?

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Background: The risk of hepatitis B virus (HBV) transfusion-transmission depends on technical, host, and viral factors including the effectiveness of screening tests in detecting viral variants, and the balance between functional and defective viral genomes in the blood of infected donors. Next-generation sequencing (NGS) offers the opportunity to directly explore the genetic of complex viral populations in infected individuals despite limited sensitivity and the need of complex bioinformatics analysis pipelines.

Aims: Two NGS procedures were evaluated to characterize the quasispecies complexity viral variants in the S gene and the ratio of functional to defective HBV genomes in donors with occult (OBI) or overt (HBsAg+) infection.

Methods: PreS/S region and whole HBV genome were PCR-amplified from 8 OBI and 18 HBsAg+ donors after viral particle concentration. HBsAg+ donors were selected to carry S mutations previously associated with immune escape. PreS/S and whole genome amplicons were sequenced on the MiSeq sequencer (Illumina) and the MinION (Oxford Nanopore Technologies) NGS systems, respectively. S amino acid sequences obtained with the two assays were analyzed and quasispecies distribution compared. Residues present in less than 5% of sequence reads were not considered in the analysis. MinION long-read single-molecule sequences were assessed for the presence of major deletions.

Results: Median HBV DNA load was 10 (range: <6-431) IU/mL in OBI and 1.09×10^5 (range: $10.1.1 \times 10^8$) IU/mL in HBsAg+ donors. HBV strains were of genotypes A3, B, D, and E in OBI, and genotypes A1-E and C/D in HBsAg+ donors. S aa consensus sequences obtained with MiSeq and MinION showed 98% to 100% identity, regardless of HBV status. A significant higher frequency of polymorphic sites was observed with MinION than MiSeq ($p = 0.003$), but no difference was observed

between OBI and HBsAg+ donors. Discrepant results between assays were mainly associated with minor variants constituting <10% of the quasispecies population within a sample. Amino acid polymorphism was observed at 20 of 22 positions previously reported associated with immune escape. NGS detected wild-type residues at these critical sites as minor species in 3 (37.5%) OBI and 8 (44.4%) HBsAg+ samples. Wild-type residues accounted for 5%–40% and 14%–43% of total sequences in OBI and HBsAg+ samples, respectively. Stop codons in S gene were detected in 5% to 79% of quasispecies from 12 HBsAg+ samples. Two HBsAg+ samples had a stop codon in 100% of their sequences. MinION whole genome sequence reads (~3,200 nt) were obtained for 8 OBI and 11 HBsAg+ samples. Complete genomes accounted for 97%–99% of reads obtained from 7 OBIs, but only 22% of the reads obtained in one OBI, with the remaining 78% showing a 2572 nt deletion. Four (36.4%) HBsAg+ samples had spliced HBV genomes representing 7.5% to 95% of the detected sequences.

Summary / Conclusions: MinION detected significantly more HBV minor species than MiSeq. Similar levels of virion-associated S quasispecies were detected with NGS in the plasmas of donors with occult and overt HBV infection. Donors infected predominantly with HBV immune escape S variants may also contained wild-type viral strains as minor species retaining the ability to produce limited amount of detectable HBsAg in the blood. The presence of a substantial proportion (7.5%–100%) of virion-associated HBV genomes carrying large deletions or a defective S gene in donor blood may be a transfusion-associated infectious risk mitigating factor to consider.

PA21-L05 | Parvo virus B19 outbreak among blood donors in Switzerland

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Background: Human Parvovirus B19 (PVB19) is a common human pathogen and is mostly a childhood infection. It is mainly transmitted by respiratory secretions but can also be transmitted via plasma derivatives and blood. In the Swiss blood donation organization, PVB19 DNA diagnostics is not a mandatory, release-relevant test. However, due to the specified quality criteria for fractionated plasma from the plasma fractionating industry, it is carried out on all blood donations. Positive PVB19 DNA results are handled in accordance with the Blood Transfusion SRC Switzerland (BTS SRC) regulations as post-donation information and the donations concerned are destroyed. If products have already been delivered, the affected customers will be informed and asked to either destroy or return the products. Pre-donations from the last 12 weeks will also be destroyed or recalled.

Aims: Minor outbreaks of PVB19 occur about every 3 to 4 years. In the course of 2023 increasing positive cases in Switzerland were observed compared to the previous years. The surveyed notified cases of the 11 Blood Transfusion Services in Switzerland are described.

Methods: Donors were tested in pools of ≤ 96 samples in a duplex format with HAV either with the Cobas DPx (Roche Diagnostics) or the Procleix Parvo/HA (Grifols) assay. Positive pools (PVB19 DNA titer of $\geq 10^2$ IU/ml or $\geq 10^3$ IU/ml, depending on the specific release criteria selected by the regional blood transfusion services) were resolved by pool deconstruction and afterwards single sample testing and confirmation will be performed. In the individual test, a titer of $\geq 10^4$ IU/ml in the single donation is considered positive. All positive B19 donations are reported to Swissmedic and BTS SRC using a defined form (hemovigilance notification protective measure).

Results: In 2023, 73 positive B19 cases were recorded via the hemovigilance notification. Titers of the positive individual samples, if available, range from 1.03×10^4 IU/ml up to 3.29×10^{13} IU/ml. PVB19 was detected in both first-time donors and repeat donors. Both genders were affected, and the age of donors ranged from 20 to 63 years. Most of the donors who could be interviewed stated that they had not experienced any symptoms. Only a few reported mild symptoms. The cases of the last 6 years are listed in comparison to the above figures: 2017: 31 cases, 2018: 9 cases, 2019: 12 cases, 2020: 21 cases, 2021: 0 cases, and 2022: 1 case.

Summary / Conclusions: The positive PVB19 cases recorded in 2023 increased over the course of the year and most cases were detected in the second half. Compared to previous years, the beginning of a new PVB19 outbreak could be observed in 2023 and continues even intensified until 2024. The fact that this follows more than 3–4 years after the mild outbreak in 2017 can certainly be attributed to the Covid 19 pandemic measures (masks, hand disinfection). The incipient outbreak in 2020, which started within the usual 3–4 years, was thus abruptly interrupted, and resulted in no or practically no cases being detected in 2021 and 2022. In view of the large number of positive blood donors, some of whom have very high PVB19 DNA titers in 2023, PVB19 testing contributes to greater safety of blood products, as customers of such products that have already been delivered, are informed, and can destroy the product.

Parallel session—blood products

Hemostasis for pediatric patients and characterizing new approaches for storing platelet concentrates

PA22-L01 | Management of pediatric trauma associated bleeding & potential role of intravenous hemostatic agents/adjuncts

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Outcomes for children with traumatic injury and life threatening bleeding are worse than in adults. This may be due to delayed recognition and treatment of hemorrhagic shock. Hemostatic Resuscitation describes a balanced blood based strategy for treating severe bleeding with the goal of improving outcomes. Recently there is data that supports the use of whole blood, increased use of plasma and platelets relative to RBCs, and the use of tranexamic acid in children with life threatening hemorrhage. There is a large multicenter definitive platform trial that has been funded to examine if whole blood compared to blood components and tranexamic acid compared to placebo in 1000 children with life threatening bleeding from traumatic injury. This presentation will review the epidemiology, practice patterns, and outcomes for children with severe bleeding from traumatic injury.

PA22-L02 | Reducing ROS production and protecting from clearance of cold-stored platelets by additional NAC

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Background: It is known the rapid clearance of refrigerated platelets was attributed to various cold storage lesions, including abnormal increase of reactive oxygen species (ROS), when platelets are exposed to the cold temperature. As an antioxidant, N-acetylcysteine (NAC) shows the prominent effect on scavenging a variety of reactive oxygen species and inhibiting cell damage and apoptosis. It was demonstrated that the addition of NAC significantly increased the in vitro viability of refrigerated platelets.

Aims: This study intended to select the appropriate concentration of NAC and explored the effects of NAC on reducing ROS production and protecting the refrigerated platelet from being recognized and cleaned by macrophages and hepatocytes.

Methods: Platelets Concentrates were divided into experimental group (N), cold storage group (4C) and room temperature group (22C), and stored at 4°C or 22°C for 5 days. The experimental groups were supplemented with 1 mM (N1), 5mM (N5) and 25 mM (N25) of NAC

into 4°C-stored PLT concentrates, respectively. Platelet ROS and other characteristics were detected by flow cytometry. Platelet phagocytosis was detected by PMA activated THP-1 cells or by primary cultured HepG2 cells.

Results: After 5 days of storage, ROS was increased significantly in group 4C than that in group 22C, and was reduced in group N5 significantly. Compared with 4C group, CD62P expression, PS exposure and β -GlcNAc expression in N5 group were significantly decreased. The platelet phagocytosis of THP-1 and HepG2 in N5 group were also significantly lower than those in 4C group. However, the addition of 5mM NAC did not significantly affect the count, PH, CD42b expression, β -Gal expression and coagulation function of cold-stored platelets for 5 days.

Summary / Conclusions: The addition of NAC to refrigerated platelets could significantly reduce the level of platelet ROS, and significantly reduce their recognition and phagocytosis by cells, suggesting that it may protect from the clearance of refrigerated platelets after transfusion.

PA22-L03 | The hemostatic function of platelets stored in hypoxic whole blood is non-inferior compared to platelets stored in conventionally stored whole blood

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Background: Hemorrhage after trauma accounts for more than 1.9 million deaths per year worldwide. Recently, whole blood was re-introduced for resuscitation from trauma-induced acute bleeding. However, oxidative stress during storage reduces the quality of red blood cells (RBC) in whole blood. To mitigate this problem, Hemanext Inc. developed a hypoxic RBC (H-RBC) storage system. H-RBC improve outcomes in rats undergoing a hemorrhagic shock model. It is unclear how hypoxic storage affects the hemostatic function of platelets (PLT) in whole blood. We addressed this question in a mouse model of hemostasis.

Aims: To compare the hemostatic function of PLT stored in H-WB to PLT in conventionally stored WB (C-WB).

Methods: CPDA-1-anticoagulated wild-type (WT) mouse whole blood was deoxygenated in a procedure closely mimicking the licensed human system (H-WB). WB undergoing a mock deoxygenation procedure served as control (C-WB). H-WB and C-WB were stored for 10 days at 1-6°C. We measured the PLT count by blood analyzer and the pH, pO₂, and pCO₂ by blood gas analyzer. We compared the ability of deoxygenated PLT in WB to achieve hemostasis in thrombocytopenic human interleukin 4 receptor-transgenic (hIL4R-TG) mice. These mice express the human IL4 receptor and were rendered thrombocytopenic by an anti-human IL4 receptor antibody. While clearing all endogenous hIL4R-positive PLT, this antibody leaves any transfused WT platelets unaffected. Whole blood was transfused, and the tail of depleted

hIL4R-TG was cut. Blood from the severed tail was collected in saline, and the experiment was terminated after 10 min or once the bleeding stopped. We compared the results to mice receiving fresh WB (F-WB). The blood loss was measured by a plate reader. Transfusions were administered to four mice in the H-WB and C-WB groups, and six received F-WB transfusions. We compared the blood loss to positive (not platelet-depleted) and negative (platelet-depleted) control groups that did not receive transfusions.

Results: PLT counts in the units did not differ significantly between H-WB and C-WB at any time. After deoxygenation, the O₂ levels in the H-WB decreased significantly (90 ± 7 to 34 ± 20 mmHg) compared to the significant increase in O₂ levels seen in C-WB (92 ± 4 to 161 ± 26 mmHg; $p = 0.003$). This difference persisted after 10 days of storage (7 ± 5 vs. 45 ± 6 mmHg, $p = 0.02$). No significant differences were observed for CO₂ and pH levels. The PLT counts in recipient animals of H-WB and C-WB did not increase significantly (13 ± 2 to 18 ± 3 × 10³/μL, $p = 0.5$, versus 15 ± 3 to 21 ± 2 × 10³/μL, $p = 0.26$). In contrast, the F-WB PLT count increased significantly (16 ± 5 to 71 ± 15 × 10³/μL, $p < 0.0001$). The average blood loss between H-WB and C-WB was not significantly different (102 vs. 100 μL, $p > 0.9$). The F-WB group lost less blood than the stored groups, albeit without significance. The negative control group lost more blood than F-WB (180 ± 88 vs. 25 ± 9 μL, $p = 0.04$). None of the mice in the H-, C-, and no-WB group stopped bleeding within 10 min. The mice in the F-WB had an average bleeding time of 7.1 min, which did not differ significantly from H-WB and C-WB. The mice in the positive control group had an average bleeding time of 4.1 min, significantly less than H-WB ($p = 0.02$) and C-WB ($p = 0.02$) and not significantly different from F-WB.

Summary / Conclusions: PLT in H-WB and stored under O₂-reduced conditions show clinically normal hemostasis in an *in vivo* mouse model.

PA22-L04 | A deep eutectic solvent is an effective cryoprotective agent for platelets

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Background: Cryopreservation is a long-term storage option, which can extend the shelf-life of platelets from days to years. The most widely used method of platelet cryopreservation uses dimethylsulphoxide (DMSO) as a cryoprotective agent (CPA), pelleting the platelets by centrifugation to remove the majority of the DMSO-containing supernatant prior to freezing, and storage at −80°C. Strategies to simplify the current cryopreservation process may lead to more widespread use of this storage technique. Deep eutectic solvents (DESs) are an emerging class of CPA, prepared by mixing hydrogen bond donors and acceptors to achieve a melting point lower than the individual components. A DES composed of Proline-Glycerol (Pro-Gly; at a molar ratio of 1:3) reportedly has similar cryoprotective efficacy to

PA22-L04 Table 1

Parameter	DES	no CPA	DMSO	p value
Recovery (%)	92 ± 16	101 ± 17	94 ± 14	0.6197
CD42b (MFI)	259 ± 20	182 ± 16*	186 ± 19*	<0.001
GPVI (MFI)	297 ± 70	108 ± 12*	208 ± 44*	<0.001
Annexin V (% positive)	81 ± 3	96 ± 1*	64 ± 8*	<0.001
CD62P (% positive)	60 ± 4	26 ± 5*	28 ± 7*	<0.001
TEG R-time (minutes)	5.2 ± 0.3	5.5 ± 0.4	5.7 ± 0.5	0.1648
TEG Maximum amplitude (mm)	50 ± 2	32 ± 2*	57 ± 3*	<0.001

* $p < 0.05$ compared to DES.

DMSO in selected mammalian cells, without toxicity, but has not been assessed for freezing platelets.

Aims: To determine the effectiveness of Pro-Gly DES as a CPA for platelets.

Methods: DES platelets ($n = 6$) were cryopreserved using 10% Pro-Gly 1:3, frozen at −80°C without centrifugation or removal of the cryoprotectant, then thawed for testing without further manipulation. Control platelets were frozen and thawed as for the DES group, but without the addition of a cryoprotectant (no CPA; $n = 6$). Platelets were also cryopreserved according to the gold-standard procedures (DMSO; $n = 6$) using 5.5% DMSO, followed by hyperconcentration to 25 mL by centrifugation to remove extracellular DMSO, and freezing at −80°C. DMSO platelets were thawed and resuspended in plasma prior to testing. Platelet quality was assessed by flow cytometry and thromboelastography (TEG).

Results: Post-thaw platelet recovery was similar between the three groups (Table 1). The abundance of labile platelet glycoproteins GPIbα (CD42b) and GPVI was higher in the DES group, compared to the DMSO and no CPA groups (Table 1). However, markers of activation (CD62P and annexin-V) were also higher in the DES platelets, compared to the DMSO group (Table 1). In terms of platelet function, the time to clot formation (R-time) was similar between the three groups. While DES improved the maximum amplitude of the clot compared to no CPA, it was lower than the DMSO platelets (Table 1).

Summary / Conclusions: DES provides a cryoprotective advantage to platelets frozen at −80°C when compared to no CPA. Importantly, when compared to the current 'gold-standard' DMSO method, most quality parameters were similar in platelets frozen with DES. The major advantage with using DES as a CPA is that it is composed of biocompatible compounds that do not need to be removed after thawing. This greatly simplifies the freezing and thawing process and avoids the toxic effects of DMSO.

PA22-L05 | Stability of dengue, zika, and chikungunya viruses in platelets stored under standard blood bank conditionsB A Cáceres¹, A Urbina², A Rodríguez³, E Calvo¹, F Delgado¹, J Castellanos¹¹Instituto de Virología, Universidad El Bosque, ²Fundación Universitaria Sanitas, ³Banco Nacional de Sangre, Cruz Roja Colombiana, Bogotá D.C, Colombia

Background: While dengue (DENV), Zika (ZIKV), and chikungunya (CHIKV) arboviruses are primarily transmitted through vector bites, high rates of asymptomatic infections and the development of viremias are factors that could potentially contribute to the transmission of these viruses through blood transfusions. Some reports have shown evidence of the presence of arboviral RNA in serum samples from blood donors and the transmission of DENV through platelet transfusion. However, it remains uncertain whether these arboviruses present in blood components remain stable when stored under standard blood bank conditions.

Aims: This study aimed to assess the stability of DENV, ZIKV, and CHIKV viruses in platelets stored under standard blood bank conditions.

Methods: Platelets obtained by apheresis from a volunteer donor were divided into T-12.5 culture flasks (three mL per flask) and infected with known amounts of DENV (1.6×10^4), ZIKV (4.6×10^3), and CHIKV (2.5×10^3) viruses. The infected platelets were then stored at 20–25°C under agitation. Over seven days (from day zero to day six), three culture flasks containing platelets infected with each virus were collected daily. The collected samples were used to detect and quantify the viral genome using RT-qPCR. Additionally, CHIKV-infected platelet samples were used to infect Vero cell monolayers, and the infection of these cells was identified by indirect immunofluorescence assay (IFI) and viral plaque-forming assays. Differences between the genomic copy number of platelets infected with the three viruses obtained on day zero and the other six days of observation were compared using Dunn's test ($p < 0.05$, Stata v16).

Results: During the initial five days of monitoring (from day zero to day four), the infected platelets exhibited a significant decrease in the initial number of copies of DENV (1.31×10^7 copies/ μ L), ZIKV (4.2×10^4 copies/ μ L), and CHIKV (1.1×10^4 copies/ μ L). On day four, the lowest genomic copy numbers of DENV (6.08×10^5 ; $p = 0.0001$) ZIKV (2.78×10^3 ; $p = 0.0001$), and CHIKV (5.42×10^2 ; $p = 0.0001$) of all the observation days were recorded. However, during the final two days of monitoring, an increase in the number of genomic copies of the three viruses was observed. Furthermore, the E protein of CHIKV was detected by IFI in infected monolayers using platelet aliquots from the seven days of observation, with no significant changes in the viral titer obtained (PFU/mL).

Summary / Conclusions: In conclusion, our findings demonstrate that the viral genome of DENV, ZIKV and CHIKV remains stable in platelets stored under standard blood bank conditions for up to seven days. In addition, we have observed that the infectious capacity of CHIKV is also maintained under these conditions.

Parallel session—management and organisation**Are we ready for anything?****PA23-L01 | Emergency preparedness**C S Cohn^{1,2}¹Laboratory Medicine and Pathology, University of Minnesota, Minneapolis, ²Chief Medical Officer, AABB, Bethesda, United States

Natural and human-made disasters may cause a surge in demand for blood components. Anticipating and preparing for this surge should be a part of emergency planning protocols. Planning should cover all aspects of blood manufacturing, including the number of donors that may be needed, phlebotomists and other staff required to process the blood collections, the number of available blood component bags, needles, etc. An analysis of each step in the process may reveal bottlenecks or other points of weakness that should be addressed. Addressing the weaknesses may require innovative ideas and/or new legislation so that changes can be made.

PA23-L02 | Suddenly, everything falls apartC Sanz¹, A García-Carulla¹, C Ruiz², M Altamirano¹, M Clarés¹, M Esteva¹, M Gomez¹, J Hernández², C López², G Hernández³, A C Pedraza¹¹Blood Bank (BST-Clinic), ²Information Systems (Centre diagnòstic biomèdic), Hospital Clinic, ³IT health consultant, GPI Iberia, Barcelona, Spain

Background: On March 5th, 2023 the Hospital Clinic of Barcelona was cyberattacked with a sophisticated ransomware that crippled all the applications and servers of the hospital.

Aims: Describe the impact of such a breakdown on the Transfusion Service operations, how we handled the situation, and the insights we gained.

Methods: Retrospective analysis of the prospectively collected daily records of all the incidents and how they were dealt with.

Results: Since the electronic transfusion request was down, a new system based on manually filled forms was urgently needed. Many requests were incorrectly filled out, so an information leaflet addressed to prescribing physicians had to be distributed to all wards and operating rooms. We had a local backup of the BBLIS (Blood Bank Laboratory Information System), updated every 24 h, but it could not be accessed immediately because the software license was not configured for local use. This license problem could not be fixed until the 4th day after the cyberattack. The electronic crossmatch could no longer be performed because connections between the immunohematology analysers and the BBLIS worked through the disabled cloud (instead of local wiring). It had to be replaced with immediate spin crossmatching, which resulted

in a rapid stockout of glass tubes. Temperature control of all refrigerators, freezers and platelet incubators worked through the now crippled network, so a protocol for manual recording and control should be instituted. Because of the crippled communication between the BBLIS and the hospital information system (HIS), all the transfusion documentation, including reports of transfusion completion, should have been managed and recorded manually. Transfusion labels with the recipient's identification had to be performed by hand until some printers were locally installed and connected to the local backup BBLISS. We need to establish an independent internet connection with the regional blood bank to incorporate into the BBLISS the data files of the supplied blood components. The increased manual workloads and the need to ensure procedural safety required assigning extra staff. It is worth noting that a full-time person was needed to answer phone calls about the availability of crossmatched blood for patients, information previously displayed at the bedside through the electronic medical record.

Summary / Conclusions: Information technologies provide transfusion services with a significant increase in efficiency and safety. Nevertheless, they are intrinsically fallible so all the possible impacts derived from their breakdown should be considered as well as the procedures to deal with them.

PA23-L03 | Mass casualty simulation—testing blood supply management

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Background: The need for emergency plans is well recognised to manage blood supply in the event of a mass disaster or casualty event. However, these are not often tested until an actual emergency event occurs.

Aims: This exercise was developed to test state and local Emergency Blood Management Plans including blood inventory levels and management, escalation and communication pathways, clinical prioritisation and decision making, and to identify where improvements can be made.

Methods: A simulated mass shooting incident at a major sporting event was developed that resulted in 209 adult and paediatric casualties with both gunshot wounds and injuries resulting from crowd panic and evacuation. To undertake the exercise, simulated patients ('peeps') that included the patient's age, gender, and basic injury information, as well as a blood product request (massive transfusion pack or individual components), were prepared. The peeps and blood product request could be attached to whiteboards using magnetic strips. Red cells (O Positive & Negative) and platelet units were also prepared to represent inventory stocks. The exercise was undertaken with senior, experienced personnel from the transfusion laboratory, haematologists, emergency staff, the ambulance service, the blood supplier, and the state incident management team. All four major hospitals, together with regional healthcare facilities, participated in the four-hour exercise. The exercise was conducted in three separate rooms: (1) the incident site

with ambulance and pre-hospital retrieval teams, (2) hospital emergency departments and their transfusion laboratories, and (3) the blood supplier and the incident management team. Large whiteboards were used to simulate ED triage bays and blood product inventories. Patients were pre-allocated to all trauma hospitals, including a children's hospital. On completion of the exercise, participants engaged in a debrief session, and a post-exercise survey was sent out to all who attended.

Results: The exercise took place over four hours, with more than fifty health service personnel participating. Prehospital staff moved the simulated patients (peeps) from the incident scene to the hospital ED. Hospital staff placed blood product requests with the transfusion laboratory. The transfusion laboratory staff issued blood products, placed orders for replacement stock, moved product between hospitals in a simulated fashion. Orders for additional blood stocks were placed with the blood supplier, and deliveries considered the transit and any quarantine times required. Within less than 90 min the laboratory blood product inventory was depleted, and blood conserving strategies had to be implemented for example, reducing MTP quantities, stock rotation between laboratories, cancellation of non-urgent surgeries, and ceasing urgent and non-urgent transfusions.

Summary / Conclusions: An exercise such as this has never been conducted in South Australia on a state-wide basis to specifically test emergency blood management plans. This proved an effective method and provided invaluable insights that are being evaluated to improve the plans and consider other strategies to manage any future unexpected, sudden demand on blood supply.

PA23-L04 | Blood donor recruitment issues in the context of the social crisis in Bangui, CAR

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Background: Donor recruitment and selection are essential links in the transfusion safety chain. With the humanitarian crises that the Central African Republic has experienced, the most affected population uses blood donation snacks as a means of subsistence. To this end, the responses of candidates for blood donation are often biased. This is because they (the candidates) avoid being excluded from the donation. This phenomenon is on the increase at sites in the capital.

Aims: The aim of our study was to assess the impact of recruiting this type of candidate on transfusion safety.

Methods: We conducted a study of blood donor recruitment and selection data for the period 2022. Pre-donation questionnaires and collection registers were used.

Results: A total of 7749 candidates for donation were recruited on the basis of questionnaires that qualified them as eligible. Of these, 567/7749 (7.31%) were HIV positive; 1515/7749 (19.55%) were HBS positive; 212/7749 (2.73%) were HCV positive and 256/7749 (3.36%) were Syphilis positive. The majority of patients were male, 5364/7749 (69.22%) and female, 2385/7749 (30.77%).

Summary / Conclusions: The prevalence of the four markers in prospective donors is associated with misinformation of these prospective donors. The post-donation snack has become a means of subsistence. As a result, local people move from one site to another to donate blood, and their answers to the questionnaire are often biased in order to avoid being turned away. The risks of infection are enormous for this type of candidate. The military-political crisis in the Central African Republic has had an impact on the social well-being of the population. The answers to the questionnaires of candidates for donations are biased in order to meet the selection criteria and obtain the collation. Difficulties of all kinds are being encountered in the recruitment of donors. A strategy that takes social cases into account would make it possible to only select reliable candidates for donation.

PA23-L05 | Utilization of fresh whole blood on remote Japanese islands

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Background: Japan is an island nation, and blood products have been used on many remote islands. Transfusion therapy on remote islands is particularly challenging when emergency transfusions are required. Fresh whole blood (FWB) would have been used as an option to save lives in these cases, but no systematic investigation has yet been conducted.

Aims: The objective was to determine the availability and effectiveness of fresh whole blood (FWB) on remote islands.

Methods: A survey of fresh whole blood (FWB) was conducted at 82 facilities on 34 remote islands where blood products had been supplied in the past. The analysis period was January 2017 to June 2019. We collected the information regarding transport system of blood products to facilities, management system for blood donors, and patients who received FWB.

Results: Characteristics of facilities performing FWB. Among 82 facilities, 44 responded (response rate: 52.7%). FWB transfusions were reported to have been performed at 9 facilities (20.5% of responding facilities). The time required for regular transport of blood products ranged from 3 h to 2 days, with a median time of 7–10.9 h. Only three facilities replied that disaster-prevention helicopters or Self-Defense Forces aircraft could be used for emergency transport of blood products. Many facilities had larger inventories of universally available O-Red Blood Cells (RBCs), while three facilities did not have blood products in stock even in FWB collection group. FWB was processed differently at each facility, with irradiation being performed in 40% of the facilities. Collection system for FWB Blood donors were often recruited in the following ways: collaborating with the local government to obtain registered donors in the community (42%), assigning hospital employees as blood donors (42%). The number of FWB

collections ranged from 0 to 30 (average 7.2, median 3) during the 2.5-year period at the 9 facilities that reported a history of FWB use. There was no significant correlation between the time required to collect FWB and the amount of blood products used, including FWB. Properties of patients receiving FWB Detailed information on 41 patients receiving FWB was obtained from 7 facilities. The survival rate of these patients was 85% at 7 days. The most common underlying diseases were gastrointestinal diseases, trauma, and malignant diseases, with these three categories accounting for 83% of the total. Gastrointestinal bleeding was a significantly more common reason for FWB transfusion in the no-inventory group than in the inventory group. In addition, pre-transfusion platelet levels were significantly higher and PT-INR levels were significantly lower in the no-inventory group. The amount of FWB used was significantly larger in the inventory group. The future requirement for FWB for similar lesions was noted to be crucial in 35 patients and undecided in 6 patients.

Summary / Conclusions: FWB was effectively utilized in emergency situations at 20% of responding facilities. It is crucial to optimize the usage of FWB and to strengthen the regular supply system of blood products depending on the healthcare environment of the remote islands.

Parallel session—clinical / education

Keep on learning

PA24-L01 | TRANSFUSION CAMP—developing a pediatric focused transfusion medicine curriculum

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Background: Physicians in almost every specialty prescribe and transfuse blood products, the most common procedure in hospitalized patients. It is estimated that of approximately 29 million units of blood and blood products, about 1% are administered to children. Reports highlight that pediatric-related transfusion errors are significant. In the UK, the Serious Hazards of Transfusion (SHOT) hemovigilance group found that 7% of transfusion related deaths and major morbidity were in the pediatric population with gaps in staff knowledge highlighted as a contributing theme. In transfusion medicine (TM), the pediatric patient varies widely from an adult patient with differing physiology, pathology, and additional considerations such as hemolytic disease of the fetus and newborn and alloimmune and autoimmune cytopenias. The pediatric patient also displays a unique response to illness or trauma and subsequent treatments, including a greater incidence of

transfusion reactions in comparison to adult patients. Transfusion protocols are centred on specific guidelines, and in critically ill children, transfusion thresholds for blood products such as plasma and platelets are not well established. Practices are complex and require individualized and patient-centered decision-making. Understanding these principles and their interplay is key to providing safe and effective patient care amongst general pediatricians and subspecialists caring for children, including emergency medicine physicians, surgeons, intensivists, and anesthesiologists. *Transfusion camp* is a longitudinal curriculum, initially designed in Canada and now scaled internationally, that aims to provide a strong baseline level of TM knowledge for non-transfusion medicine trainees. *Transfusion Camp* has been shown to increase trainee TM knowledge and impact trainee reported confidence in their transfusion practice. Pediatric TM knowledge and its application is a known gap among trainees across numerous specialties, including pediatrics. Performance of 330 pediatric trainees across 19 sites in 6 countries on the BEST-TEST 3, an internationally validated assessment used to assess pediatric transfusion knowledge, displayed an overall mean score of 37%, with less than 25% of respondents correctly answering 5/9 questions related to transfusion reactions. These results highlight a need for further initiatives aimed at improving pediatric TM education. With the success of *Transfusion Camp's* didactic and team-based learning (TBL) curriculum for postgraduate non-hematology trainees, there is an opportunity to develop a similar pediatric-focused TM curriculum. During this presentation, the process of adapting the *Transfusion Camp* structure to develop and implement a pediatric TM curriculum aimed at general pediatric and subspecialty pediatric trainees will be discussed. In addition, details related to the development and validation of an assessment tool used to evaluate pediatric specific transfusion knowledge will be reviewed.

PA24-L02 | Community priorities for future transfusion medicine research in Australia—phase one of a community consultation from the Blood Synergy program

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Background: Australia spends EUR 910 million annually on the national blood supply, yet many transfusion practice evidence gaps remain. Engagement with clinicians, patients and the community, including donors, is important to identify priority areas for research to inform transfusion practice. The Blood Synergy is a program to provide new knowledge on how blood products are used in Australia, and how their use can be improved and made more cost-effective.

Aims: To identify transfusion medicine research priorities from the Australian community, including research that could make a difference in the delivery of care to people receiving blood products.

Methods: In 2023 we conducted a cross-sectional community survey. We collected brief demographic information and suggestions for

transfusion research priorities using open-ended questions. Snowball recruitment occurred through the Blood Synergy website and professional, social and advocacy networks. We undertook a content analysis, whereby: open codes were developed and applied to each suggestion to group like suggestions together ideas that had substantial existing evidence were removed axial coding developed overarching research priority statements and the priority statements were classified into domains. Double coding, researcher discussion and consensus provided rigour.

Results: We received responses from 155 participants, who identified as recipients ($n = 43$), carers ($n = 12$), blood donors ($n = 32$), healthcare professionals ($n = 34$) or other professionals ($n = 34$). Respondents could choose more than one category. The lived experience of transfusion was captured by the 87 respondents who identified as recipients, carers, or blood donors. Participants provided 232 transfusion medicine research priorities, which were grouped into 54 research priority statements across 11 domains: Clinical safety and effectiveness research—guiding transfusion decision-making Clinical safety and effectiveness research—transfusion support for specific patient populations. Clinical safety and effectiveness research—optimising use of existing products and practices. Clinical safety and effectiveness research—new products or new ways to use products. Understanding and improving blood utilisation and future demand. Guidelines and standards. Registries, data linkage and networks. Delivering resources and information. Clinician factors influencing blood use. Blood donations and donors. Patient knowledge, barriers, and experiences of transfusion to improve care

Summary / Conclusions: Participant suggestions covered a wide range of topics. Most common statements related to clinical effectiveness questions for use of existing products, and priority populations. Participants and additional experts have been invited to contribute to a modified Delphi process using the results of this consultation to progress the development of national research priorities.

PA24-L03 | Effects of training on leadership skills and teamwork in transfusion medicine—the synergy of scholarly prowess and proficient leadership

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Background: Effective leadership is a multifaceted and highly valued component of healthcare education, increasingly recognized as essential to the delivery of high standards of education, research and clinical practice. To meet the standards of healthcare in this era, creating competent leaders is the need of the hour. Consequently, incorporation of leadership training and development should be a part of all health professional curricula. Healthcare education leaders are required to work effectively and collaboratively across disciplines.

Aims: This study explored the dynamics related to a leadership development programme (LDP) and its impact on the Resident doctors, the nursing team and technical team in Immunohematology and Blood Transfusion Department.

Methods: A single instrumental case study was conducted in a unit of a large academic hospital where an LDP was implemented successfully in the Department of Immunohematology and Blood Transfusion. Data were collected through individual interviews, focus group discussions and scoring of the participants through OSCE (Objectively structured clinical examination) before and after the implementation of this programme. A Total of 16 participants including 5 resident doctors, 7 technical officers and 4 nursing officers were enrolled for the study. The data were categorized and analyzed to assess the impact of LDP on the Resident doctor, the nursing team and technical staff.

Results: Leadership development is an ongoing, interactive process between the Transfusion medicine physician, co-workers and donor-patient populations. The Resident doctors became more effective in areas of self-awareness (29.2% improvement), communication (42.1%), donor management skills (17.7%), patient management (26.2%), Academic (11.5%) and bed-side (18.6%) clinical performance, hands-on skills (12.4%), better donor screening leading to significant reduction in Transfusion Transmitted Infections (0.75 % decrease). They also had better understanding about importance of mental health. The nursing team showed improvement in communication (27.3%) with donors and patients, greater responsibility, empowerment and job clarity. With promotion of good practical skills, they were able to perform better phlebotomies leading to a lesser number of adverse donor reactions (12% reduction) and manage emergencies related to donor or patients. Technical team showed significant improvement in hands-on skills (15.8%), re-learning the basics of test principles leading to better understanding of test results, increased work efficiency leading to lesser Turn-Around Time (TAT) [improvement for 4 min] and ultimately better patient care. Overall better management and leadership skill promoted upbeat ambience and a sense of camaraderie in the department of Immunohematology and Blood Transfusion.

Summary / Conclusions: The study gives a deeper insight of the importance of a good stewardship towards attaining a higher level of competency among the participants. The content of clinical leadership development must encompass a holistic conceptualization and should use simulation-based learning, and team-based approaches, to improve proficiencies among healthcare professionals.

PA24-L04 | Can medical students use AI to learn transfusion? ChatGPT and the ASH medical student transfusion learning objectives

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Background: ChatGPT, a large-language A.I. released in 2022, has been studied as a tool for medical education. Some authors have reported that ChatGPT performs reasonably well on medical student examinations,

and it is already used by some medical students as an educational resource. It is however well known that ChatGPT invents facts which do not exist, and is not able to express uncertainty: it may provide learners with incorrect answers which seem convincing. It is unclear whether medical students should rely on ChatGPT to learn transfusion material.

Aims: To determine whether ChatGPT provides correct or incorrect answers when presented with transfusion queries at the medical student level.

Methods: The American Society of Hematology (ASH) has published online 199 medical student hematology course learning objectives intended for use by second-year medical students, and which may also be the basis of further study by more advanced trainees. These include 29 transfusion learning objectives, covering topics in basic transfusion science and clinical transfusion medicine. These transfusion objectives were revised into 31 separate queries which were entered into the ChatGPT text-based interface. ChatGPT responses were then separately graded by 3 expert transfusion medicine physicians using a 4-point scale analogous to US/Canadian university grades (A = 4, E = 0). Marking discrepancies were resolved at a consensus conference. Answers evaluated as incorrect were reviewed to assess the type of question asked and the type of error generated by ChatGPT.

Results: The overall grade of ChatGPT in answering the transfusion questions was 2.27 out of 4.0, equivalent to a C grade on an A/B/C/D/E scale. It performed well (grades of A or B) in naming or listing transfusion tests, indications, or complications; for example a grade of "A" for "Name the four major blood groups in the ABO system". ChatGPT performed moderately (graded at C) on questions which were somewhat more complex or more clinical, such as when presented with a specific clinical scenario about an Rh+ fetus. ChatGPT's results were worst (grades of D or E, both representing unsatisfactory performance) on questions asking for details about current/evolving clinical practice, such as explaining platelet alloimmunization and its prevention and management, and quantifying specific transfusion risks. Overall, ChatGPT shows the most difficulty in answering questions about clinical practice where details have changed over time, or where transfusion concepts may be understood differently by non-experts compared with transfusion physicians: for example, ChatGPT gave outdated figures for risks of Transfusion Transmitted Infections, and showed an incorrect/outdated understanding of the use of plasma and cryoprecipitate—suggesting for both that Hemophilia A was an appropriate current indication.

Summary / Conclusions: As a representation of modern computer programming, ChatGPT's performance on transfusion questions is remarkable: on average, it scored a passing grade on a series of 31 transfusion questions taken from the ASH medical student objectives. As a potential learning resource for medical students and other trainees, ChatGPT's answers were very often superficial, vague, incomplete, outdated, incorrect, and even unsafe. Medical students should not use ChatGPT to learn transfusion medicine.

PA24-L05 | Designing and evaluating the impact of a competency-based training program in transfusion medicine for MBBS interns

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Background: The traditional method of delivering medical education has some limitations in both design and approach. Competency-based medical education (CBME) is a modern approach being applied to medical training across the Indian subcontinent. Transfusion medicine (TM) training is no exception and requires a structured competency-based training program that leads the way for others to follow. This discipline bridges both clinical and laboratory aspects of benign haematology and blood medicine. The National Medical Commission (NMC) in New Delhi, India advocates for a structured CBME for undergraduates in TM across the Indian subcontinent. The purpose of this research is not only to provide an overview of CBME but also to design the application of CBME to MBBS interns in transfusion medicine.

Aims: To assess their knowledge of both pre-and-post CBME-based training cum workshop programs for MBBS interns regarding Transfusion Medicine.

Methods: This study was conducted at the Department of Immuno-hematology and Blood Transfusion using a questionnaire-based cross-sectional approach. Three faculty members from TM designed the questionnaire and used the Content Validity Ratio (CVR) to determine which questions to include. The questionnaire consisted of six sections: Section I—Indication of blood component therapy, Section II—Immunohematology, Section III—Blood Banking, Section IV—Bedside transfusion practices, Section V—Transfusion reaction and Management, and Section VI—Transfusion-transmitted Infection (TTI). Each section contained five questions, making a total of 30 questions. All participants underwent a pre-training test followed by a CBME-based training workshop program and a post-training test. The scoring was out of 30. The data was analysed using SPSS software by entering the data and test scores into a Microsoft spreadsheet.

Results: Out of the total of 113 interns, only 91 (80.53%) participated in the workshop. The average age of participants was 23.7 ± 1.3 years old, with a female-to-male ratio of 50:41. Overall, the scores of participants before and after the workshop were 13.95 ± 3.8 and 23.3 ± 3.2 out of 30, respectively, and there was a significant difference between the two scores ($p < 0.0001$). In each section, the mean scores before and after the workshop were as follows: section (2.3 \pm 0.8 and 3.6 \pm 0.8, respectively, $p < 0.0001$, CI: 95% 1.0–1.5), section II (3 \pm 1.1 and 4.6 \pm 0.7, respectively, $p < 0.0001$, CI: 95% 0.3–1.0), section III (1.9 \pm 1.1 and 4.1 \pm 0.9, respectively, $p < 0.0001$, CI: 95% 1.9–2.4), section IV (2.9 \pm 1.5 and 4.6 \pm 0.7, respectively, $p < 0.0001$, CI: 95% 1.3–2.0), section V (1.6 \pm 0.9 and 3.4 \pm 1.1, respectively, $p < 0.0001$, CI: 95% 1.5–2.09), and section VI (2.0 \pm 0.9 and 3.7 \pm 0.9, respectively, $p < 0.0001$, CI: 95% 1.4–1.9). These section-wise scores indicate a significant improvement in all sections after the workshop. Section III

showed the greatest improvement, indicating the largest knowledge gap.

Summary / Conclusions: To conclude the knowledge among interns significantly improved with workshops emphasizing the need of such competency-based initiatives. Mandating their posting in the blood transfusion services will not only increase their knowledge but also increase their confidence in managing adverse events in their clinical practice.

Parallel session—donors and donation

Innovations in donor management

PA25-L01 | Civilian walking blood banks—crisis preparedness for when banked blood is unavailable

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Background: Early balanced blood transfusion is recommended in Civilian and Military guidelines for patients with hemorrhagic shock. To enable lifesaving blood transfusions to patients in hemorrhagic shock when banked blood is unavailable, a system for emergency whole blood collection in the framework of a civilian walking blood bank can be established. In the Norwegian Blood Preparedness Program, four civilian walking blood banks have been established by March 2024.

Definition: In a civilian walking blood bank, whole blood is collected on site from a prescreened emergency donor pool for immediate transfusion to patients with severe bleeding.

Methods: In the Norwegian Blood Preparedness program we have established civilian walking blood banks based on the following principles: Implementing a civilian walking blood bank requires a system for education, training, supervision and revision. The civilian walking blood banks must be subject to the supervision from a responsible blood service, the “Mother Blood Bank”, and regulated according to national/regional regulations. Routines on donor selection, whole blood collection, and emergency transfusion must be established and trained in collaboration with all stakeholders. Whole blood donors are selected based on donor interview, physical examination and laboratory investigations. It is important to educate the emergency blood donors in donation routines and donor selection criteria, so that the safety of donors and patients are maintained. Emergency donors undergo testing for transfusion transmissible disease (TTD) and donor interviews at inclusion, every 6 months, and at donation (post-transfusion testing). It is important to educate the emergency blood

donors in donation routines and donor selection criteria, so that the safety of donors and patients are maintained. By using a whole blood collection bag with integral access ports for connection of infusion sets, transfusion can be given without further processing of the blood. The blood bags must be labelled with a unique donation identification number to ensure traceability. Adverse donor or patient events are to be recorded and monitored according to national/regional guidelines.

Summary: We conclude that implementation of a Whole Blood based emergency collection and transfusion program is feasible also in the civilian health services, and that a civilian walking blood bank may enable life-saving blood transfusions in crisis situations when banked blood is unavailable.

PA25-L02 | Chat GPT as a tool for blood donor recruitment

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Background: In Brazil, where the demand for blood is constant and the genetic diversity of the population poses additional challenges, donation campaigns are vital to ensuring an adequate blood supply to meet various medical needs. However, despite significant efforts, we face persistent challenges such as shortages in certain blood groups and reliance on periodic campaigns. It is in this context that innovative technologies emerge, particularly artificial intelligence (AI). The ability of AI to analyze large datasets, identify complex patterns, and optimize processes offers unique opportunities to revolutionize blood donation promotion. The emergence of generative AI, such as Chat GPT, has made its use more accessible, enabling the execution of strategic campaigns.

Aims: To evaluate the use of Chat GPT in promoting blood donation at a university hospital blood center.

Methods: In addition to employing usual strategies, we developed a campaign for the National Blood Donor Week in 2023 using Chat GPT 3.5. For this, Chat GPT was trained through questions about blood donation, and then the elaboration of the campaign was requested, considering specific service characteristics such as limited budget, collection unit in a university hospital, and low number of staff. We also requested that the AI develop and detail the suggested actions, which were then put into practice. We compared the number of donors attendances, the quantity of first-time donors, and the deferral rate during the same period (third week of November) of the 2023 and 2022 campaigns, the year in which artificial intelligence was not used.

Results: The total number of attendances at the blood center during that week in 2023 was 88, compared to 74 in 2022, representing an increase of 18.9%. The quantity of first-time donors was also higher in 2023: 56 compared to 41 in the previous year. The deferral rate in 2022 was 28.4%, and in 2023, it was 25%, indicating a decrease of 3.8%.

Summary / Conclusions: The use of AI led to an 18.9% increase in donor attendance at the blood center compared to the same period in the previous year, without worsening the deferral rate. Although the quantity of donors was small, the study indicates that Chat GPT can be used as an

additional tool for blood donation campaigns, especially in the context of public hospitals with limited budgets, such as ours. Further studies are necessary for evaluation of AI full impact in blood donation.

PA25-L03 | Artificial intelligence in blood donation—implementation of an automated and efficient system to optimize the call for donors in three hospitals of Catalonia

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Background: Blood establishments, such as the Blood and Tissue Bank (BST) of Catalonia, are responsible for collection, testing, processing, storage and distribution to health institutions of blood and blood components, as well as promoting donation to face this need. Artificial intelligence (AI) offers a disruptive and powerful solution to optimize these processes through the efficient analysis of massive data and providing strategic knowledge on which to base decision-making.

Aims: The aim of this work was to develop and implement an automated system based on AI to optimize blood donation and guarantee a more efficient and sustainable distribution of donors and donations in Catalonia.

Methods: We used the information of the BST database from 2015 to 2023. Blood donors were categorized using principal component analysis (PCA) and the clustering algorithm HDBSCAN. Donation dynamics in Catalan municipalities was characterized by PCA and agglomerative clustering. Prediction of donor response to the donation call was addressed with the classification algorithm XGBoost. These developments were integrated into an automated workflow that returned the most optimal combination of donors to contact to achieve the donation goal for each campaign. This system was validated in twelve donation points and, since October 2022, it has been implemented in the weekly donation call of three Catalan hospitals. To assess the benefits of this strategy, the performance of these centres during the first year was compared with that to the previous 12-month period.

Results: Clustering analysis revealed 20 different categories for blood donors according to their sociodemographic and donation profiles. Catalan municipalities were found to cluster into eight distinct groups with very different dynamics in donation and of the

donor cohort. The prediction model was capable of discriminating those donors that would respond to a call from those who would not, and was optimized to maximize the capture of potential responders (recall = 98.6%, accuracy = 84.2%, precision = 76.4%). The application of this model in targeted campaigns led to a notable increase in donor response in all cases, being more than double in half of them. The implementation of the automated system in the weekly donation call in three hospitals improved the efficiency of the process in one of them, increasing donor response from 8.7% to 11.0% considering donations in the same centre, and from 9.8% to 15.3% including any donation point, while it had a neutral influence in the remaining two. These results revealed the impact of this strategy to be dependent on the active donor cohort of the hospital, the potential donor population of the area, and the coordinated planning of surrounding mobile campaigns.

Summary / Conclusions: The automated, AI-based system developed has demonstrated great potential to optimize the calling process for donation and to improve the efficiency and sustainability of the distribution of donors and donations in Catalonia. Successfully addressing these goals allows the BST to respond more effectively to foreseeable fluctuations in donations and to anticipate the progression of the donor cohort, as well as to address unexpected shortage situations such as the one caused by the COVID-19 pandemic. Therefore, the AI-based system implemented definitely contributes to improving the processes that guaranty BST's mission of supplying blood components to the population and is a powerful asset applicable to the activity of other blood establishments.

PA25-L04 | Building community—the AR revolution in blood donation in Catalonia

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Background: In January, after the Christmas holidays, one of the periods in which blood donations decrease the most, it is essential to focus on the discourse of the importance of blood donation. There arises the need to work together, to build a community. However, living in a hyper-informed society makes it increasingly difficult to reach the public and generate this community. In this sense, innovation is key to fostering community participation.

Aims: To forge a cohesive community and enhance social ties within the realm of blood donation. To use augmented reality technology to redefine the narrative surrounding blood donation, shifting it from a solitary act to a collective effort. To promote the idea that blood donation is the responsibility of the entire society.

Methods: We generate a campaign slogan rooted in this concept of community "Doing it together is truly ours," and from there, we develop a dynamic augmented reality sculpture that is built with each blood donation. Donors condition the evolution of this sculpture with their donations. Additionally, it symbolizes the shape of a human tower, a typical construction of Catalan culture. In this way, we reinforce the message "it's truly ours," where we

merge tradition and technology. It is an experience accessible through mobile devices, located in high-traffic areas throughout Catalonia, ensuring participation and commitment. From this initiative arise all the following actions: 1. **Explaining the project on the website where appointments for blood donation are reserved.** This way, we do not divide traffic and focus efforts. 2. **Street marketing. Installing large totems** in areas where the augmented reality monument can be seen through a QR code. 3. **Newsletter** to the database. 4. **Social media** publication strategy and promotion of MGM. 5. Putting **media** into play to echo the pioneering initiative. 6. **Associative entities** that share DNA with this project, acting as mouthpieces.

Results: We succeed in having municipalities want to host this integrating and innovative project at the same time. We exhibit it in the 4 most important cities in Catalonia. And during its inauguration, we detect a significant increase in commitment and community participation in blood donation. The dynamism of this project engages the population, and its human tower shape fosters a sense of belonging. The fact that it is a sculpture representing Catalan culture causes users to identify with it more and emotionally connect with the initiative. Consequently, interactions on social networks increase by 56%, improving the engagement ratio from 4.4 to 6.9. Although it is an initiative that does not seek immediate conversion but rather positioning, while the initiative occupies conversation spaces, donations increase by 30% more than in a normal week.

Summary / Conclusions: The power of augmented reality allows us to generate omnichannel conversation around blood donation. Visualizing the collective impact of the sum of individual gestures using innovative applications fosters a culture of collaboration and collective responsibility. Additionally, the use of AR positions the Blood and Tissue Bank as a modern, innovative brand focused on the donor experience.

PA25-L05 | Genome-wide association study reveals the unique genetic structure of blood donors

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Background: Blood donors are considered as an excellent option for healthy control cohort in genetic research. Voluntary and often frequent and continuous blood donation provides unique possibilities for longitudinal sampling. However, blood donation criteria that must be met to be eligible to donate blood, leads to a highly selected population, that is, to the *healthy donor effect*. In addition, blood group-based donor recruitment may result in enrichment of certain blood group antigens, with well-known disease associations, in a donor pool.

Aims: We conducted a genome-wide association study (GWAS) between the blood donors of Finnish Red Cross Blood Service Biobank and a mainly hospital-based population, FinnGen, to reveal the possible genetic differences between the two populations and to understand the possible genetic impact of the healthy donor effect.

Moreover, our aim was to discover genetic variants possibly effecting donor health.

Methods: Additive GWAS was performed for 53 688 blood donors and 228 060 controls using Regenie v2.2.4 in FinnGen pipeline. Sex, age, BMI, PC1-10, birth region, and FinnGen genotyping array version were used as covariates. All the genome-wide significant ($p < 5e-08$) hits were fine-mapped with FinnGen pipeline using SuSiE. Further association analyses were conducted on fine-mapped variants and blood groups, BMI, hemoglobin, HLA and 227 disease phenotypes predefined by FinnGen. Genetic correlation on detected phenotypes is ongoing by LD Score Regression v1.0.1 in FinnGen pipeline. Protein quantitative trait loci (pQTL) analysis was performed using multiplex antibody-based immunoassay database and multiplex aptamer-based immunoassay database, Olink and SomaScan, respectively, to detect statistically significant association between GWAS hits and protein expression levels.

Results: Genome-wide association analysis revealed 2973 genome-wide significant ($p < 5e-08$) genetic loci associated with blood donorship. After fine-mapping, 5 coding and 36 non-coding variants were detected. Strongest association was seen in blood group genes ABO in chromosome 9, Kell in chromosome 7 and rs55794721 in chromosome 1 which was associated with Rh antigens. Strong associations were seen in blood donors in iron metabolism: risk towards hemochromatosis conferred by putative novel rs9968910 and lower allele frequency of iron deficiency anemia risk increasing RNF43 variant. Various significant associations with autoimmune diseases, cardiovascular conditions, and medication with mental disorders were seen. Several HLA-alleles were detected by HLA association analysis, that is, the well-known autoimmune risk allele HLA-DRB1*04:01 ($p = 5, 8e-15$) and HFE C282Y associated HLA-B*07:02 ($p = 8, 6e-08$). 112 different statistically significant ($FDR \leq 0.05$) protein expression level variations were detected by the pQTL analysis which resulted in enrichment for biological processes for cell adhesion and antigen binding in Gene Ontology.

Summary / Conclusions: We demonstrate, to our knowledge for the first time, the genetic impact of the healthy donor effect and blood group-based selection of blood donors. The unique genetic structure of blood donor population may enable new perspectives and possibilities in disease-associated research. Enrichment of certain blood group antigens in blood donor population may also be a challenge if not taken into consideration. Moreover, the results reveal genetic factors affecting on blood donor health and blood donation suitability.

Parallel session— immunobiology

Blood groups—new discoveries and old mysteries solved

PA26-L01 | Exonic deletions in the MAL gene, encoding myelin and lymphocyte protein, define the rare inherited AnWj-negative phenotype

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Background: AnWj (ISBT 901009) is a high prevalence blood group antigen present on red cells and epithelial tissues of more than 99.9% of individuals but absent on cord cells. Antibodies directed to AnWj antigen are rare and are usually associated with transient suppression of AnWj expression, although a small number of individuals have a persistent, autosomal recessive inherited AnWj-negative phenotype. Anti-AnWj may be clinically significant and has been associated with acute hemolytic transfusion reactions. The carrier molecule for the AnWj antigen has remained elusive; although CD44 and Smyd1 histone methyltransferase have both been reported to be associated with AnWj expression, the mechanism for this has not been elucidated. Myelin and lymphocyte protein (Mal), encoded by the MAL gene on chromosome 2, is an integral 17KDa multi-pass membrane proteolipid, expressed on T-lymphocytes, myelin-forming cells and polarised epithelial cells. It has also been identified as the receptor for *Clostridium perfringens* epsilon toxin on erythrocyte membranes.

Aims: We report serological and genetic investigations from patients with inherited and transient AnWj-negative phenotype, in addition to expression studies to elucidate the carrier molecule for the AnWj antigen.

Methods: Whole exome sequencing was performed, and variants were filtered to search for rare, homozygous variants consistent with loss of high prevalence AnWj antigen. Alignments of MAL, CD44 and SMYD1 were visualised using Integrative Genomics Viewer. Expression of proteins of interest and AnWj antigen was assayed using

serology, flow cytometry and western blotting on red cells and BEL-A erythroid cell-lines modified by CRISPR gene editing.

Results: Exome sequencing of two unrelated inherited AnWj-negative individuals revealed no coding mutations in *CD44H* (encoding erythroid CD44), and only one carried the homozygous *SMYD1* mutation previously reported to cause the AnWj-negative phenotype. However, both patients were homozygous for the same large, 6646nt, deletion in *MAL*, encompassing coding exons 3 and 4 (NC_00002.12, NM_002371.4; c.262-423_462+2348del). A further three unrelated inherited AnWj-negative patients were also homozygous for the *MAL* deletion, whilst AnWj-positive controls and those with transient AnWj-negative phenotype had wild-type *MAL*. AnWj-positive red cells were shown to be Mal-positive by serology, flow cytometry and western blotting, whilst AnWj-negative red cells, whether inherited or transient, had no detectable Mal surface expression. Binding of anti-Mal to AnWj-positive red cells was inhibited by binding of human anti-AnWj. No expression of Mal or AnWj could be detected in wild-type or Mal knockout (KO) BEL-A erythroid cells, but over-expression (OE) of Mal restored expression of AnWj antigen. No effect on Mal or AnWj expression was observed following either OE or KO of *CD44*, showing that *CD44* is not required for AnWj expression.

Summary / Conclusions: We demonstrate Mal proteolipid to be the carrier molecule for the AnWj red cell antigen, with homozygosity for the same large deletion in *MAL* underlying the inherited AnWj-negative phenotype in all patients studied. Lack of Mal protein was shown on red cells of both inherited and transient AnWj-negative phenotype, and over-expression of Mal protein in an erythroid cell-line resulted in AnWj expression, showing that Mal is required for expression of AnWj. Our data finally elucidate the genetic background of the AnWj antigen, establishing a new blood group system.

PA26-L02 | Disruption of the ATP11C flippase causes a rare blood phenotype and underlies a novel erythroid blood group system

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Background: The International Society of Blood Transfusion (ISBT) officially recognizes 390 blood group antigens (BGAs), 362 being distributed in 45 systems. Besides, the so-called "orphan" antigens, of unknown molecular basis, are classified in collections and the 700 and 901 series. However, other orphan BGAs do exist, but with no name yet assigned, pending their molecular characterization. An alloantibody of undetermined specificity was found in 7 male patients (from 4 families), all but one suffering from severe hemophilia B. They were all found to be serologically cross-compatible, which means they all likely shared the same rare blood type.

Aims: To elucidate the molecular basis of the rare blood type shared by the 7 patients; to investigate the function of the protein carrying the related antigen.

Methods: A whole-exome sequencing followed by a variant filtering strategy was performed on genomic DNA in 4 unrelated patients. The reactivity of the alloantibody was investigated by flow cytometry analyses in the patients' RBCs and in a CRISPR-Cas9 K562 model. Protein studies were carried out by western blot, immunoprecipitation and comparative global proteomic analysis. Morphological and rheological characteristics of the patients' RBCs were explored.

Results: Exome sequencing revealed different large deletions in Chr X, including *F9* in the 6 patients with hemophilia B. Only 3 common genes were deleted in all patients: *MCF2*, *CXorf66* and *ATP11C*. *ATP11C* was considered the best candidate, as it is known to encode a flippase present in the RBC membrane, responsible for phosphatidylserine (PS) transport. We then hypothesized that the antibody was directed against *ATP11C*. Western blot analysis with a polyclonal anti-*ATP11C* confirmed the absence of the protein in patients' RBCs. The absence of reaction of the antibody with *ATP11C* KO K562 cells was consistent with an *ATP11C*-related antibody. *ATP11C* KO cells stably transfected with an *ATP11C* plasmid showed a full restoration of antigen expression. The antibody failed to immunoprecipitate *ATP11C* from KO cells. Previous studies described *ATP11C* as the major (90%) erythrocyte membrane flippase, and the exposure of PS on the RBC surface is considered a marker of senescence. Unexpectedly, no severe erythroid disorders were detected in our *ATP11C* null patients, including rheological and morphological properties, and PS exposure. However, a mild anemia was observed in most patients. Analysis of differentially expressed proteins between the control group and patients did not reveal any compensation by another flippase or scramblase. Consistently, the inactivation of *ATP11C* in CD34+ cells did not alter erythroid differentiation and proliferation. Interestingly, the investigation of B lymphocytes from our *ATP11C* null patients confirmed the role of this protein in the immune response, in accordance with the murine model. Of note, 3 patients from 2 families also showed a neurodevelopmental disorder, which is likely due to the fact that the large deletion also encompasses the *SOX3* gene.

Summary / Conclusions: The rare phenotype in our patients is caused by the absence of the flippase *ATP11C* (null phenotype), defining a novel blood group system. Interestingly, RBC integrity is preserved and in-vitro erythropoiesis unaffected despite the absence of *ATP11C*.

PA26-L03 | Luke, may the FORS be with you? A premature stop codon in GBGT1 is associated with loss of the sialylated high-prevalence antigen LKE

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Background: Luke (ISBT no. 901017) was first reported in the mid-1960s as a high-prevalence antigen (Tippett, Vox Sang, 1965). The Luke-negative (LKE[−]) phenotype is found in 1-2% of populations but anti-LKE is a rare specificity. LKE is part of the globo series of glycolipids and is an extension of Gb5 (Stage-Specific Embryonal Antigen 3 = SSEA-3) to monosialosylgalactosylgloboside (MSGb5 = SSEA-4 = LKE). LKE varies in strength on RBCs and its expression affects other glycolipid levels. The genetic basis of LKE[−] remains unknown, even if candidate loci have been proposed, for example, *B3GALT5* and *ST3GAL2*, which encode glycosyltransferases (GTs) extending Gb4 to Gb5 and Gb5 to MSGb5, respectively.

Aims: To elucidate the genetic basis of LKE[−].

Methods: Whole exome sequencing (WES) was performed on DNA from 4 LKE[−] reagent RBC samples (one from a donor with anti-LKE). PCR and Sanger sequencing was used to investigate SNVs in selected genes (*A3GALT2*, *B3GALT5*, *GBGT1*, *ST3GAL2*). Flow cytometry was performed with anti-SSEA-4 for LKE detection (clones MC813-70, REA101). Additional testing was performed with anti-P, -P^k, -PX2, -P1 and -ExtB. GT co-transfections of cell line HL-60 were performed. ColabFold (Mirdita, Nat Methods, 2022) was used to assess interactions between GTs. Screening for LKE[−] donor samples was done by pheno- (*n* = 239) or genotyping (*n* = 100). 19 samples from a previous genetic screening cohort (*n* = 1,108) were available for phenotyping.

Results: WES revealed a SNV, c.363C>A, p.Tyr121Ter (rs35898523) in the *GBGT1* (FORS1 synthase) gene to correlate with lack of LKE expression. Two of four samples were homozygous for c.363C>A and a 3rd sample was heterozygous. Two SNVs in *A3GALT2* (rs72889865, rs201715975) were found in the 4th sample, not carrying c.363C>A. Screening identified 5 LKE[−] and 2 LKE very weak (LKE^{+vw}) samples. Four of 5 LKE[−] samples were c.363A/A while the others were c.363C/A. A LKE^{+vw} sample had both the above *A3GALT2* SNVs. Compound heterozygosity for c.363C>A and a rare SNV in *GBGT1* (c.197A>G; rs117595304) was found in one initial LKE[−] sample and one LKE[−] found by screening. Additional genetic testing was performed on samples with high and weak expression of LKE. All LKE^{+high} samples were wildtype c.363C/C whereas >50% of LKE^{+w} samples were c.363C/A. Elevated levels of P^k were observed in both LKE[−] and LKE^{+vw/w} samples. Low levels of P, PX2 and ExtB were also noted (all antigens made by P synthase, *B3GALNT1*). Co-transfection of *B3GALNT1*, *B3GALT5* and wt or c.363C>A *GBGT1* indicated higher expression of LKE with the wt construct. ColabFold analysis of FORS1 synthase and P synthase indicated a possible

heterodimer, but results were not conclusive due to disordered loops close to the interacting GT surfaces.

Summary / Conclusions: We show here that a premature stop codon in *GBGT1*, a gene governing the FORS blood group system, is associated with loss of LKE, one of three remaining 901 series antigens without a known genetic basis. Of nine LKE[−] samples, six were homozygous for c.363A/p.Tyr121Ter, and three were not fully explained but two heterozygous for c.363C/A. Furthermore, both LKE^{+vw} and most LKE^{+w} samples were c.363C/A. We hypothesize that FORS1 synthase acts as a chaperone for other globo series GTs, especially P synthase since its products are weakened and its acceptor substrate P^k increased. Further studies are needed to prove or refute this idea, although overexpression indicated synergy between implicated GTs. That aside, *GBGT1* genotyping can now be applied to identify LKE[−] donors.

PA26-L04 | A Leu151Phe substitution in Choline transporter-like protein 2 (CTL2), adjacent to Arg152Gln, encoding Csa/Csb antigens, encodes a novel high prevalence red cell antigen

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Background: The CTL2 blood group system, until recently, comprised two high prevalence antigens, VER and RIF, carried on Choline transporter-like protein 2 (CTL2), encoded by *SLC44A2*. CTL2 is also expressed on neutrophils, carrying HNA3a and HNA3b antigens, encoded by Arg152 and Gln152 respectively. All numbering is based on the shorter, 704 amino acid isoform of CTL2 (TV2; NM_001145056.2), known to be expressed in neutrophils; equivalent to Arg154Gln in longer isoform (TV1; NM_020428.3). Anti-HNA3a and anti-HNA3b are clinically significant, implicated in both Neonatal Alloimmune Neutropenia and Transfusion-related Acute Lung Injury. It was recently reported that Cs^a, an antigen of moderately high prevalence, and the antithetical Cs^b antigen (formerly of the Cost collection) are encoded by the same amino acid substitution as HNA3a/HNA3b and the antigens were moved to the CTL2 system. Anti-Cs^a and anti-Cs^b are not clinically significant, but only one example of anti-Cs^b has been reported.

Aims: Samples from a male patient for preoperative testing, were subjected to serological investigations and whole exome sequencing for identification of a weak to moderate strength pan-reactive antibody in his plasma.

Methods: Serological investigations were performed by standard LISS tube IAT and direct agglutination techniques. Whole exome DNA sequencing was performed, and data was filtered to search for rare, homozygous coding variants consistent with lack of a high prevalence

antigen. Flow cytometry was performed on transfected HEK293 cells expressing different variants of CTL2.

Results: The antibody present in the plasma of the male patient was found to react weak to moderate strength with all untreated and papain treated cells tested by LISS IAT, including Cs(a-) cells and RIF- cells. Exome sequencing revealed a rare homozygous *SLC44A2* mutation; c.451C>T, encoding Leu151Phe (NM_001145056.2; rs147820753; freq. 0.005). The patient was homozygous c.455G in exon 6 (Arg152), predicting a Cs(a+b-) phenotype, and had normal expression of Cs^a antigen. Exon 14 sequence showed homozygosity for wildtype c.1192C (Pro398), predicting a RIF+ phenotype, and no further coding mutations were found in *SLC44A2*. The patient's antibody was shown to bind only HEK cells expressing CTL2 Leu151 and not CTL2 Phe151 (irrespective of the presence of either Arg (Cs^a) or Gln (Cs^b) at position 152), demonstrating this substitution encodes a novel antigen.

Summary / Conclusions: We report a new high prevalence antigen carried on CTL2, encoded by c.451C (Leu151), adjacent to the substitution encoding the Cs^a/Cs^b antigens. Homozygosity for c.451C>T resulted in lack of this antigen and associated alloantibody production in a male patient. The mutation was carried on a Cs(a+) genetic background in this case but did not alter expression of the Cs^a antigen. This novel antigen will become the fifth antigen in the CTL2 blood group system, subject to ratification by the ISBT Working Party on Red Cell Immunogenetics and Blood Group Terminology. The patient was transfused multiple times, with red cell units positive for the newly identified antigen and therefore incompatible, without any signs of haemolysis. The Leu151Phe substitution has been shown to impair anti-HNA3a binding in neutrophils, although has not been demonstrated, as yet, to encode a neutrophil antigen. As with CD36 antigen, on both platelets and erythrocytes, discovery of further crossover between platelet, neutrophil and red cell antigens seems likely.

PA26-L05 | Novel SC/ERMAP Variant c.217C>G (p.Arg73Gly) associated with expression of a low-prevalence Scianna blood group system antigen implicated in HDFN-affected pregnancies

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Background: Antibodies (Ab) to low-prevalence antigens (LPA) causing HDFN can be difficult to identify. A G4P2 pregnant patient with

consistently negative antibody screens required intrauterine red cell transfusion (IUT) for fetal anemia in two consecutive pregnancies starting at 28 (third gestation, G3) and 25 weeks (fourth gestation, G4), respectively. Maternal plasma (MP) during G3 and G4 showed the presence of an Ab that reacted with paternal red blood cells RBCs (PRBCs) and an Ab to a low prevalence antigen present on PRBCs was suspected. With no alternate cause for fetal anemia identified, the positive DAT on fetal RBCs (FRBC) prior to first IUT and the incompatibility of maternal plasma with paternal RBCs all pointed to an alloimmune process. Prior first trimester miscarriage and twin gestation complicated by intrauterine fetal demise were probable sensitizing events. Parents are of Indian (Gujarati) descent.

Aims: To determine the specificity of the maternal antibody causing HDFN.

Methods: Serologic testing was by standard hemagglutination methods. MP was used for Ab ID and was diluted in PBS for titration against PRBCs. Antigen typing and enzyme/DTT treatment of RBCs was done with in-house reagents. Genomic DNA was isolated from WBCs and used for amplification and Sanger sequencing of SC/ERMAP coding regions and flanking introns. Fetal blood was obtained by cordocentesis. MP was tested with FRBCs treated with EDTA-glycine acid (EGA).

Results: Maternal RBCs typed as group O, D- with an unremarkable common RBC phenotype. MP was negative by IAT with panel cells expressing common RBC antigens including a battery of panel RBCs covering 37 LPAs but reacted 3+ with PRBCs. MP reacted 3+ by IAT with papain or 0.2M DTT treated, 2+ with α-chymotrypsin treated and 1+ with trypsin treated PRBCs suggesting Ab against a Sc antigen. MP was negative with Sc2+, Sc4+, and SCAC- negative RBCs. The Ab titers in MP were 64 at 9- and 25-week gestation points. PRBCs typed SC:1,-2,3 and SCAC+. FRBCs were 4+ DAT+ (IgG). MP reacted 3+ by IAT with EGA- treated (DAT-) FRBCs. SC sequencing of paternal and fetal samples identified novel heterozygous change c.217C>G (p.Arg73Gly), which is not listed on gnomAD v.2.1.1. No other changes in SC were found.

Summary / Conclusions: We identified an undescribed SC variant, c.217C>G (p.Arg73Gly), in paternal and offspring samples associated with expression of a novel, low prevalence Sc antigen for which we propose the name SCAB. SCAB brings the total Sc antigen count to ten and the LPA count to three. Previous 3D protein structure modeling and analysis predicts position p.73 to reside extracellularly on the surface of the IgV domain of the Sc glycoprotein. Interestingly, the novel SC variant c.217C>G (p.Arg73Gly) described here is associated with expression of a LPA while variant amino acid change p.73Cys (encoded by c.217C>T, c.219C>T), has been reported to result in loss of the high-prevalence antigen SCAC. Our Testing showed that SCAC- negative RBCs do not express SCAB, and that SCAB+ RBCs express SCAC. Our study highlights the importance of investigating LPAs in cases of fetal anemia in the presence of a negative maternal antibody screen. This study implicates anti-SCAB as causative in two HDFN-affected pregnancies requiring intrauterine transfusions. Anti-SCAB joins antibodies to the low- prevalence Sc2 and Sc4 antigens as causes of HDFN.

PA26-L06 | In-depth proteomic analysis and structural modelling of protein-protein interactions identify known and novel molecular binding partners for EMP3 in erythroid cells

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Background: The absence of Epithelial Membrane Protein 3 (EMP3) on red blood cells (RBCs) underlies the MAM-negative (MAM⁻) phenotype. EMP3 is a ubiquitous protein found at low levels in most tissues but with higher expression in hematopoietic cells. Low levels of CD44 are a signature of MAM⁻ RBCs due its proposed physical interaction with EMP3, indicating that EMP3 stabilises CD44 in the membrane (Thornton, Nature Communications, 2020). Erythroid culture of CD34⁺ cells from peripheral blood from MAM⁻ individuals showed an unexpected expansion of erythroid cells, suggesting that EMP3 acts as a brake on the erythropoietic pathway. However, the function of EMP3 in erythroid cells is yet to be elucidated.

Aims: To enhance our understanding of the function of EMP3 by investigating its molecular partners in erythroid cells, including but not limited to CD44.

Methods: Global proteomics were performed on three examples of MAM⁻ RBCs from our in-house collection of rare reagent RBCs and compared to three anonymized MAM-positive (MAM⁺) samples. Flow cytometric analyses were performed on RBCs and cultured cells. *In silico* analysis of the expression differences of blood group proteins incl. EMP3 was performed in a dataset from CD44 overexpression in HEK293 cells. 3xFLAG-tagged EMP3 (fused to either the N- or C-terminus) was overexpressed in erythroid cells cultured *in vitro* from CD34⁺ hematopoietic stem and progenitor cells and used in co-immunoprecipitation (Co-IP) experiments with anti-FLAG-conjugated beads. A library of all detected proteins during erythroid maturation, reticulocytes and RBCs was compiled. This library was used for *in silico* co-folding analysis of EMP3 with the entire spectrum of erythroid proteins utilizing AlphaFold protein co-folding prediction technology.

Results: Using comprehensive proteomics analysis of MAM⁻ versus MAM⁺ RBC samples, we confirmed that CD44 is one of the significantly down-regulated proteins in the RBC proteome. In addition, we identified numerous other proteins that are significantly up- or down-regulated in MAM⁻ RBCs. When analysing RBCs of MAM⁺ individuals using flow cytometry, we found that CD44 is highly correlated to MAM antigen expression levels on RBCs. EMP3-mRNA was significantly upregulated in HEK293 cells when CD44 is overexpressed. The co-IP experiments identified multiple potential EMP3 binding partners, including CD44, LGALS3, LAPTM5 and TFRC. Certain proteins were preferentially precipitated with N-terminally FLAG-tagged EMP3, for example, CD36, indicating interactions at the EMP3

C-terminus. Utilizing AlphaFold2 we found a high co-folding score between EMP3 and CD44 mediated by salt bridges between EMP3 (residues Glu35; Asp42) and CD44 (Arg643). Furthermore, despite stringent conditions, EMP3 was predicted to interact with several other erythroid proteins. However, only two candidate proteins overlapped between the two principal experimental approaches (co-IP and two AlphaFold2 prediction scores), CD44 and ENO1, both expressed early in erythropoiesis.

Summary / Conclusions: We confirm that EMP3 influences CD44 expression in erythroid cells and predict the precise site of protein-protein interaction. Utilizing different *in vitro* and *in silico* experimental approaches, we also found that EMP3 interacts with proteins expressed during early stages of development, suggesting potentially important roles in erythroid biology.

Parallel session—blood products

Transfusion out of hospital and pathogen inactivation

PA27-L01 | Transfusion out of hospital—where are we?

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Introduction: Most transfusions are given to patients treated in hospitals, either during their admission or at day hospitals on an outpatient basis. Most transfusions performed outside the hospital are administered at patients' homes or in nursing homes. Home transfusion (HT) derives from the need to provide continuous care to patients with acute or chronic pathology, because out-of-hospital treatment also has advantages both in reducing complications and in health care costs. Patients may need chronic transfusion, due to fragility or dependency that prevents them from accessing hospitals. HT can be part of a conventional therapy within an out-of-hospitalization approach for acute patients, who are treated at home by hospital specialized teams.

Current situation: It is difficult to know where HT is performed. In a survey (Shaw, Vox Sanguinis, 2021) Canada, Spain and Sweden offered TaD in a structured manner, while Australia, South Africa, Mexico, USA, Brazil, and India offer it occasionally. HT is available other European countries and occasionally in the UK or Japan. It is difficult to know if the patient populations that benefit (e.g., oncology, myelodysplastic syndromes). The difficulties mentioned to implement HT are staff shortages, geographical dispersion, high costs, the lack of monitoring systems, the difficulty in managing data, and maintaining the cold chain to avoid the loss of blood products. Another problem is the absence of standards or regulations for HT, even in countries where HT is used.

Implementation requirements for HT: Key aspects were summarized (Shaw, Transfusion Medicine Reviews, 2021): Which clinical unit is responsible for the program? Who indicates transfusions? How patients and caregivers enrolled and selected for HT? How is an adequate training provided to patients and caregivers? Pre-transfusion collection of samples for crossmatch—what, who, and when? Who transports the units? How is the cold chain maintained? Which components can be transfused? How many transfusions a day to a single patient? Are pre-medications routinely given? Who is present during the transfusion, how long? How is the patient monitored? What are the follow up arrangements after the transfusion? How is emergency medical care delivered? Are there physicians / nurses on call? How and when can they be contacted? How is waste disposed? How are transfusion reactions investigated and reported? How is the transfusion documented? How is HT included in a patient's records?

Conclusion: Home transfusion is growing worldwide, due to its potential to help improve the patients' wellbeing and reduce costs in a changing context, provided it is implemented in a controlled manner with an adequate process design, and an accurate risk and cost analysis. However, still some issues hinder its inception: regulation, technology and safety perception must improve. More research is needed, with improved data gathering by healthcare organisations.

PA27-L02 | Home Transfusion, a viable and secure approach for complex, frail patient population

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Background: Home hospitalization provides specialized care within patient's comfort environment. There is a current interest in decentralizing hospital care to improve patient convenience, minimize costs to the health service, and to prevent nosocomial infections, especially after the COVID-19 pandemic. Although home transfusion (HT) is an increasingly used strategy, safety concerns may be a perceived barrier to its implementation.

Aims: The objective of this study was to assess HT feasibility and safety.

Methods: A HT multidisciplinary protocol was implemented in our hospital during the first COVID-19 pandemic wave and it has been consolidated in subsequent years. To be eligible for HT, all cases were previously evaluated by Blood Bank and Home Hospitalization teams. Inclusion criteria comprised informed consent, stable patient condition, availability of a caregiver, proximity of patient's home to the hospital, and absence of adverse transfusion reactions or erythrocyte alloantibodies in previous transfusions. Cases not

PA27-L02 Table 1: Patient characteristics

Outpatient location-n (%)	Value
Family Home	20 (67.7)
Residential care home or convalescence center	38 (32.2)
Clinically significant transfusion history-n (%)	
Mild or moderate transfusion reaction	3 (2.5)
Red blood cell alloantibodies or weak positive DAT	
At baseline time	5 (4.2)
After the first HT	5 (4.2)

meeting all criteria underwent multidisciplinary assessment considering. Nursing staff remained with the patient during the first 30 minutes of the red blood cell (RBC) infusion and until the end of platelet transfusion (PLT). The caregiver was trained to identify and report alarm signs. We prospectively reviewed data collected from March 2020 to January 2024. We evaluated demographics, clinical and transfusion history, reason for the request, and transfusion adverse side effects.

Results: During the inclusion period we received 437 HT requests. In 83 cases (18.9%), the transfusion was not performed. The main reasons for unacceptance were: unmet inclusion criteria ($n = 21$; 25.3%) and patient finally transferred to a hospital center ($n = 20$; 24.0%). 118 adult patients received 354 transfusions, 602 RBC units, and 9 PLT concentrates. Mean patient age was 85 years-old (range: 22–99) with 60.1% were females. Regarding comorbidities, 60 patients (50.9%) had cancer, 54 (45.8%) heart disease and 29 (24.6%) dementia. 92.3% of the cases presented more than 5 points in the Comorbidity Charlson Index. Other characteristics of the transfused patients are shown in Table 1. 42 patients (35.6%) received more than one transfusion and in most cases (64 %) 2 RBC units were administered per day. Given the high risk of TACO, with more than 2 risk factors in 54.6% of the studied population, in 312 (88.1%) transfusions, preventive diuretic administration was considered. Regarding adverse reaction incidences, a single potential side effect was made aware, which was the worsening of a previous heart failure symptomatology in a palliative care patient.

Summary / Conclusions: Home transfusion is feasible and safe even in frail complex patients if performed on selected cases and under specific multidisciplinary protocols and assessment.

PA27-L03 | Quantifying residual red blood cells in platelet and plasma components—flow cytometry and a visual inspection tool support implementation of pathogen inactivation

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Background: Platelet concentrates (PC) and plasma components may contain small numbers of residual RBCs (rRBCs), an important

consideration for use of pathogen inactivation (PI) technologies. The INTERCEPT PI blood system (Cerus Corporation) requires that input PC and plasma components have rRBC counts $< 4 \times 10^6/\text{mL}$. However, enumeration of rRBC counts can be challenging and there is no consensus method. To support Canadian Blood Services' implementation of the INTERCEPT blood system to produce apheresis and pooled (whole blood (WB)-derived) platelets, psoralen treated, we developed a flow cytometry assay to count rRBCs and an in-house visual inspection tool (VIT) to assess suitability of platelet components for INTERCEPT treatment.

Aims: To describe a flow cytometry assay and a method for development of a visual inspection tool to assess rRBCs levels in non-RBC blood components prior to pathogen inactivation using INTERCEPT.

Methods: To count rRBCs, we developed a flow cytometry protocol based on the BD Leucocount Kit. The approach used BD Trucount™ Tubes, which contain a known number of fluorescent beads, as an internal reference to determine absolute cell counts. PCs (apheresis or pooled in 40% plasma/60% SSP+) or plasma (apheresis or WB-derived) were diluted $10\times$ in PBS and added ($20 \mu\text{L}$) to Trucount™ tubes. RBCs were labelled with CD235a (Glycophorin A)-PE for 30 min in the dark at room temperature. Samples were diluted with 1 mL PBS, vortexed and acquired (stopping collection: 2500 beads) on a BD FACS Canto II running FACSDiva Software version 8.0.1. A gating strategy based on glycophorin-A positive events was applied consistently across samples to ensure only RBCs were counted. To develop a visual inspection tool, platelet components were spiked with known RBC concentrations from an ABO-matched red cell concentrate (1×10^6 rRBC/mL; 4×10^6 rRBC/mL; and 6×10^6 rRBC/mL), photographed and colour-true prints were generated. The visual inspection tool was validated using PCs spiked with known rRBC counts. These PCs were visually assessed next to the VIT by a minimum of 10 users to determine pass or fail against the 4×10^6 rRBC/mL limit.

Results: Serial dilutions of plasma or PCs demonstrated linearity of the flow cytometry assay up to 1500 RBC events. At counts greater than 1500, the assay undercounted rRBC. Within- and across-run measurements of assay precision resulted in intra-assay coefficients of variation (CVs) ranging from 2.55% to 3.36% ($n = 5$ samples; 9 runs on each) and inter-assay CVs ranging from 2.02% to 5.12% ($n = 5$ replicate samples counted in independent runs). Spiking apheresis PCs with known concentrations of RBCs between 0 and 6×10^6 rRBC/mL demonstrated the assay's accuracy (correlation coefficient 0.9997). PCs spiked with known RBC counts were prepared and photographed to develop the visual inspection tool. Validation of the tool by multiple users confirmed its utility to distinguish whether spiked units passed or failed against the 4×10^6 rRBC/mL limit.

Summary / Conclusions: The flow cytometry assay was found to be suitable for counting low numbers of rRBCs in platelet and plasma components. The visual inspection tool was successfully deployed and is being used operationally at Canadian Blood Services to determine the suitability of platelet components intended for INTERCEPT treatment.

PA27-L04 | Effect of pathogen reduction using riboflavin and UV light, subsequent cryopreservation and thawing on red blood cells and platelets concentrates

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Background: Pathogen reduction technology (PRT) may improve the safety of RBCs for transfusion and also PRT is increasingly used in the preparation of platelets for therapeutic transfusion.

Aims: This work describes the effect of PRT on the recovery and function of cryopreserved platelets and erythrocytes after thawing. The study was divided into two parts, the first part describing the cryopreservation of erythrocytes obtained by collecting whole blood, which was treated with riboflavin and UV light before being processed into erythrocytes. The second part of the study describes the cryopreservation of platelets obtained by apheresis and treated with riboflavin and UV light.

Methods: In the first part of the study, 24 Group O whole blood (WB) transfusion units (T.U.) were treated with PRT before cryopreservation; 20 similarly-collected units were untreated controls. All T.U. were subsequently processed into erythrocytes, then cryopreserved with 40% glycerol (wt/vol), frozen at -80°C , and long-term stored. After reconstitution with deglycerolization, the erythrocyte T.U. were resuspended in AS-3 and stored at $4 \pm 2^\circ\text{C}$ for 21 days. Erythrocytes were sampled before PRT, after PRT and further after thawing on days 0, 7, 14 and 21. The following measurements were taken from the collected samples: hematocrit, volume, hemoglobin per unit, pH, % hemolysis, hemoglobin in the supernatant, potassium, phosphorus, NH_3 , osmolality, ATP, and 2,3-DPG. The findings show that cryopreserved erythrocytes made from Riboflavin and UV light-treated fresh whole blood, meet the criteria for clinical use and provide additional protection against infectious threats during long-term storage. In the second part of the study, 16 Group O apheresis platelets transfusion units (T.D.) were treated with PRT before freezing; 15 similarly collected T.D. of trombocytes were frozen without PRT as controls. 5%–6% DMSO was added to all units and then the supernatant was

removed, then frozen at -80°C , stored for 14 days and then reconstituted in thawed AB plasma. After reconstitution, all units were assessed for: platelet count, MPV, platelet recovery, thromboelastography, thrombin generation time, endogenous thrombin potential, glucose, lactate, pH, pO₂, pCO₂, HCO₃, CD41, CD42b, CD62, Annexin V, CCL5, CD62P, Kunicki score, and aggregates $>2\text{ mm}$.

Results: PRT with leukoreduction caused a 5% loss of RBC followed by a 24% freeze-thaw-wash related loss for a total 28% loss but treated units contained an average of 45 g of hemoglobin, meeting European Union guidelines for cryopreserved RBC. Treated cryopreserved RBCs displayed higher post-wash hemolysis, potassium, and ammonia concentrations, and lower ATP at the end of storage. PRT treated platelet T.D. had lower platelet number ($247\text{ vs. }278 \times 10^9/\text{U}$), reduced thromboelastographic MA (38 vs. 62 mm) and demonstrated aggregates in all units.

Summary / Conclusions: Cryopreserved RBCs from Riboflavin and UV light-treated WB meet the criteria for clinical use for 7 days after thawing and provide additional protection against infectious threats. Cryopreserved platelets from apheresis platelets treated with riboflavin and UV light showed reduced platelet number, reduced function greater than the reduced number would cause, and aggregates. While the platelet numbers are sufficient to meet the European standard, marked platelets activation with weak clot strength suggest reduced effectiveness.

PA27-L05 | Impact of no agitation on buffy coat platelet concentrates during reduction of residual amotosalen (CAD) after pathogen inactivation

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Background: The process of pathogen inactivation in platelet concentrates (PC) (INTERCEPT® Blood System, Cerus) uses the principle of photoreaction of amotosalen to UVA illumination. It has a Compound Adsorption Device (CAD) that reduces the residual amotosalen contained in the PC after illumination. This stage, the aim of which is to obtain a final concentration of amotosalen under $7.5\text{ }\mu\text{M}$ in the PC, requires a period of agitation between 6 and 16 h.

Aims: To assess the quality of Buffy Coat-PC (BC-PC) without any agitation during CAD and stored until 7 days in order to determine what measures to take in case of agitation failure during CAD.

Methods: 24 buffy coats were pooled and split in order to obtain 3 identical BC-PC ($n = 12$). They were pathogen inactivated with INT250 DS process. CAD was performed during 16 h in 3 conditions: A: control with agitation. B: without agitation. C: without agitation for 16 h followed by 6 h of agitation. Residual amotosalen was measured

after CAD (D2) and the following parameters were analyzed after 2, 3, 4 and 7 days of storage: swirling, platelet count, pH, pO₂, pCO₂, glucose, LDH, soluble p-selectin.

Results: Residual amotosalen post-CAD was superior to $7\text{ }\mu\text{M}$ only in B condition with no agitation. However, the condition C had similar results to control condition with values far under $7\text{ }\mu\text{M}$. During PC storage, platelet count, swirling (always +++) and blood gases were similar to all conditions while LDH concentration was lower in conditions B and C compared to control condition reflecting a slower platelet lysis without agitation. The same profile was observed with p-selectin, a platelet activation parameter. Concerning metabolism parameters, we observed a lower level of pH (but still >6.4) at D2, D3 and D4 and higher glucose consumption in conditions B and C. This may be due to lower oxygen availability without agitation during CAD. Soluble p-selectin level was similar in all condition indicating a moderate platelet activation according time.

Summary / Conclusions: In case of PC agitation failure during CAD, 6 h of additional agitation has a positive impact on the residual amotosalen concentration while maintaining the other biological parameters compliant for the delivery of CPs.

Parallel session—blood safety

Revisiting the big three viral TTI

PA28-L01-01 | Tenofovir Measurement in Whole Blood and Plasma: Implications for PrEP Monitoring in Blood Safety

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Background: Tenofovir (TFV) prodrugs, tenofovir disoproxil fumarate (TDF), and tenofovir alafenamide (TAF), combined with emtricitabine (FTC), are widely used for oral HIV pre-exposure prophylaxis (PrEP). Despite TDF and TAF having different pharmacokinetic profiles, in both instances, TFV is intracellularly metabolized into tenofovir diphosphate (TFV-DP), which accumulates in cells, including red blood cells. With increasing availability of PrEP, there is interest in the utility of TFV testing to assess undisclosed PrEP use in blood donor samples. Although PrEP breakthrough HIV infections have not yet been reported in blood donors, they pose a

PA28-L01-01 - Table: Whole Blood and Plasma OCT-ELISA detection rates according to participants' self-reported days since the last dose

	Today	1 - 2 days	3 - 7 days	8 - 14 days
OCT-ELISA	n = 460	n = 229	n = 22	n = 16
Whole Blood, n (%)				
Detected	444 (97%)	223 (97%)	18 (82%)	13 (81%)
Not detected	16 (3.5%)	6 (2.6%)	4 (18%)	3 (19%)
Plasma, n (%)				
Detected	422 (94%)	199 (88%)	11 (52%)	6 (38%)
Not detected	27 (6.0%)	27 (12%)	10 (48%)	10 (63%)
Not tested	11	3	1	0

potential risk to the safety of the blood supply if blood screening assays fail to detect these infections due to low-level viremia or blunted seroconversion.

Aims: Evaluate the performance of a novel enzyme-linked immunosorbent assay (ELISA) method for detecting TFV in whole blood (WB) and plasma, considering the timing of the last dose and different PrEP drug regimens.

Methods: WB and plasma samples from a cross-sectional behavioral and biomarker study (known as ADVANCE) of sexually active men who have sex with men (MSM), interested in donating blood, from 8 U.S. metropolitan areas, were analyzed using the OraSure Colorimetric TFV (OCT) ELISA. The lower limit of detection is between 50-100 ng/mL of TFV (from TFV and cleaved intracellular TFV-DP) in WB and 25-50 ng/mL of TFV in plasma.

Results: Of 1,548 HIV-negative participants, 743 (48%) reported oral PrEP use in the previous month. The WB assay accurately identified 95% of these PrEP users, while the plasma assay identified 88%. Detection rates for WB and plasma decreased as the days since the last PrEP dose increased. WB detection remained good up to 14 days post-dose, while plasma detection decreased starting 2 days post-dose (Table). Receiver operating characteristics curve showed an area under the curve of 0.96 (WB) and 0.88 (plasma). Participants on TDF/FTC vs. TAF/FTC who took PrEP in the last 2 days had higher detection rates (99% vs 93% in WB, 98% vs 86% in plasma) and significantly higher percentage of inhibition ($p < 0.001$). In this first-generation assay, the specificity of the TFV ELISA was not ideal (80% for WB; 66% for plasma).

Summary / Conclusions: Superior detection of TFV using WB due to the longer intracellular half-life of TFV-DP compared with the parent drug in plasma was evident in our study. The performance of this immunoassay using WB demonstrates its value as a tool for measuring TFV as part of PrEP regimens among those who have blood collected as part of HIV infection testing. In communities with high PrEP uptake, the OCT-ELISA could be used to monitor community-level oral PrEP use more affordably. In communities with low PrEP uptake, such as among blood donors, use of the WB OCT-ELISA presents a promising avenue for quantifying undisclosed oral PrEP use.

PA28-L01-02 | Residual risk of transfusion-transmitted HIV in the US during different eligibility policy periods before the adoption of individual donor assessment

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Background: From 1985 to 2016, US blood collection organizations (BCOs) indefinitely deferred male donors who reported sex with other men (MSM) since 1977. Following revised FDA guidance, in 2016 BCOs implemented a 12-month MSM deferral policy (MSM12m) and in 2020 a 3-month MSM deferral policy (MSM3m). In 2023, individual donor assessment (IDA) was implemented in the US. The TTIMS program monitors HIV prevalence, incidence and transfusion-transmitted (TT) residual risk in the US by collating donor-donation data and conducting additional laboratory testing on infection-reactive samples from four major BCOs representing about 60% of the US blood supply. Here, we evaluate TT-HIV residual risk (RR) in each of the three major time-based MSM deferral policy periods before IDA implementation.

Aims: To assess changes in TT-HIV RR in the US following MSM donor eligibility policy changes compared to the policy of indefinite deferral.

Methods: HIV incidence was estimated in first-time donors (FTD) and repeat donors (RD) during the 15 months before MSM12m implementation, the MSM12m period, and the MSM3m period. HIV incidence was estimated in FTD and RD using biomarkers of recent infection and classic methods, respectively. Residual risk was estimated based on the average of FTD and RD incidence, weighted by the number of collections from each group. We estimated the infectious window period (IWP) using a previously-described model (Grebe et al, Blood 2020), incorporating viral ramp-up dynamics, transmissibility as a function of inoculum size, and viral concentration dependent NAT sensitivity (in minipool 16 format). In this model, the IWP depends on the volume of plasma transfused, so separate estimates are reported for red blood cell (RBC) and fresh frozen plasma (FFP) products. Uncertainty in IWP and residual risk estimates were evaluated by bootstrapping from distributions of input parameters including incidence estimates.

Results: The infectious window periods for RBC and FFP products were estimated at 4.49 and 7.33 days, respectively. TT-HIV RR for RBC products was estimated at 1 in 3.65 million transfusions during the indefinite deferral period, declining to 1:5.01m in the MSM12m period, and to 1:8.63m in the MSM3m period (95% CIs shown in table). For FFP products, TT-HIV RR was higher, owing to the greater plasma volume for an FFP product, and was estimated at 1:2.24m, 1:3.07m and 1:5.29m in the three periods, respectively. The

differences between the MSM3m and indefinite deferral periods were statistically significant.

PA28-L01-02 - Table 1

	RBC estimate (95% CI)	FFP estimate (95% CI)
IWP (days)	4.49 (3.56,6.90)	7.33 (6.38,9.48)
Indefinite deferral period	1:3.65m (4.26m,1.89m)	1:2.24m (2.44m,1.34m)
MSM12m period	1:5.01m (6.03m,2.90m)	1:3.07m (3.42m,2.06m)
MSM3m period	1:8.63m (10.17m,4.63m)	1:5.29m (5.82m,3.28m)

Summary / Conclusions: Residual risk of TT-HIV in the US has declined continuously despite expansion of donor eligibility, indicating that the policy changes have had no adverse impact on TT-HIV risk. The TT-HIV RRs are very low, specifically, less than 1 transmission in 8 million RBC transfusions and less than 1 in 5 million FFP transfusions in the MSM3m period. These findings are reassuring and help to support the adoption of IDA in the US.

PA28-L01-03 | Blunted HIV detectability using contemporary donor screening serological and nucleic acid assays following early antiretroviral treatment initiation

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Background: Initiation of antiretroviral therapy (ART) during acute or very early stages of HIV infection results in undetectable HIV RNA and p24 antigen (Ag) and suppressed antigenic stimuli that may lead to blunted antibody (Ab) responses. A reduced ability to identify such infections using nucleic acid-amplification testing (NAT) and Ag/Ab serological assays may compromise blood transfusion safety.

Aims: We used samples from two early ART initiation studies to investigate the altered dynamics of HIV RNA detectability and sero-conversion using FDA-licensed blood donor screening NAT and Ag/Ab combo assays.

Methods: RV254 (RV254/SEARCH 010) and SIPP (SeroIncidence Panel Project) identified participants with acute or recent HIV infection in Bangkok and San Francisco, with early ART initiation and serial sample collections. From RV254, we obtained samples from the date of ART initiation (visit 1), and from visits conducted 12 and 24 weeks afterwards (visits 2 and 3). From SIPP, we obtained samples collected at a median of 10 visits after ART initiation (median follow-up 394 days). Samples were tested using cobas® MPX Test for NAT detection, and three Ag/Ab assays (Abbott Alinity s HIV Ag/Ab Combo; Elecsys® HIV Duo; and Ortho VITROS HIV Combo). We present percentages and 95% confidence intervals (CI) of reactive results for RV254 participants initiating ART at Fiebig stages 1&2 (prior to Ab detection) or 3&4 (initial days following seroconversion).

Results: Among 55 RV254 participants, 25 initiated ART at Fiebig stages 1/2, and 30 at Fiebig stages 3/4. Reactivity in Ag/Ab assays was <100% at all timepoints, and lower among those initiating ART at Fiebig stages 1/2 (Table). Seroreversion (nonreactive tests in samples from participants with previously reactive results on the same assay) were observed in 16 RV254 participants (29%, 95% CI 18-43%). For SIPP participants, who started ART at later timepoints (2 participants at Fiebig 4; 15 at Fiebig 5), Ag/Ab assays identified all infections with no evidence of reactivity waning over time.

To investigate if blood screening tests might fail to detect HIV infection in RV254 participants, we identified samples with nonreactive results in NAT and Ag/Ab assays. Blood donation screening using these assays would have missed 7 cases at visit 2, of whom 6 cases would also have been missed at visit 3, regardless of the Ag/Ab test adopted for testing.

PA28-L01-03 - Table 1

	Visit 1 - ART initiation	Visit 2 - 12 weeks	Visit 3 - 24 weeks
cobas® MPX Test			
Fiebig 1/2	100% (86-100)	29% (13-51)	32% (15-54)
Fiebig 3/4	100% (88-100)	67% (47-83)	57% (37-75)
Abbott Alinity			
Fiebig 1/2	64% (43-82)	32% (15-54)	44% (24-65)
Fiebig 3/4	86% (68-96)	76% (57-90)	80% (61-92)
Elecsys® HIV Duo			
Fiebig 1/2	60% (39-79)	64% (43-82)	72% (51-88)
Fiebig 3/4	80% (61-92)	90% (74-98)	90% (74-98)
Ortho VITROS			
Fiebig 1/2	60% (39-79)	76% (55-91)	76% (55-91)
Fiebig 3/4	97% (82-100)	97% (83-100)	97% (83-100)

Summary / Conclusions: Our findings suggest reduced HIV detectability due to very early ART initiation, particularly for ART initiation at or before Fiebig stage 4. For blood transfusion services, surveillance and further research are needed to: 1) evaluate the frequency of donations from persons with HIV who initiated ART during acute HIV infection in different settings; 2) understand the infectivity of plasma RNA- and Ab-negative donations; and 3) determine the need for novel screening algorithms, including modifications in the donation interview, more sensitive NAT and Ag/Ab tests, or antiretroviral testing of donation samples.

PA28-L02 | Assessment of HIV prevalence and incidence among blood donors in five Brazilian blood centers before and after the discontinuation of deferral policies for men who have sex with men

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Background: The time-based blood donation deferral policies for men who have sex with men (MSM) have been replaced by individual risk assessment regardless of gender or sexual orientation in many countries, including Brazil.

Aims: To compare HIV prevalence, markers of recent infection, and incidence among blood donors before and after the withdrawal of the time-based deferral policy for MSM across five public blood collection organizations (BCOs) located in São Paulo, Belo Horizonte, Rio de Janeiro, Recife, and Manaus.

Methods: Donations at BCOs are screened for HIV using 4th generation HIV antigen/antibody assays and HIV-1 RNA using a minipool-6 NAT assay (Bio-Manguinhos, Brazil), followed by pool resolution. The Recipient Epidemiology and Donor Evaluation Study-IV-Pediatric (REDS-IV-P) Brazil component collects blood donor screening data. Covering the period January 2017 to June 2023, we report the frequencies, rates per 10⁵, and 95% confidence intervals (CI) of confirmed HIV-positive donations (NAT-reactive and serology reactive or indeterminate) among first-time donors (FTD), and the HIV NAT-yield (serology nonreactive) rates for FTD and repeat donors (RD) for more than a 3-year period before and 3-year period after the policy change. We also report incidence of confirmed HIV per 10⁵ person-years before and after the policy change among RD and results from multivariable regression analysis to assess factors associated with prevalence and incidence.

Results: In May and June 2020, the BCOs modified their donor history questionnaire to align with changes in the donor selection policy.

PA28-L02 – Table 1

Blood Center	Before		After	
	FTD	RD	FTD	RD
Overall	3.2 (15)	3.9 (33)	2.1 (9)	1.2 (10)
Hemope, Recife	2.8 (3)	4.4 (11)	2.0 (2)	1.5 (3)
Hemominas, Belo Horizonte	1.4 (1)	0	0	0
FPS, São Paulo	2.9 (4)	1.2 (3)	1.6 (2)	0.9 (2)
Hemorio, Rio de Janeiro	5.7 (7)	12.7 (19)	0.9 (1)	2.7 (4)
Hemoam, Manaus	0	0	8.1 (4)	0.8 (1)

The overall prevalence of confirmed HIV infection in FTD was 72/10⁵ (95% CI 64-79) before and 94/10⁵ (95% CI 84-103) after the policy change. Differences in HIV prevalence among first-time male donors, particularly in younger age groups, and variability between blood centers, with notable increases in Manaus, were observed. The HIV NAT-yield rates significantly decreased after the policy change for RD ($p = 0.001$), while no change was observed for FTD ($p = 0.41$); variability was evident between blood centers (Table). Before the policy change, HIV incidence in RD was 14.54/10⁵ person-years (95% CI 13.04-16.21); after the policy change, it was 13.54/10⁵ person-years (95% CI 12.08-15.17). In regressions adjusted for sex, age, race, education, donation type, and BCO, FTD had 1.21 times the prevalence of HIV after the policy change compared to before (95% CI 1.04-1.40, $p = 0.015$) and RD had 0.93 times the incidence of HIV after the policy change compared to before (95% CI 0.79-1.09, $p = 0.37$).

Summary / Conclusions: After withdrawing the MSM deferral policy, HIV incidence among RD remained stable, while HIV prevalence increased among FTD. No evidence of significant increases in NAT-yield donations for both RD and FTD were observed. Together these findings suggest residual risk of transfusion-transmitted HIV has not increased. As in other large countries, regional differences in HIV infection rates highlight complexities in the HIV epidemic in Brazil which are reflected in the donor population before and after the policy change.

PA28-L03 | For the Assessment of Individualised Risk (FAIR)—low numbers of HIV and good reported compliance suggests maintenance of lower risk donor pool under FAIR in England

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Background: In England, since June 2021, blood donation eligibility is based on an assessment of individualised risk (FAIR). The number of

PA28-L03 Table 1: HIV confirmed positive donors in England

	3-month deferral 2018 to 2020	FAIR 2021 to 2023*
Total n (recent including seroconversion)	21 (4)	22(4)
per 100,000 donations	0.46	0.48
First-time n (recent)	18 (3)	19 (1)
per 100,000 donations	4.24	4.97
Repeat within 3 years (seroconverter)	3 (1)	3 (3)*
per 100,000 donations	0.07	0.07
Gender		
Male	16	17
Female	5	5
Median age (range)	37 (19-60)	36 (21-62)
Reported source (non-compliant)		
Sex between men (SBM)	3	8
Sex between men and women (SBMW)	13	10
SBMW and partner may have had sex in sub-Saharan Africa	2 (1)	0
Other blood contact	1	1
Source not known	2	3 (1)
Dual HIV syphilis case	2	1

* Results for 2023 are preliminary and include a repeat donor with 13-month donation interval, avidity awaited.

people estimated to be living with undiagnosed HIV in the general UK population was estimated to be low and declining, key to the evidence-based recommendations to introduce FAIR. The pre-donation form now includes questions to all donors about syphilis and gonorrhoea, drug use during sex, and anal sex with new or multiple partners within 3 months; with syphilis deferral in place before FAIR. This has removed the 3-month deferral for some gay and bisexual men who have sex with men (GBMSM) and also for women with male partners who had sex between men (SBM). With this suite of questions in place, the selection question for donors with partners who may have had sex in an HIV-endemic area which in practice was applied to the area of sub-Saharan Africa was also removed from December 2021.

Aims: To describe the impact of the FAIR policy on rates of HIV, sources of HIV and compliance in HIV positive blood donors in England in the post-implementation period compared to the period when 3-month deferral for sex between men was in place.

Methods: Data on number of donations screened for HIV antibody/antigen and NAT and confirmed positive were extracted from the routine surveillance database. Details included reported source of HIV, partners and compliance from the clinical team completing pro-forma based on post-test discussions. Seroconversion was defined as confirmed HIV in a repeat donor with either a negative donation within

12 months or avidity indicating recency and negative donation within 3 years. HIV rates were calculated per 100,000 donations.

Results: The number and rate of HIV were similar for both policy periods (Table 1). No NAT yield donations were seen for the 6-year period in over 9 million donations. Four recent HIV cases were seen under both policies, 3 had seroconverted under FAIR, all 3 reported sex between men and women and appeared compliant but one did not fully engage. Under FAIR, 8 donors reported SBM, all were compliant, 2 were newly eligible, none had recent HIV. One donor was clearly non-compliant to FAIR but did not engage.

Summary / Conclusions: The number and rate of HIV positive donations were low with a 3-month deferral and remained low under FAIR. Few seroconversions were seen, unrelated to introduction of FAIR. Additionally, lack of NAT yield, low number of dual syphilis cases and good reported compliance suggests maintenance of a low risk donor pool under FAIR. However, under both donor selection policies there are a small number of donors who do not fully engage, and we need to continue to encourage appropriate self-deferral or full disclosure.

PA28-L04 | Hepatitis C epidemiology in Canadian blood donors—insight into the low-risk undiagnosed population

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Background: Increased diversity of the donor base is desirable to ensure rare red cell phenotypes more common in certain ethnic groups are available. Hepatitis C is a blood-borne infection caused by the hepatitis C virus (HCV) that can progress to cirrhosis and liver cancer. About 70% of infections become chronic. Individuals presenting with both anti-HCV antibodies and HCV RNA might represent a late acute/chronic infection (LACI). Direct acting antiviral treatment has been available since 2014 initially based on disease stage, and in 2018 for all infected. While the United States recommends all adults be tested once for HCV, in Canada clinical screening is risk-based. About 1% of Canadians have had hepatitis C, with 0.5% chronically infected (about 25% are unaware). Higher chronic infection prevalence is associated with birth between 1945 and 1964, intravenous drug use and birth in a high prevalence country. Blood donors are in good general health and deferred for risk behaviours such as intravenous drug use and sexual risks. Blood donor data may be important for public health to provide insight into the low-risk undiagnosed population.

Aims: We aimed to describe HCV epidemiology in first-time blood donors over 28 years of monitoring.

Methods: All first-time blood donors in all provinces except Quebec (1993 to 2021) were included. All donors were tested for anti-HCV. Since late 1999 all were also tested for HCV NAT. Donors both anti-HCV and HCV NAT positive were assumed to have LACI. The Pampalon Material Deprivation and the CanMarg Ethnocultural indices were

based on the donor's residential postal code. Separate logistic regression models were fitted with anti-HCV positivity and LACI as the dependent variables. Independent variables were year, age cohort (born before 1945, 1945–1964, 1965 or later), sex, region, material deprivation and ethnocultural quintiles. The quarterly percentages of HCV positive donations also HCV NAT positive were calculated. An ARIMA model was fitted with interruption in 2014.

Results: There were 2,334,238 first-time donors from 1993 to 2021. Prevalence for anti-HCV 0.33% (0.30, 0.37) in 1993 and 0.07% (0.05, 0.09) in 2021. In 2021 0.03% (0.01, 0.04) had LACI HCV. Predictors for both anti-HCV positivity (with or without HCV NAT) and LACI were similar. For LACI predictors were male sex (OR 1.8, 1.6, 1.9), birth between 1945 and 1964 (OR 7.0, 6.2, 7.8), living in the western provinces and living in material deprived (OR 2.7, 2.2, 3.2) and more ethnocultural concentrated neighbourhoods (OR 1.7, 1.4, 2.1). The ARIMA model showed modest decline prior to 2014, with greater decline afterwards ($p < 0.0001$).

Summary / Conclusions: Since 2014 the percentage of anti-HCV positive donors who were also HCV NAT reactive (LACI) has declined in blood donors. Blood donors LACI prevalence is 16 times lower than the general population in keeping with deferral for high-risk behaviour. The decline in the proportion of LACI donors after 2014 may be related to introduction of direct acting antivirals. Donors largely mirror population trends and highlight the ongoing prevalence of untreated infections in groups without obvious risk factors missed by risk-based clinical screening. Higher proportions of donors in ethnoculturally concentrated neighbourhoods may suggest that HCV infections rates could increase as a more ethnically diverse donors are recruited.

PA28-L05 | Occult hepatitis B among blood donors and associated transfusion transmitted infections in England, 2009–2023

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Background: Occult hepatitis B infection (OBI) is a form of chronic hepatitis B virus (HBV) infection where an individual has HBV DNA present in the hepatocytes but at low and fluctuating levels in the blood. They also typically have undetectable levels of HBsAg. These two factors mean that it is difficult to detect donors with OBI presenting a risk for transfusion transmission to occur.

Aims: Our aim was to characterise blood donors with OBI and to describe HBV transmissions which have led to an identification of further blood donors with OBI, identified in England before and after anti-HBc screening.

Methods: We have collated the data on OBI cases and associated HBV transmissions in England, covering the period from the introduction of HBV DNA for blood donation screening in 2009 (in pools of 24) to the full implementation of individual anti-HBc testing of all previously non-tested or new donors in 2023.

Results: During the 15-year study period, 53 cases of OBI have been detected in blood donors of which most were males ($n = 41$, 77%), over the age of 45 years ($n = 42$, 79%), repeat donors ($n = 30$, 57%) and linked to an HBV endemic country ($n = 45$, 85%). Prior to anti-HBc implementation, 41 donors with OBI were identified over a 13-year period (average 2.7 cases per year; ranging from 2 to 5 per year), 5 of them through post-transfusion investigations. Four of the 5 HBV transmissions were reported for investigation as the recipient had developed acute hepatitis without other source identified, whereas the remaining was triggered by identification of a new HBV infection in dialysis patient. All patients developed chronic HBV infection, including one identified via subsequent lookback and one of them died. Five recipients were infected by red cell transfusion and one from fresh frozen plasma. All were linked to donors with OBI; one transmission involving two recipients was confirmed via sequence analysis. Comparison of virological properties of OBI donations which led to a transmission to those where no evidence of transmission will also be presented.

Summary / Conclusions: Unidentified occult HBV infection among blood donors represents a risk for HBV transmission to recipient even in high-income countries with low incidence of HBV, often leading to a chronic, life-long infection, if not to a fulminant outcome. Anti-HBc screening is an effective way of reducing that risk; over 4× more blood donors with OBI have been identified and removed from a donor panel via the anti-HBc screening (10 in 2023) than previously found by HBV DNA screening. Furthermore, no cases of HBV transmission via blood transfusion have been identified since the implementation of anti-HBc screening in England.

Parallel session—donors and donation

Getting to know you and your health

PA29-L01 | Modelling personalised inter-donation intervals for post-donation testing

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Background: Deferrals due to low haemoglobin are time-consuming and costly for blood donors and donation services. Furthermore, accepting donations from those with low haemoglobin could represent a significant safety issue. One approach to reduce them is to use haemoglobin concentration alongside donor characteristics to inform personalised inter-donation intervals.

Methods: We developed a discrete event simulation model comparing personalised inter-donation intervals using “post-donation” testing (PDT; i.e., estimating current haemoglobin from that measured by a haematology analyser at last donation) versus the current approach in England. The model was informed by data from 17,308 participants in the COMPARE study, a large study of blood donors in England, originally designed to compare different methods to measure haemoglobin concentrations. We assessed five different strategies: Current strategy in England: pre-donation testing with fixed inter-donation intervals (12 weeks for men, 16 weeks for women). PDT with personalised inter-donation intervals based on medium ($\geq 70\%$) certainty of being over the regulatory threshold to donate, plus an adaption adding on-session testing for those with lower certainty. PDT with personalised inter-donation intervals based on high certainty ($\geq 90\%$) of being over the regulatory threshold to donate, plus an adaptation allowing early return for those with higher certainty. Personalised inter-donation intervals were defined using mixed-effects modelling to estimate haemoglobin trajectories and the probability of crossing regulatory haemoglobin thresholds for donation. The simulation for each strategy was based on 1000 hypothetical donors drawn from the COMPARE population, tracked for 1 year. We then estimated the impact of each strategy on total donations, low haemoglobin deferrals, donations made below the regulatory threshold, and blood service costs.

Results: The model had generally good internal validation, with predicted events similar to those observed. Over 1 year, a personalized strategy requiring high probability of being over the haemoglobin threshold, minimized adverse events (low haemoglobin deferrals and

below-threshold donations) in both sexes and costs in women. Donations per adverse event improved from 3.4 (95% uncertainty interval 2.8, 3.7) under the current strategy to 14.8 (11.6, 19.2) in women, and from 7.1 (6.1, 8.5) to 26.9 (20.8, 42.6) in men. In comparison, a strategy incorporating early returns for those with high certainty of being over the threshold maximized total donations in both men and women, but was less favourable in terms of adverse events, with 8.4 donations per adverse event in women (7.0, 10.1) and 14.8 (12.1, 21.0) in men.

Discussion: Personalised inter-donation intervals using post-donation testing combined with modelling of haemoglobin trajectories can help reduce deferrals, below-threshold donations, and costs.

PA29-L02 | Long-term risk of lymphoma and autoimmune disease following red-cell transfusion in childbirth—a Swedish nationwide cohort study

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Background: Blood transfusions introduce foreign cells into the recipient, which induce immunological responses. These immunological responses have been speculated to increase the risk of manifest immunological disorders, such as non-Hodgkin lymphoma (NHL) and autoimmune disease. Studies of these diseases in transfusion recipients are methodologically challenging and results have been ambiguous. Women transfused during childbirth constitute a distinct group of recipients, who are typically younger and healthier than other transfused patient groups. This offers an opportunity to study associations between red-cell transfusion and future adverse immunologic outcomes, with less confounding by indication.

Aims: Assessing the long-term risk of lymphoma or autoimmune disease in Swedish women who received red-cell transfusions during childbirth.

Methods: Using the SCANDAT-3S database, we formed a cohort of women aged 18–50 years with a first and any subsequent registered birth between 1987 and 2017, excluding women with histories of autoimmune disease, lymphoma, or previous blood transfusions. We used Cox regression to evaluate the risk of NHL, rheumatoid arthritis (RA), systemic lupus erythematosus (SLE), and systemic sclerosis, in association to red-cell transfusion at child delivery. To circumvent surveillance bias and reverse confounding, follow-up commenced 180 days post-delivery. Adjustments were made for time-varying and

PA29-L02 - Table 1: Events per person-years, adjusted cause-specific hazard ratios and risk differences for diagnosis of non-Hodgkin lymphoma or autoimmune disease in relation to red-cell transfusion in delivery, using a 180 days latency in follow-up.

	Transfusion in childbirth	Events / person-years	Adjusted HR* exposed / non-exposed (95% CI)	Risk differences at 25 years of follow-up (95 % CI)
Non-Hodgkin lymphoma	Yes	17 / 420,460	0.86 (0.53-1.40)	−0.02% (0.10%–0.05%)
	No	540 / 11,805,084		
Systemic lupus erythematosus	Yes	43 / 420,460	1.38 (1.01-1.87)	0.13% (0.00%–0.26%)
	No	824 / 11,805,084		
Systemic sclerosis	Yes	15 / 420,460	1.89 (1.12-3.21)	0.06% (0.00%–0.12%)
	No	208 / 11,805,084		
Rheumatoid arthritis	Yes	185 / 420,460	1.07 (0.92-1.24)	0.16% (−0.08–0.39%)

constant covariates. We calculated disease incidence as a function of time since last delivery, to determine absolute risk.

Results: The study included 1,999,013 registered births by 1,043,713 women, with 4.1% having had red-cell transfusion therapy during any delivery. Main results are displayed in table 1. The time to diagnosis averaged around 10 years following the last birth for all outcomes. Sensitivity analyses, incorporating factors like calendar period, start-of-follow-up modifications, general blood product transfusion, number of transfused units and gestational factors did not significantly alter the results.

Summary / Conclusions: In this study with extensive follow-up, women who received red-cell transfusion during childbirth were at an increased risk of SLE and systemic sclerosis compared to non-transfused women. This association persisted across unadjusted, adjusted, and sensitivity analyses. No evidence of such associations was observed for NHL and RA.

PA29-L03 | Characterizing viral profiles in eligible plasma—insights from blood donor biorepository samples metagenomics

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Background: Blood donated by volunteers undergoes rigorous screening for pathogens like HBV, HCV, HIV, and Treponema pallidum. Yet, despite these measures, blood products may still harbor microbes, including both established endemic pathogens and those associated with emerging infectious diseases. As representatives of the general population, blood donors offer valuable insights for large-scale epidemiological studies. Recently, there's been a growing recognition of the significance of utilizing stored blood donor biorepository samples. These samples are proving instrumental in epidemiological surveillance, the development of screening and diagnostic assays, as well as in advancing our understanding of disease epidemiology and pathogenesis.

Aims: This study aimed to investigate the utility of metagenomics sequencing in detecting the plasma virome and characterizing viral profiles in

eligible plasma biorepository samples. Such analyses are crucial for enhancing blood safety protocols and understanding pathogen prevalence.

Methods: A total of 1200 plasma samples were randomly selected from the blood donor biorepository, with each set of 100 samples pooled together. Subsequently, cell debris and bacteria were eliminated using PVDF filtration membrane. Viral nucleic acids were then extracted, followed by the construction of DNA and cDNA libraries. Metagenomic sequencing and subsequent data analysis were conducted.

Results: From the sequencing efforts, 2336 viral reads via DNA sequencing and 61985 viral reads via cDNA sequencing were obtained. A total of seven DNA viruses belonging to two families and one RNA virus were identified from these reads. Among them, DNA viruses include four viruses of the family *Anelloviridae* (sequences accounting for 92.1%, 2145/2336), namely Torque teno virus (TTV), Torque teno midi virus (TTMDV), Torque teno mini virus (TTMV) and TTV-like mini virus (TLMV). Additionally, three DNA viruses (sequences accounting for 7.9%, 184/2336) from the family *Herpesviridae* were identified, including human herpesvirus 6A (HHV-6A), human cytomegalovirus (HCMV), and Epstein-Barr virus (EBV). The predominant RNA virus (96.2%, 59602/61985) was GB virus C/human pegivirus (GBV-C/HPgV). Anelloviruses and GBV-C, generally considered non-pathogenic viruses, were the predominant DNA viruses and RNA viruses respectively presented in eligible plasma. Conversely, the three identified herpes viruses are known opportunistic pathogens, with EBV notably associated with nasopharyngeal cancer, prevalent in southern China. Notably, distinct virus distribution patterns were observed among different plasma pools, with non-pathogenic viruses detected in 12 pools, while herpes viruses were found in two pools.

Summary / Conclusions: This study indicated the potential of utilizing blood donor biorepository samples in conjunction with metagenomics sequencing to characterize plasma virus profiles, especially those beyond the scope of routine blood screenings. Regular monitoring of these profiles could contribute to infectious disease surveillance and epidemiological research, ultimately enhancing blood transfusion safety and public health outcomes.

PA29-L04 | Hypotensive adverse events among U.S. source plasma donors Plasmavigilance II

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Background: Source Plasma (SP) is the starting material for 87% of plasma-derived medicinal products globally, especially Immunoglobulins. The Plasma Protein Therapeutics Association (PPTA), an international trade and voluntary standard-setting association representing the SP industry, has been instrumental in setting the Plasmavigilance program to promote donor safety. The PPTA International Quality Plasma Program (IQPP) Standard for Recording Donor Adverse Events (DAE Standard) provides a common language to define and classify donor adverse events (AEs) across the SP collection industry. PPTA and its member companies are committed to providing essential data to its members, regulators, patients, and donors to monitor donor health.

Aims: This study analyzes hypotensive AE (HAE) rates and SP donor characteristics that may be predictors of a hypotensive AE.

Methods: Donation data for 1.1 million donors making 12,183,183 SP donations between May 1 and August 31, 2018, were analyzed. This represented approximately 72% of the donations collected by the U.S. plasma industry. The PPTA DAE Standard was used for AE definitions and classifications. A logistic regression analysis was performed individually for males and females to compare donors with HAE to donors without HAE based on donor attributes such as age, donation status, estimated blood volume (EBV), body mass index, and pre-donation screening tests, including height, weight, systolic blood pressure (SBP), diastolic blood pressure (DBP), total protein, hematocrit (HCT), and pulse. Unadjusted (univariate) odd ratios and 95 percent Confidence Intervals were calculated.

Results: The female and male rate for HAE was 16.18/10⁴ donations and 3.56/10⁴ donations, respectively. HAE rates were the highest at the lighter end of the weight ranges for all three nomograms, but the rates were generally higher, and the association was more pronounced in female donors compared to male donors. EBV and donation status were the attributes associated with having the highest predictors of HAE. Donors aged less than 24 and older than 65, first time donor status, low EBV, and high pre-donation pulse. There appears to be no association with pre-donation SBP, DBP, or HCT, for females, with the odds of having a HAE.

Summary / Conclusions: At donation frequencies and volumes allowed by the US Food and Drug Administration, SP donors have low HAE rates. Special attention and mitigation strategies could be directed to donors who are young, light weight, low EBV, female, or first-time donors to further reduce the odds of HAE, continue to ensure the donor has a safe experience.

PA29-L05 | Long COVID in blood donors—prevalence, associations with post-infection SARS-CoV-2 antibody levels, and clinical subphenotypes

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Background: Long COVID became a common, sometimes debilitating condition; yet, the case definition, determinants, and pathogenesis remain incompletely understood. Recognizing how antibody (Ab) responses correlate with long COVID could support the development of preventive and therapeutic interventions.

Aims: We aimed to investigate the: (1). prevalence of long COVID in blood donors, comparing self-report and a literature-based definition derived from a large study; (2). factors associated with long COVID, with a focus on levels of anti-SARS-CoV-2 Ab at pre-defined time points; and (3). patterns of distinct subphenotypes of long COVID symptoms in the donor population.

Methods: People who donated blood during the period of universal testing for SARS-CoV-2 (June/2020-July/2021) at our blood center were invited to respond to a survey addressing comorbidities; COVID-19 infection; acute/persistent symptoms; and vaccinations. We compared the prevalence of long COVID as defined by self-report and a second definition adapted from the RECOVER study. Univariable and multivariable modified Poisson models were used to evaluate factors associated with long COVID, focusing on anti-SARS-CoV-2 Ab categorized into quartiles measured at 4–12; 12–24; and 24–48 weeks post-infection. Anti-nucleocapsid Ab were measured by Roche Elecsys NC Total Ig and VITROS N Total Ig; anti-spike IgG Ab levels were measured by Ortho VITROS IgG; total anti-spike Ab were measured by Ortho VITROS S Total Ig. We investigated subphenotypes of long COVID using hierarchical clustering.

Results: Of 33,610 participants, 16,003 (48%) reported having had COVID-19. Of those, 1853 (12%) self-reported long COVID; 685 (4%) had long COVID as defined by an adapted RECOVER definition; 488 (3%) met both criteria; and 2,050 (13%) met at least one long COVID definition. Multivariable models adjusted for age, sex, race, ABO/Rh blood type, hospital care during acute infection, number of comorbidities, vaccination before infection, and time between infection and donation showed that higher anti-nucleocapsid Ab levels measured 12–24 weeks post-infection were associated with higher risk of self-reported long COVID (adjusted risk ratio [aRR] for 4th vs. 1st quartiles 1.32, $p = 0.042$) and higher risk of adapted RECOVER definition (aRR for 3rd vs. 1st quartiles 2.01, $p = 0.026$). Higher anti-spike IgG Ab levels measured 12–24 weeks post-infection were

associated with lower risk of self-reported long COVID (aRR for 2nd and 4th vs. 1st quartile 0.32 [$p = 0.018$] and 0.21 [$p = 0.017$], respectively). Higher total anti-spike Ab measured 24–48 weeks post-infection was associated with lower risk of adapted RECOVER definition (aRR for 2nd and 4th vs. 1st quartile 0.46 [$p = 0.019$] and 0.42 [$p = 0.006$], respectively). Cluster analysis identified four clinical subphenotypes: neurological and psychiatric symptoms for cluster 1; neurological and respiratory symptoms for cluster 2; multi-systemic symptoms for cluster 3; and neurological symptoms alone for cluster 4.

Summary / Conclusions: Even among relatively healthy blood donors compared to the general population, long COVID is common using two imperfectly overlapping definitions. Results showed time-dependent associations between SARS-CoV-2 Ab and long COVID risk. Specifically, higher levels of anti-nucleocapsid Ab were associated with higher risk, whereas higher levels of anti-spike Ab were associated with lower risk of long COVID. Clinical subphenotypes suggest distinct pathophysiologic mechanisms in specific subgroups.

Parallel session—blood products

Removing plasmatic ABO incompatibility and RBC in-vitro production

PA30-L01 | Self-anticoagulant sponge for whole blood auto-transfusion

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Clinical use of intraoperative auto-transfusion requires the removal of platelets and plasma proteins due to pump-based suction and water-soluble anticoagulant administration, which causes dilutional coagulopathy. Herein, we develop a carboxylated and sulfonated heparin-mimetic polymer-modified sponge with spontaneous blood adsorption and instantaneous anticoagulation. We find that intrinsic coagulation factors, especially XI, are inactivated by adsorption to the sponge surface, while inactivation of thrombin in the sponge-treated plasma effectively inhibits the common coagulation pathway. We show whole blood auto-transfusion in trauma-induced hemorrhage, benefiting from the multiple inhibitory effects of the sponge on coagulation enzymes and calcium depletion. We demonstrate that the transfusion of collected blood favors faster recovery of hemostasis compared to traditional heparinized blood in a rabbit model. Our work not only develops a safe and convenient approach for whole blood auto-transfusion, but also provides the mechanism of action of self-anticoagulant heparin-mimetic polymer-modified surfaces.

PA30-L02 | Universal human plasma for blood group independent transfusion

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Background: Isoagglutinins in the plasma require for ABO blood group compatibility. We developed a GMP conform procedure to deplete the isoagglutinin titer of fresh frozen plasma (FFP) units group A, B, and O. The method is based on isoagglutinin adsorption onto added red cells from red cell concentrates (RCC). A requirement for the approval of universal plasma from blood group A units was an extensive validation study to be able to assess the impact of the plasma treatment on coagulation and immune parameters.

Aims: GMP conform production of universal plasma for blood group independent transfusion

Methods: We developed a bag system, which enables pooling of three FFP units, addition of the adequate RCC volume and subsequent removal of isoagglutinin red cell sediment in a closed system. After incubation at 20–24°C for 2 h the isoagglutinin red cell sediment was removed from the plasma pool by centrifugation (4000 g, 10 min) and separation. Within the validation study we compared plasma units of 12 production processes (= 36 units) with or without the addition of RCC for isoagglutinin adsorption with regard to the following parameters: anti-B titer, hemoglobin, residual red cells, factors II–XIII, fibrinogen, vWF antigen + activity, protein C + S, antithrombin, ADAMTS13, aPTT, Quick value, lactate dehydrogenase and complement activation.

Results: Starting from maximum values of 1:16 in the plasma pools, total anti-B titers were reduced to < 1:1 in all units of the treatment arm. Free hemoglobin was similar between the control and treatment group ($24.8 \pm 19.7 \mu\text{M}$ vs $22.9 \pm 15.4 \mu\text{M}$). No residual red cells were found by microscope counting. All other coagulation and immune parameters showed no significant differences between the control and treatment arms (e.g. factor VIII $75.4 \pm 19.0\%$ vs $76.0 \pm 19.0\%$, fibrinogen $2.7 \pm 0.5 \text{ g/L}$ vs $2.6 \pm 0.4 \text{ g/L}$, protein C $92.3 \pm 6.6\%$ vs $88.9 \pm 6.1\%$, protein S $85.8 \pm 13.2\%$ vs $83.8 \pm 9.8\%$).

Summary / Conclusions: We present a fully automatic GMP conform procedure for the production of universal plasma for blood group independent transfusion. With regard to all coagulation and immune parameters, we were able to show that our method has no impact on the quality and safety of the universal plasma.

PA30-L03 | How high is high?—an assessment of anti-A/B titres in the UK donor population

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Background: In the UK, NHS Blood and Transplant (NHSBT) test all donors at each donation for high titre (HT) anti-A/B IgM at a single dilution of 1:128, or equivalent, by observing agglutination patterns in

PA30-L03 Table 1: Percentage of donors testing positive at titres above 1:128 with repeat screening.

Dilution	High titre positive/query percentage	
	Group A N = 100	Group O N = 100
1:128	93%	95%
1:512	17%	25%
1:1024	6%	7%
1:2048	1%	1%
1:4096	0%	0%

microplate wells on an automated platform (PK7300, PK7400 Beckman Coulter). If agglutination is present, the donation is considered HT positive, if not, the donation is considered HT negative. For pooled components containing plasma e.g., pooled cryoprecipitate or pooled platelets, a unit is considered HT negative if all the constituent donations have tested as HT negative. Currently, UK guidelines recommend that group specific plasma components should be transfused wherever possible. Unfortunately, there is limited availability of specific blood groups due to the distribution of blood groups amongst the UK donor population. Furthermore, a patient's blood group may not always be known at the point of transfusion. As such, where transfusion of group specific plasma is not possible, the least incompatible ABO group is used. In such events, the unit must have been tested and found negative for HT anti-A/B. At present, data to understand the precise titre of anti-A/B in donors is lacking, so the exact titre of a unit of HT plasma is not known.

Aims: This study aimed to understand the precise anti-A/B titres of HT positive UK donor samples.

Methods: A total of 200 samples, 100 group O and 100 group A, all testing HT positive at 1:128 for IgM anti-A/B during standard mandatory screening were selected. The plasma of each HT positive sample underwent a series of doubling dilutions with phosphate buffer solution (PBS), to give a result equivalent to 1:128 (neat), 1:512, 1:1024, 1:2048 and 1:4096, and subjected to standard screening methodology again.

Results: Only 17 % of Group A HT samples tested were still positive at 1:512, with 25% positive in Group O. At the highest titrations measured, only 6%–7% demonstrated agglutination at 1:1024, 1% at 1:2048 and 0% at 1:4096, for both ABO groups.

Summary / Conclusions: In the UK donor population, the incidence of high titre anti-A and B during mandatory testing in donor samples (<1:128) is around 10%–15%. In this study, only a small percentage of these HT donations had very high anti-A/B (e.g., between 17% and 25% of the HT positive samples were positive at 1:512, only 1% at 1:2048 and 0% at 1:4096). These data will contribute to understanding the risk associated with the transfusion of high anti-A/B titre plasma. This is particularly important in pooled components containing plasma, which are currently labelled as HT positive if any one of the constituent units has been assessed as HT positive for anti A/B IgM, even if the final pooled component may not be >1:128. It will also inform the capacity required in the development of medical

devices to remove antibodies, to produce universal blood components.

PA30-L04 | Metabolic profiling and media component optimization of cultured erythroblast for the production of transfusion-ready cultured red blood cells

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Background: Transfusion of donor-derived red blood cells (RBCs) is the most commonly used form of cell therapy and although generally safe, suffers from risks of alloimmunization against blood group antigens and blood borne diseases. Cultured RBCs (cRBC) could secure a safe transfusion product that additionally can be designed for specific target groups. Erythrocyte precursors can be cultured from hematopoietic progenitors, which can subsequently be differentiated into transfusion-ready cRBCs with >90% enucleation.

Aims: Although the current state-of art allows to produce cRBC compliant with all regulations required for the production of cellular products, the limited competitive value due to production costs will remain a problem if challenges of upscaling and costs management are not addressed. Here we discovered and evaluated significant media component cost-drivers using a combination of metabolomics and media component mimics.

Methods: The in vitro erythropoiesis model system was initiated from peripheral blood mononuclear cells using a factor combinations of Epo, SCF and dexamethasone followed by differentiation leading to >90% enucleation. Measurements include but were not limited to flow cytometry, ARCA (deformability), osmotic shock assays, metabolomics, and capillary microfluidics morphological assessment.

Results: Increasing erythroblast concentrations in media led to reduced support for erythroblast proliferation. However, a 1:1 mix with fresh medium restored growth. Separating fresh and spent media with a 3 kDa cut-off revealed that exhaustion of the <3 kDa fraction limited growth. Metabolomics analyses at 12, 24, and 36 h post-seeding showed nucleoside degradation, amino acid depletion, and decreased cycle intermediates, indicating oxidative stress in high-density erythroblast cultures. Removing nucleosides lowered purine salvage intermediates, boosting cell productivity by 30%. In addition, we show that the high cost-driver human serum albumin can be replaced by polyvinylalcohol (PVA) during expansion phase of CD71+CD235low erythroblasts but not during differentiation. Supplementation of human serum albumin only marginally rescues

differentiation, however was completely rescued by addition of human plasma (5%). Plasma contains a significant amount of cholesterol. The erythrocyte membrane contains 50% cholesterol. Surprisingly, media supplementation with cholesterol (1-5 mg/dL) at the onset of terminal erythroblast differentiation inhibited enucleation compared to standard differentiation medium (SDM). In contrast, the addition of cholesterol at the point of enucleation in differentiation resulted in significantly increased filter recovery, stability of reticulocytes, enhanced deformability, and osmotically more stable reticulocytes compared to SDM, without compromising culture purity (>97%) or enucleation. We also investigated the capillary passaging of cRBCs using a lab-on-the-chip microfluidic system to quantify specific shape changes. The temporal addition of cholesterol to the culture medium significantly improved the yield and quality of enucleated cultured RBC while improving their downstream processing ability and purification process.

Summary / Conclusions: In conclusion, we have identified and amended specific media components that allow for a more cost-effective production of cultured RBC and introduced a tool for their quality control.

PA30-L05 | Improving the stability of cultured red blood cells during refrigerated storage

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Background: Red blood cells (RBCs) produced by *in vitro* culture of CD34⁺ cells are more fragile than peripheral blood RBCs. Cultured RBCs (cRBCs) are mainly immature reticulocytes, containing excess plasma membrane, that prevents the final interactions for the formation of the mature biconcave disc. Consequently, cRBCs are unstable and more fragile during storage. cRBCs stored for 10 days in SAG-M at 4°C show high haemolysis (2%-13%) compared to the accepted level in standard RBC units (<0.8%), containing less than 2% native reticulocytes. Various additive solutions have been optimised for the storage of mature RBCs, giving donated units a shelf life of 35-days in UK and Europe and 42 days in USA, but limited research has been carried out on the storage of reticulocytes. We evaluated several additives with the potential to improve the stability of cRBCs during storage, to make these cells viable for clinical or diagnostic applications.

Aims: To evaluate different additive solutions to improve the stability of cRBCs in storage. To increase the storage capacity of cRBCs during longer-term storage and assess the survival and maturation of stored cRBCs *in vivo* in a mouse model.

Methods: Peripheral blood CD34⁺ haemopoietic stem cells were cultured using a three-stage culture medium. After 18-21 days of

culture, filtered reticulocytes were resuspended at 1.5×10^9 /mL, aliquoted and resuspended in SAG-M or SAG-M plus 10%-60% citrate-phosphate-dextrose (CPD), 10% colloids Dextran-40, Ficoll-70 (Dx, Fi) or 10% human serum albumin. Some aliquots were rejuvenated (Rj) by the addition of 16.67 mM sodium pyruvate and 16.67 mM inosine followed by incubation at 37°C for 1hr prior to storage. The cRBCs were then stored at 4°C for 10, 16 or 20 days. At the end of storage, the supernatant was analysed for haemolysis and the cell morphology was assessed by confocal microscopy. For the *in vivo* assays, macrophage-depleted NOD.Cg-Prkdc^{scid}Il2rγ^{tm1Wjl}/SzJ mice were transfused with 2×10^8 cRBCs stored in SAG-M or SAG-M+10%Dx on day 1, 9 and 21 of storage. Peripheral blood aspirates were collected at different intervals post-inoculation and analysed by flow cytometry and confocal microscopy.

Results: The addition of 60% CPD to SAG-M reduced haemolysis from 8% to 2% ($p < 0.001$) but caused the cRBCs to dehydrate. The addition of colloids to SAG-M maintained cell morphology, and reduced haemolysis by approximately 50% at day 10 of storage compared to those stored in SAG-M alone ($p < 0.01$ and $p < 0.05$, respectively). At longer-term storage, the average haemolysis of cRBCs in SAG-M alone increased from 2.5% on day 10 to 8% on day 16, whilst haemolysis of cRBCs in SAG-M+10%Dx+Rj remained around 2% for the 20 days ($p < 0.0001$). In the *in vivo* study, human cRBCs stored in either solution could be detected in the mouse circulation from 10 min up to 48 h. Overall, the storage solution did not affect cRBCs survival in murine circulation. A decrease in the expression of the reticulocyte maturation marker CD71 was observed, accompanied with a reduction in diameter.

Summary / Conclusions: We tested different additives to SAG-M and found that the addition of 10%Dx to SAG-M significantly reduces cRBCs haemolysis during storage. We showed that stored cRBCs survive *in vivo* 48hrs post-infusion and appear to mature in circulation. These results are, to our knowledge, the first report showing an increased stability of cRBCs during storage making the future applications of cRBCs a real possibility.

Parallel session—management and organisation

Plasma—the big picture!

PA31-L01 | SUPPLY in a nutshell

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Background: Donated plasma is the only source to produce plasma-derived medicinal products (PDMPs) which are indispensable for the treatment of many patients. The Covid-19 crisis has shown how vulnerable the world is in supply of life-saving medicines. PDMPs,

particularly immunoglobulins, are facing shortages in Europe today, with their availability driven largely by a world-wide competitive market. Sufficient collection of plasma by a country does not automatically mean sufficient supply of PDMPs for its inhabitants, and there are also large differences in the usage of IgG between countries. Simultaneously, there is an imbalance in the global collection of plasma needed for the products, with a high dependence on plasma coming from the USA.

Aims: By assessing the entire plasma-to-PDMP-to-patient chain—from plasma donor recruitment, retention, and health, through plasma collection and processing, to procurement, demand, and use of PDMPs—the SUPPLY project provides evidence-based recommendations to both strengthen voluntary non-remunerated plasma collection capacity in Europe and to help ensure safe and adequate access for EU patients to essential PDMPs. SUPPLY thereby aims to contribute to the EU becoming more strategically independent in its need for plasma medicines and to help address the global imbalance in the collection of plasma.

Results: The SUPPLY project provides a set of recommendations and guidance for the EU, National Governments, blood establishments, competent authorities, medical societies, and other professional stakeholders to support them in being able to both increase plasma collection in the EU by the public health sector and to achieve optimal availability of plasma medicines for patients, both in a general situation as well as in times of crises. Among the key recommendations are: Plasma is a critical medical raw material and a public resource that requires strategic management. EU member states should create action plans to optimise investment in the increase and improvement of the plasma collection, building a solid donor base while focusing on retaining donors and increasing donation frequency within scientifically proven safe limits. National commitments to collect sufficient volumes must be accompanied by sufficient control over the plasma-PDMP-patient chain to ensure that the patient population needs are met, including in times of crises. Policymakers, healthcare providers, and stakeholders must work collaboratively to proactively address risks identified and adopt a systemic approach that combines different solutions and leverages the synergies between them. Carry out a large prospective study in plasma donors to examine the health consequences of plasma donation at varying frequencies. Take consideration of the IgG level to determine the value of plasma. Introduce legal provisions at national level which link collected plasma to the usage of products manufactured from this plasma by the public health sector. Create national databases on Ig usage at patient level. Create an EU wide harmonized management plan on shortages.

PA31-L02 | Creating a roadmap for the appropriate and prioritised use of immunoglobulins in Europe

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Background: With the rising demand of immunoglobulins (Ig) worldwide and high dependency on the U.S. market, Europe's plasma supply chain faces constant shortages. Therefore, the SUPPLY project was established to increase and strengthen the resilience of plasma collection in Europe to enable a stable and adequate supply of plasma-derived medicinal products (PDMPs) by assessing the entire plasma value chain from donor recruitment to demand and use of PDMPs.

Aims: As part of SUPPLY, Work Package (WP) 6 focused on demand and use of PDMPs, particularly Ig. WP6 sought to provide recommendations on the appropriate and prioritised use of Ig in Europe and the UK during baseline and times of crises (specifically, on the impact of COVID-19).

Methods: Five steps were conducted: (1) a survey for clinicians; (2) semi-structured interviews of clinicians and pharmacists; (3) an extensive grey literature search focusing on the five biggest markets of Ig; (4) a French case study using the national claims database; (5) a multi-stakeholder virtual workshop.

Results: The clinicians' survey resulted in 183 respondents in 18 countries primarily in the fields of haematology, neurology, and immunology. The interviews were carried out with 14 clinicians and 2 pharmacists from 8 countries in the fields of haematology, neurology, and immunology. Both the survey and interviews revealed what is used to guide Ig decision-making in the clinical environment, which ranged from various guidelines, clinicians' expertise, and the existence of an Ig approval process for off-label usage. Additionally, most respondents had faced increased Ig shortages during the pandemic, activating similar mitigating measures such as altering treatment paradigms. Interview respondents also shared lessons learned, such as the need for improved communication, data collection measures, and collaborations between specialties and countries. The grey literature search focused on France, Germany, Spain, Italy, and the UK which showed a diversity of Ig indications, prioritisation plans, management plans, and shortage communication measures across (and, sometimes, within) these countries. Accessing Ig dispensation data was difficult due to a lack of centralised databases and data protection laws. The French case study assessed general trends in Ig usage in France over a 10-year period; however, assessing Ig specifically regarding number of patients treated, volume used, and cost was not possible and would require additional data (collection). The results from all these steps were presented during a virtual workshop to 47 participants representing clinicians, patient representatives, national competent authorities, European Medicines Agency, and the European Commission. Preliminary recommendations regarding the creation of a comprehensive national database on Ig usage on the patient level and the need for a harmonised prioritisation and management plans across the EU were presented to participants. Feedback was obtained, identifying pertinent steps to these recommendations and similar ongoing initiatives on an EU level.

Summary / Conclusions: Our results contributed towards the creation of five recommendations regarding improved understanding of Ig usage, harmonisation of prioritisation and shortage management plans, and enhanced collaborations and linkages. These are a roadmap

for next steps, but further work is needed to make these recommendations into actual Ig prioritisation plans and management strategies.

PA31-L03 | Quality management, processing and cost modelling for efficient plasma collection with a minimum of wastage—recommendations from the SUPPLY Project

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Background: The European Union's EU4Health Programme (2021–2027) funded the SUPPLY Project “Strengthening voluntary non-remunerated plasma collection capacity in Europe” to find solutions to increase the volume, efficiency, and resilience of plasma collection for fractionation, in times of crisis.

Aims: Among 7 Work Packages, WP3 studied Plasma donor collection best practices and supply chain. Two Tasks “Characterization of the waste of recovered plasma and missed opportunities for plasmapheresis” and “Plasma collection & processing chain and cost-modeling” aimed at finding solutions for efficient, cost-effective collection processes and minimize waste of plasma.

Methods: A survey was sent to competent authorities in the 27 EU member states (MS), to describe volumes and information about both recovered and apheresis plasma supplied for fractionation (PpF), and if the case, the reasons for not collecting by apheresis, or not sending PpF for fractionation. Visits to Blood establishments (BE) collecting apheresis plasma compared practices and provided insight on best practices to collect in the most efficient way.

Results: 15 MS replied to the survey. Volumes of plasma collected are shown in Table 1. Proportion of MS supplying apheresis PpF according to volume in Table 2.

From the survey and visits at BEs, possible optimisations for plasmapheresis were explored and some gaps identified in existing quality management systems on collection of PpF for each MS/ BE organisation. The task groups provided recommendations: Practical

PA31-L03 Table 1 Volumes of recovered and apheresis plasma collected

15 EU MS	Total (l)	Range (l)
PpF recovered from WB donations	3.226.593	
Apheresis PpF	2.991.826	from 60 to 2M
Plasmapheresis PpF per 1000 inhabitants	2 to 42	Mean 15; median 13.5

PA31-L03 Table 2 Volumes of apheresis plasma sent for fractionation per MS

Volume sent (1000 l)	Number of MS
0	2
<10	1
<50/100	7
>100 <1000	4
>1000	1

recommendations for BEs for the collection and processing of apheresis plasma highlight the importance of digitalisation and automation, approval of donations from First-Time Tested Donors, reduced plasma content in platelet product, use of bottles for storage, and questionnaire/virus testing done in accordance with PpF destination to reduce unnecessary resources. Political recommendations for EU institutions to increase plasmapheresis in the public sector included: Government pragmatic support for plasmapheresis program, Regular national campaigns for plasmapheresis; Simplification and modification of regulatory requirements related to trained healthcare personal; physician on call; best donation frequency for high IgG level and donor protection; haemoglobin and ferritin thresholds; and making sure EDQM guideline are interpreted the same way in all European BE's.

Summary / Conclusions: Best Models for implementing/improving processes in hybrid or plasma-only centers, a Transfer Plan with toolbox for implementing quality management recommendations, and a Plasma Collection Accreditation System unified at EU level for the public sector would allow initiation of plasmapheresis collection in MSs who do not have a program and harmonisation of high-quality procedures of best practices in all European BEs. Harmonisation of PpF collection practices in the EU public sector would lead to a better attractiveness of its PpF to fractionators and ultimately, pooling small MS' volumes that would otherwise being wasted. This would contribute reducing the dependency of EU from the US by increasing volumes of PpF collected by the public sector.

PA31-L04 | Enhancing plasma donor retention—an exploration of factors shaping new donors future donation commitments

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Background: Retaining new source plasma donors in voluntary non-remunerated settings is imperative to meeting domestic and worldwide demand for plasma-derived therapeutic products. Existing research suggests that retention is impacted by the particular characteristics of plasmapheresis, including donation time, needle size, and the accessibility of the plasma center. Studies on plasma donation have largely focused on the experiences and perspectives of whole-blood donors who transition to plasma donation. Little attention has been given to new plasma donors with a long-lapsed or no prior whole blood donation history.

Aims: To understand the factors impacting the intentions of new plasma donors to make future donations and develop a habit of plasma donation.

Methods: Using a sociological approach that interrogates the experiences of plasma donors within a broad set of social relationships and contexts, this study investigates the experiences of new plasma donors at three Canadian Blood Services source plasma collection centers. Purposive sampling was used to identify individuals who became first time plasma donors between March 2021 and February 2023 and who were not previously registered donors in the CBS system. Data from 46 individual semi-structured interviews were coded using NVivo software and analyzed using thematic analysis.

Results: The majority of new plasma donors express their intention to donate in the future, often citing multiple reasons. Positive donation experiences and an ongoing desire to help others through plasma donation serve as key motivators for subsequent donation practices. New plasma donors construct their motivations for future donation in relation to others. They are encouraged by positive interactions with staff at the donation center and perceive the donation process as an opportunity to strengthen existing relationships and expand their social network. Additionally, the desire to help others is explained in relational terms, seen as a commitment to assisting those who are ill or in need, which also brings a sense of personal fulfillment to the donor. New plasma donors express a desire to receive more information about the impact of their donations, particularly regarding the recipients of plasma products. Few new plasma donors establish a regular donation routine or commit to a fixed donation schedule. Instead, they set goals or flexible plans for donation. This nuanced approach is situated within the complexity of their everyday lives and informed by established social relations. Participants emphasize they must work with and around work, school, family and other social responsibilities to engage in ongoing donation practices.

Summary / Conclusions: Our study highlights the significant role of the plasma donation experience in influencing new plasma donors' intentions to make subsequent donations. Blood services should enhance donor retention by building upon the relationships that donors establish with staff and other donors and emphasize the stories of recipients to provide donors with a sense of connection though a clear understanding of their impact on others. Additionally, blood operators should support new donors in establishing goals and/or flexible donation habits that are aligned with insights into how others incorporate donation into their complex lives.

PA31-L05 | The impact of plasmapheresis frequency on donor health—a systematic review of controlled experimental and observational studies

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Background: Most plasma used for manufacturing plasma-derived medicinal products (PDMPs) such as albumin, immunoglobulins (Ig), and clotting factors is obtained from source plasma collected via plasmapheresis, the majority of which is contributed by the United States where donors can donate plasma up to twice per week and up to 100 times per year. SUPPLY is a project co-funded by the European Union that aims to strengthen voluntary non-remunerated plasma collection capacity in Europe, to enable a stable and adequate supply of PDMPs.

Aims: To conduct a systematic review to identify, analyze, synthesize, and critically appraise the best available scientific evidence that investigates the impact of plasmapheresis frequency on donor safety and health.

Methods: Six databases and two trial registries were searched until 4 December 2023. (Non-)randomized controlled trials (RCTs) and controlled observational studies investigating the impact of plasmapheresis frequency (higher frequency regimen versus lower frequency regimen) on adverse events (primary outcome), cardiovascular health, or protein levels (secondary outcomes) were eligible for inclusion. The Grading of Recommendations, Assessment, Development and Evaluation (GRADE) approach was used to assess the risk of bias and overall certainty of the evidence.

Results: Four cohort studies, 1 non-RCT and 2 RCTs (of which one is ongoing) were included. The 4 included cohort studies were graded as providing very low-certainty evidence (due to methodological limitations and imprecise results because of limited sample sizes). The non-randomized controlled trial showed no statistically significant or clinically meaningful differences in total protein or immunoglobulins (IgG, IgA, IgM) after weekly compared to bi-weekly plasmapheresis during 6 months (low-certainty evidence). The completed RCT compared high-frequency plasmapheresis (3 donations per month) or very-high-frequency plasmapheresis (2 donations per week) to donors undergoing monthly plasmapheresis or a sham intervention during 12 weeks: Few (minor) adverse events were reported in high-frequency and very high-frequency groups with adverse event rates (per 50 donations) <1 for haematoma, vasovagal reactions, and anaemic events. (low-certainty evidence). Plasmapheresis twice per week may result in a large reduction in ferritin and IgG levels (low-certainty evidence) (Very) high-frequency plasmapheresis may result in little to no difference in other protein levels (Hb, IgA, IgM, albumin, CK, CRP) or in cardiovascular health (low-certainty evidence). Preliminary results from the ongoing RCT demonstrated that total protein and IgG levels were significantly reduced by plasmapheresis over 15 weeks with both high frequency (3 times per 2 weeks) and 2-week intervals and this reduction increased with increasing donation frequency. For high-frequency donors, total protein and IgG levels had not recovered baseline concentrations even 4 weeks after donations.

Summary / Conclusions: More RCTs and prospective cohort studies are needed to examine the (long-term) health consequences of plasmapheresis at varying frequencies. Pending these high-quality studies and more conclusive evidence on the impact of very frequent plasma donations on donor health, not-for-profit blood establishments should continue to use the precautionary principle to maximize safety for plasma donors. This implies that frequent plasmapheresis should be defined with a broad safety margin.

Parallel session—clinical

EHA / ISBT joint session on management of immune thrombocytopenia - how low can it go?

PA32-L01 | New insights into ITP Pathophysiology and therapies

J W Semple

Abstract not available

PA32-L02 | New approaches for the management of immune thrombocytopenia

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Immune thrombocytopenia (ITP) is an autoimmune disorder that results in a low platelet count due to a combination of inadequate platelet production and increased peripheral platelet destruction. Due to the improved understanding of the pathophysiology of the disease, in recent years there have been significant advances in therapeutic management of the ITP in adults, which we will review here. When platelet counts falls below $20\text{--}30 \times 10^9/\text{L}$, as initial treatment, corticosteroids (CS) continue to be the standard, although the adverse effects associated with their long-term use make it advisable to try to use them only for a limited period of time. They have a response rate between 40% and 70%, however 80% of patients eventually relapse or become CS-dependent. There have been attempts to increase the response rate to CS by adding a second drug such as mycophenolate mofetil and with all-trans retinoic acid. In patients with bleeding, at high risk of bleeding, who require a surgical procedure or are unresponsive to CS, the administration of intravenous (IV) immunoglobulin G (IgG) or IV anti-D (not available in Europe) may be indicated. Other second lines of treatment currently used, include anti-CD20 monoclonal antibodies (for inhibiting the production of antibodies) or

thrombopoietin receptor agonists (eltrombopag, avatrombopag, or romiplostim) for increasing the production of platelets, although we must bear in mind that this approach do not alter the disease's immune mechanisms. Interestingly the cells producing antibodies, plasma cells in general do not express CD20 so the positive role of anti-CD20 in autoimmune disease in general and in ITP in particular, are thought to be due to an indirect effect through the removal of B lymphocytes. However, plasmablasts and plasma cells express CD38 at high levels. There are two monoclonal antibodies directed against CD38 being studied as a treatment of ITP. One is mezagitimab and the second is daratumumab, widely used in the treatment of multiple myeloma Spleen tyrosine kinase (Syk) has become a novel target for the treatment of ITP due to its role in the pathophysiology of the disorder. Fostamatinib is non-specific tyrosin kinase inhibitor currently approved for the treatment of ITP although a sustained response is only found in a 18% of the patients. Another oral SYK inhibitors, solevlenib, is currently being developed. Another target recently identified in the management of the ITP is the Bruton's tyrosine kinase (BTK). BTK plays a key role in the mechanisms involved in antibody production and also mediates the activation of macrophages and regulates phagocytosis. Several inhibitors are being developed such a rilzabrutinib and orelabrutinib, both active orally. Neonatal fragment crystallizable receptor (FcRn) plays a critical role in maintenance of the physiological levels of IgG in the circulation. Inhibiting FcRn provokes a reduction in IgG concentration without affecting the levels of IgM and IgA. Currently there are two different FcRn inhibitors being studied for treating CS-refractory ITP patients. Rozanolixizumab, a humanized IgG4 anti-FcRn monoclonal antibody and efgartigimod, a monoclonal IgG1 Fc fragment, both to be administered subcutaneously. In summary, over the last twenty years we have seen a tremendous increase in therapeutic alternatives for the management of CS-refractory or relapsing forms of ITP. As a result, splenectomy, previously indicated in the early stages of the disease, is now considered a last resort.

PA32-L03 | Blood donor SARS-CoV-2 infection is associated with increased platelet transfusion effectiveness in recipients without acute COVID-19

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Background: SARS-CoV-2 infection causes platelet activation which may cause thrombosis in COVID-19 patients. We recently showed that blood donor SARS-CoV-2 infection was not associated with thrombosis in platelet transfusion recipients without COVID-19.

PA32-L03 – Table 1

24-h RBC transfusion events in relation to blood donor SARS-CoV-2 N-Ab positivity				
Days from initial donor N-Ab positivity				
RBC Tx Events	N-Ab negative (n = 7542)	0-60 days (p < 0 = 368)	61-180 days (p < 0 = 182)	>180 days (p < 0 = 115)
Adj. RBC Tx [95% CI]	38% [37-39]	32% [28-36]	41% [35-47]	39% [31-46]
Adj. RBC units [95% CI]	0.7 [0.7-0.8]	0.5 [0.4-0.7]	0.7 [0.5-1.0]	0.7 [0.4-1.0]
Groupings of N-Ab S/C ratio				
RBC Tx Events	N-Ab negative (n = 7542)	Low S/C (n = 438)	Intermediate S/C (n = 146)	High S/C (n = 81)
Adj. RBC Tx [95% CI]	38% [37-39]	36% [32-40]	38% [31-44]	27% [18-36]
Adj. RBC units [95% CI]	0.7 [0.7-0.8]	0.7 [0.5-0.9]	0.7 [0.5-0.9]	0.5 [0.3-0.7]

Abbreviations: RBC: red blood cell; S/C: signal-to-cutoff; N-Ab: SARS-CoV-2 nucleocapsid antibody; Tx: transfusion; CI: confidence interval.

However, the impact of blood donor SARS-CoV-2 infection on platelet transfusion efficacy is unknown.

Aims: Utilizing a linked vein-to-vein database, we examined associations of blood donor SARS-CoV-2 nucleocapsid antibody (N-Ab) results on a measure of platelet transfusion effectiveness.

Methods: We linked blood donor SARS-CoV-2 serology data with Kaiser Permanente Northern California adult patients without active SARS-CoV-2 infection who were transfused apheresis-derived platelet components between 6/1/2020 and 3/31/2022. Using multivariable regression, 24-h red blood cell (RBC) transfusion events following single-unit platelet transfusion were examined as an indicator of effectiveness in relation to the timing and signal-to-cutoff (S/C) ratios of donor SARS-CoV-2 N-Ab data, adjusting for other donor, component, and recipient factors. N-Ab S/C ratios were defined as: negative (<1), low (1-99), intermediate (100-150), and high (>150). We calculated the adjusted incidence and number of RBC units transfused in the 24 h following platelet transfusion.

Results: We identified 2808 adults who received 8207 single-unit platelet transfusions for which donor SARS-CoV-2 serology data were available. Over the study period, 8.1% (665/8207) of units were from SARS-CoV-2 N-Ab positive donors, and the number of days from index seropositivity to donation was 56 (IQR 0-141). The pre-transfusion platelet count was 32 (IQR 12-60), and the frequency and number of RBC transfusions in the 24 hours after platelet transfusion was 37.7% (3094/8207) and 0.7 units (standard deviation:1.4), respectively. Compared to N-Ab negative units in adjusted analyses, 24-h RBC transfusion requirements were decreased in recipients of platelet units from recently seroconverted (<60 days) (0.5 [0.4-0.7]) donors and platelet units with high S/C ratio of N-Ab titers (0.5 [0.3-0.7]).

Models adjusted for donor (age, sex, BMI), component (irradiation, pathogen reduction, storage solution and duration), and recipient (age, sex, ABO, BMI, pre-Tx platelet counts and hemoglobin levels, and prior RBC Tx). Adj. RBC Tx: adjusted incidence of RBC Tx for the 24-h period after platelet Tx; Adj RBC units: number of transfused RBC units for 24-h period after platelet Tx.

Summary / Conclusions: Platelet transfusions from blood donors with recent SARS-CoV-2 infections and high nucleocapsid antibody S/C ratios were associated with decreased RBC transfusion requirements suggestive of increased platelet transfusion effectiveness.

PA32-L04 | Towards improved understanding of platelet transfusions in preterm infants using mass spectrometry-based proteomics

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Background: Prophylactic platelet transfusions are administered to thrombocytopenic preterm infants to prevent bleeding. Unfortunately, evidence to guide practice is limited. More importantly, a recent randomized trial showed transfusion associated harm, of which the pathophysiological mechanisms are yet to be unveiled. Importantly, studies have shown differences in hemostasis between pediatric and adult populations, and large variations regarding bleeding risks associated to the etiology of thrombocytopenia. This highlights the lack of knowledge regarding the molecular process associated with platelets functions and transfusions in infants. Sampling for mechanistic studies in this population is difficult due to their small total blood volume. Unbiased proteomics provides a unique opportunity to capture a global snapshot of circulating proteins using limited material, including residual material from routine care. Thus, this technique might be an important tool to study transfusion-related questions in preterm infants. This study is a first step towards mapping the proteomic landscape of preterm infants, as a basis for more targeted studies investigating transfusion associated harm

Aims: We explored the potential of MS-based proteomics to (1) improve understanding of molecular processes associated with

platelet count, (2) identify differences in protein levels between two groups in infants focusing on patterns in often thrombocytopenic severely small for gestational age (SGA) infants compared to appropriate for gestational age infants (AGA)

Methods: Longitudinal samples from two cohorts (1) 55 infants (24-41 weeks GA, AMC biobank) and (2) 67 infants (24-30 weeks GA, PRIDICT-BPD study) were analyzed using unbiased MS-based proteomics. Associations of proteins to platelet count were evaluated with correlation. Protein profiles were compared using statistical analysis in cord blood and longitudinal monitoring of SGA-specific changes

Results: We show that residual material is of high sample quality with limited erythrocyte lysis (CA1, CA2), coagulation (FGA, FGB, FGG) and platelet activation (PPBP, PF4). Analysis of protein dynamics between MS and laboratory measurements revealed a high correlation of CRP (r : 0.97) indicating analytical reliability. Good correlations of platelet count to platelet proteins (PF4, r : 0.60; PPBP, r : 0.60), apolipoproteins (APOA1, r : 0.65; APOA2, r : 0.62) and zinc finger proteins (ZNF782, r : 0.71; ZNF256, r : 0.66) were observed. Although we found limited plasma proteomic differences at birth between SGA and AGA infants, longitudinal monitoring revealed 69 proteins with deviations in protein level trajectories over time. This included the fat-accumulating hormone adiponectin, platelet proteins and proteins associated with innate immune system

Summary / Conclusions: Here, we show the potential of MS-based proteomics in two independent cohorts by defining circulating protein trajectories in the preterm infant population associated with platelet count and showed relationships to platelet proteins and apolipoproteins. We also identified associations with two zinc fingers, of which the implication should be further explored. Furthermore, we highlight the ability of MS-based proteomics profiling to identify longitudinal alterations in protein levels associated with SGA infants. These findings provide a stepping stone to improve our understanding of platelet transfusions in infants and other populations, including hematology patients.

Working party session— Granulocyte immunobiology WP

Adverse events—TRALI

WP01-L01 | Reverse TRALI—why do we believe?

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Transfusion-related acute lung injury (TRALI) is known to be the most serious consequence of a blood transfusion and refers to respiratory distress that occurs during or after a blood transfusion. Depending on the trigger, TRALI is divided into antibody-mediated and non-

antibody-mediated forms. In antibody-mediated TRALI, the transfusion of blood components containing alloantibodies against human leukocyte antigens (HLA class I and II) or human neutrophil antigens (HNA) activates antigen-positive cells after binding and triggering reactions that lead to lung injury. In non-antibody-mediated TRALI, biological response modifiers (BRMs) formed in blood products during storage trigger an inflammatory cascade in recipients, leading to TRALI. Recent studies following the transfusion of stored blood products to volunteer recipients cast doubt on the involvement of BRMs in triggering TRALI. In 10% of TRALI cases, the recipient's antibodies to the transfused antigen may trigger a reaction, leading to reverse TRALI. While the introduction of leukoreduction of blood components has reduced the likelihood of reverse TRALI in previously immunized recipients, it has not eliminated the incidence of reverse TRALI due to transfused soluble antigens. The current presentation will explain the possible mechanisms for reverse TRALI, the incidence and prevention strategies.

WP01-L02 | Clinical impact of SLC44A2/CTL2 expression in peripheral blood cells

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The SLC44A2 (solute carrier family 44 member 2) protein, also known as choline transporter-like protein 2 (CTL2), is found in various human tissues including kidney, lung, inner ear, endothelial cells and blood cells. SLC44A2 is claimed to be responsible for choline transport into mitochondria, an important step in the oxidative pathway of choline metabolism. SLC44A2 expression in peripheral blood cells has an important impact on human diseases, especially because this protein carries the HNA-3 neutrophil system and the CTL2 erythroid blood group system. 1. Polymorphisms in SLC44A2 are responsible for Transfusion-related acute lung injury (TRALI). A SNP in SLC44A2, c.461G>A (p.Arg152Gln, 1st extracellular loop of SLC44A2), results in the HNA-3a and HNA-3b antigens. Severe TRALI is mostly due to antibodies present in plasma-rich components directed against HNA-3a. Anti-HNA-3a causes active aggregation of granulocytes which may damage pulmonary vessels. Other allelic forms of SLC44A2 were incriminated but more rarely encountered. 2. SLC44A2 may be involved in neonatal alloimmune neutropenia (NAIN). NAIN is caused by maternal alloimmunisation to fetal neutrophil antigens. The most implicated antibodies are directed against HNA-1a and HNA-1b, present on Fy_g receptor IIIb and CD177, respectively. Anti-HNA-3a/-HNA-3b are more rarely involved, with immunogenicity of HNA-3b being lower than that of HNA-3a. 3. SLC44A2 is associated with venous thromboembolism (VTE). Several studies recently confirmed SLC44A2 to promote thrombosis in a mouse model and suggested this may be related to platelet-neutrophil interaction. Genome-wide association studies in humans linked expression of HNA-3b

(rs2288904-A, minor allele) with a 30% decreased risk of VTE when compared to those expressing HNA-3a (rs2288904-G, major allele). SLC44A2 was identified as a receptor for von Willebrand factor (VWF). HNA-3a was found to play an important role for the adhesion and activation of neutrophils in veins under inflammation, and when submitted to specific shears. The fact that neutrophils expressing HNA-3b show a different response to VWF could explain the association between HNA-3b and reduced risk for VTE. 4. SLC44A2 carries the CTL2 erythroid blood group system. SLC44A2 is expressed in RBCs and carries the CTL2 blood group system (#39). Two high-prevalence antigens, VER and RIF, were initially discovered, with VER- (CTL2 null type) and RIF- being rare blood types. The RIF polymorphism (p.-Pro398Thr) is located on the 3rd extracellular loop of SLC44A2. Anti-VER was found in a patient with no expression of SLC44A2 (large homozygous deletion, exons 1-14). Anti-RIF was discovered in many patients of Moroccan ancestry. Interestingly, VER- siblings all suffered from age-related hearing loss, as found in *Slc44a2*^{-/-} mice; one also suffered from epilepsy and another one from intracranial giant aneurysms. VER- patients, with no SLC44A2 in platelets and RBCs, are not subject to any apparent haematological disorder. Anti-VER and anti-RIF are able to promote activation of neutrophils and cause their adhesion on endothelial cells, thus being both potentially responsible for TRALI. The Cs^a and Cs^b RBC antigens, from the COST collection, were recently found to correspond to the HNA-3a and HNA-3b antigens, respectively. As a result, the uncommon Cs(a-) phenotype (5% in Europeans, <1% in Africans), corresponds to the *HNA-3b/b* genotype.

WP01-L03 | Role of complement in TRALI

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The administration of blood-products is an established and often life-saving approach in a broad range of clinical settings. Unfortunately, rare life-threatening adverse transfusion reactions can occur which remain difficult to predict and which lack therapeutic interventions, including Transfusion-related acute lung injury (TRALI). TRALI occurs within 6 h of administration of a blood transfusion and remains one of the leading causes of blood transfusion-related fatalities. TRALI clinically manifests as non-cardiogenic respiratory distress, observed as pulmonary bilateral infiltrates on a chest x-ray. The pathophysiology of TRALI is complex and involves incompletely understood

immunological cellular reactions which eventually damage the pulmonary endothelium, resulting in pulmonary edema. This includes pathogenic recipient cells as macrophages, polymorphonuclear neutrophils (PMNs) and protective cells such as CD4⁺ T regulatory cells. TRALI is, in general, seen as a 2-hit model where the underlying clinical condition of patients (e.g. reflected by a state of inflammation) is defined as the first hit and the second hit is delivered by the blood transfusion product, which may contain anti-leukocyte/endothelial-reactive antibodies or biological response modifiers.

Here we investigated the role of the complement system in TRALI by using a murine antibody (anti-major histocompatibility complex (MHC) class I clone 34-1-2S)-mediated TRALI model, in combination with analyses of TRALI patient plasma samples. We found that *in vitro* complement activation correlated with *in vivo* capacity to induce TRALI in a Fc-dependent manner and that C5-deficient mice were protected from TRALI development. To further dissect the effect of the antibody Fc-region, we generated chimeric hIgG1 variants of 34-1-2S either unable to activate complement and/or Fcγ-receptors (FcγRs). This revealed an essential role for the complement system, and not for FcγRs, in the onset of TRALI in mice. We found a relationship between IgG-mediated complement activation and increased macrophage trafficking from the lungs to the blood. *In vitro* we found that neutrophil extracellular traps (NETs) could be formed in a setting mimicking TRALI with lipopolysaccharide (LPS) and C5a stimulation. In plasma of human TRALI patients we found increased levels of C1q-C4 complexes, C5a and NETs compared to healthy controls, with a correlation between C5a levels and NETs. Collectively, this reveals a critical role for Fc-mediated complement activation in TRALI, with a direct relation to macrophage trafficking from the lungs to the blood and with NET formation, suggesting that targeting the complement system may be an attractive therapeutic approach for combatting TRALI.

Working party session—Quality management WP

Quality requirements for SoHO—the risk-based approach

WP02-L01 | Proposal of new EU-SoHO regulation—the potential impact on the quality management system (QMS) of blood establishments

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The proposal of the new EU-SoHO (Substance of Human Origin) Regulation aims to ensure high protection for EU citizens by guaranteeing safe and effective blood, tissue and cells including gametes (BTC), taking into consideration the commonalities among them. More in detail, it is directed at (1) defining safety and quality requirements for all stages of the chain from donor to recipient; (2) ensuring effective regulatory oversight of the sectors; (3) achieving a degree of harmonisation of safety and quality requirements of SoHO that facilitates inter-MS exchanges; (4) establishing a high level of legal certainty at Union level; (5) achieving community sufficiency through the encouragement of voluntary unpaid donations (VUD). In this context, the quality management system (QMS), a formalised system documenting processes, procedures, and responsibilities for the performance of SoHO activities, plays an essential role. Among the general obligations of SoHO establishments, Chapter V of the new EU-SoHO Regulation highlights the minimum requirements of the QMS and the commitment of all the establishment staff in the achievement of an effective organization. More in detail, article 50 is dedicated to the QMS that SoHO establishment shall establish, maintain and update to ensure high quality SoHOs by following the Good Practice Guidelines, published by the EDQM. Moreover, the QMS shall ensure that SoHO activities are carried out by qualified personnel and in facilities organized to avoid SoHO contamination, cross-contamination by infectious agents or loss of traceability, in order to guarantee donor, patient and staff safety. To realize it, it is important to put in place procedures and instructions, to specify clearly role and responsibility of different staff, to assess the adequacy of premises and equipment, operating their preliminary qualification and a systematic check of the maintainance of the qualified status. The processes evaluated as critical for the quality and safety of the final SoHO products, according to a risk-assessment approach, have to be validated and re-validated at regular intervals, and monitored through pre-defined critical process parameters (CPP). The quality and safety of products coming from SoHO preparation processes shall be monitored through critical quality attributes (CQA) in order to demonstrate their compliance with the consistent specifications. The QMS shall include continuous quality improvement as well as the management of any contracted third

parties in charge of activities impacting on quality and safety of SoHO. SoHO establishments are required to review the QMS at regular intervals to verify its effectiveness and introduce corrective measures if deemed necessary. The Commission may adopt implementing acts regarding further details on the procedures and specifications of the QMS. Overall, the QMS in the view of the EU-SoHO Regulation appears to be substantially based on the risk assessment approach in order to ensure the highest effort where a major risk has been assessed for donor and patient health.

WP02-L02 | Requirements for effective hemovigilance systems

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Adverse reactions following blood transfusion can be caused by quality defects that appear along the transfusion chain from donor screening, testing, processing, storage, transport, issuing the blood to the patients and follow up on them. Haemovigilance is an important step in the provision of safe and efficacious blood components and blood products, because it allows to relate adverse reactions of the donor and/or the recipient with potentially systemic failures during the process. It thereby helps identifying and correct these root causes. This can only be achieved, when a clearly defined communication is established between the sites of application of the products (e.g., hospitals), the sites of manufacturing (donation sites and blood establishments performing manufacturing, storage, and transport), and ideally the national regulatory authority surveil the functioning of the system and to impose measures to prevent this in the future. The blood transfusion systems are very heterogeneously developed in different parts of the world. This also includes the existence and functionality of the haemovigilance systems. While in high income countries different but functional approaches to haemovigilance exist, this is especially not the case in low-income or low to middle income countries. This problem may be addressed by a step-wise approach starting out with a minimal system that is adapted to the local situation and that can be further developed over time. Such a minimal system would need to fulfil at least some requirements to be functional, which will be discussed in detail in this presentation. To address the requirement of a functioning haemovigilance system, the WHO published “a Guide to Establishing a national Haemovigilance System” in 2016. Within this document several aspects are laid out that can be adopted to the local situation, but also the minimal requirements are described. This was supplemented with the “User guide to navigate resources on stepwise implementation of haemovigilance systems” by WHO in 2022. Both documents build the basis of this presentation.

WP02-L03 | Quality monitoring in support of a risk-based quality system

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Despite progressive development in transfusion medicine, based on scientific and technological achievements, hemovigilance and international cooperation, some known risks are still present, while some risks are an emergent threat. Nowadays, more and more risks are recognized as a consequence of increased globalization, climate change, natural disasters, political instability, cyberattacks, and so forth. Some of them can be a threat to blood safety, blood supply, staff availability and supply of materials and equipment. Considering the biological origin of blood components and the specificity of associated risks, the growing interest and attention paid to quality risk management (QRM) is not surprising. Accordingly, QRM is appropriately addressed in various international guidelines, standards, and regulations. Due to the scope and complexity of activities in transfusion medicine, quality monitoring in the entire transfusion chain is necessary to achieve and maintain safe and high-quality transfusion treatment. This systematic process applies various tools and techniques to collect, analyze and evaluate data. In case of deviations from specified requirements, it is necessary to assess their significance and the need for corrective actions. Quality monitoring is achieved through quality control programs, use of quality indicators, regulatory audits/inspection, customer satisfaction surveys and benchmarking. QRM and quality monitoring are interrelated and complement each other in the achievement of a common goal. Quality monitoring enables the identification of potential risks to the achievement of quality objectives, while risk assessment assists in making decisions about the scope and frequency of quality monitoring activities. The results of quality monitoring are essential input data for the periodic management review, the aim of which is to assess the effectiveness of the quality system and opportunities for improvement. Quality indicators are a powerful tool in identifying potential quality concerns and risks, and monitoring trends over time. Their use in the field of transfusion medicine has been supported by the ISBT Working Party on Quality Management since 2012. The selection of quality indicators as well as the setting of quality objectives and control limits should consider the risks associated with the monitored activity/process. Risk based approach should also be used in monitoring customer satisfaction. It is important to identify possible risks for different categories of customers and use them appropriately in creating surveys. Examples of risks include not only quality and safety issues but also disruptions in supply, IT and other communication failures, and so forth. Quality control of blood components is based on statistical process control (SPC), with a defined number and frequency of sampling. Based on the volume of blood component preparation, different processing conditions and associated risks, modifications to the sampling plan may be required. Risk based approach is a strategy that should be applied in the planning and implementation of audits/inspections. This means that the available

resources are primarily focused on risky areas that can threaten the achievement of the strategic goals. According to the above, it can be concluded that QRM is an indispensable and increasingly important tool in quality management, and its proactive and preventive approach has proven to be very useful in quality monitoring activities.

WP02-L04 | WHO guidance on Implementation of a quality system in blood establishments

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Background: Blood transfusion is an essential part of modern health-care. An effective quality system (QS) contributes towards ensuring that all blood and blood products are safe, clinically effective and of appropriate and consistent quality. The *WHO Action Framework to Advance Universal Access to Safe, Effective and Quality-Assured Blood Products 2020–2023* has identified a functioning QS across the entire blood transfusion chain as a desired outcome for the strategic objective to promote efficiently managed blood services.

Aims: The WHO Guidance document was developed to provide guidance on the activities needed to implement a functioning QS in the BE. The objective is to assist and enable countries in planning, mobilising and utilising the resources available to put in place an effective QS, including leveraging existing tools and programmes. It is important that the QS is clearly defined, structured and organised, and relevant to the activities of the BE and national standards or regulatory requirements. The WHO Guidance document is to be used in conjunction with other existing documents such as the *WHO Aide Memoire on Quality Systems for Blood Safety* and the *WHO Guidelines on Good Manufacturing Practices for Blood Establishments*.

Method: A working group comprising experts in quality systems, blood regulatory systems and transfusion medicine across all the WHO Regions was appointed by WHO to develop the guidance document. The working group was divided into smaller sub-groups of authors who were designated to draft the individual chapters. The drafted manuscripts underwent multiple cycles of reviewing, editing and revision by the working group until general consensus was reached. Two rounds of consultation were conducted to further improve and refine the content and text of the document. The first round involved a small group of international quality experts and their feedback was incorporated before the document was circulated for a second wider consultation involving individuals and organisations in the field of transfusion. Further revisions and improvements were incorporated by the working group and a WHO-appointed editor ahead of final publication.

Discussion: The guidance document opens with a general introduction on quality and QS in blood services and WHO quality initiatives, followed by the potential benefits and the challenges of implementing a QS in BEs. The essential elements of an effective quality system are described, and key elements are further elaborated in chapters on: Organisational management Standards, guidelines and references Documentation Training and staff development Assessment—

management of process and improvement Assessment—monitoring of performance. This is followed by a chapter explaining how to approach implementation in a step-by-step manner, with practical considerations and development of a roadmap. The document concludes with a discussion on the importance of continuous quality improvement. A comprehensive list of references provides information on additional resources.

Conclusion: Implementation of an effective QS in BEs will lead to potential benefits which include the safety, consistency, availability and quality of blood and blood products provided to patients, improvements in operational efficiency and cost-effectiveness, and better products and processes. Development of the Guidance document will support these efforts.

Working party session—Blood components WP

Cold stored platelets

WP03-L01 | Haemostatic function of cold-stored platelets—in-vitro studies

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As haemostatic cells, platelets perform many discrete functions, including identification of damaged endothelium, followed by adhesion, activation, initiation of aggregation and localisation of the coagulation process to result in a firm platelet plug and cessation of bleeding. In addition, platelets coordinate clot resolution, via retraction and fibrinolysis. Storage of platelets differentially affects these haemostatic properties. Platelet components for transfusion are conventionally stored at room temperature (RT; 20–24°C), which limits their shelf-life to between 5 and 7 days. This short shelf-life presents challenges to inventory management and may result in both unavailability and/or excessive wastage. Refrigeration, at 2–6°C without agitation (cold storage), is advantageous as it negates several of the limitations of RT storage, thereby affording an extension of the shelf-life to at least 14 days. Extensive pre-clinical laboratory studies have been carried out to investigate the impact of cold storage on platelet quality and function, with the general consensus being that cold-stored platelets represent an equivalent or superior haemostatic product compared to RT platelets. Cold-stored platelets are phenotypically different to platelets stored at RT, expressing a higher abundance of the activated form of the fibrinogen receptor (GPIIb/IIIa), which contributes to the potentiated agonist-induced aggregation responses. Cold-stored platelets also externalise phosphatidylserine and release procoagulant platelet microparticles, which increase proportionally as storage progresses. These factors likely contribute to the faster clot initiation and thrombin generation properties of cold-stored platelets.

The contractile function of cold-stored platelets is also maintained throughout extended storage, as observed by clot retraction studies. When considering cold-stored platelets, the storage solution and length of storage influences the extent of these phenotypic and functional changes. Specifically, storage in plasma increases the risk of aggregate formation, metabolic rate and activation status as storage progresses, compared to platelets stored in PAS. In addition, the composition of PAS may differentially affect aspects of *in vitro* platelet quality. A summary of the laboratory studies assessing the haemostatic function of cold-stored platelets will be presented.

WP03-L02 | What can animal models tell us about cold stored platelets?

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Stored platelets are transfused to prevent and treat bleeding. Platelets are currently stored at room temperature to maximize circulation time. Their quality declines during storage, and the risk of bacterial contamination increases, leading to short storage times. Cold-stored platelets (CSP) have recently regained attention because they could alleviate these shortcomings. In CSP, the platelet quality is better preserved, and the risk for bacterial growth is lower, allowing for prolonged storage times at the expense of shortened circulation time. However, testing of platelet quality *in vitro* has limitations, as platelets change in circulation upon transfusion, a phenomenon termed “rejuvenation.” Therefore, *in vivo* testing is indispensable. In humans, quantifying blood loss is notoriously difficult and requires carefully crafted clinical trials with suitable endpoints. Mouse models have a long history in platelet biology and are suitable for testing stored platelet quality. Previous publications highlight how these models can be used to assess circulation time, platelet quality changes upon transfusion, blood loss, adhesion to endothelial cells, and three-dimensional thrombus formation at the site of injury. Furthermore, mouse models have helped to characterize the immune response to platelet transfusions. This presentation will summarize recent developments and publications on mouse models in platelet transfusion medicine, focusing on cold-stored platelets. For example, mouse models have been used to characterize the cold exposure-response of platelets. In addition, they have been instrumental in describing the clearance mechanism of cold-stored platelets involving Von Willebrand Factor (VWF), hepatic macrophages, and hepatocytes. When developing approaches to circumvent the cold stage lesion, mouse models are frequently the first approach to test the efficacy of preventing premature clearance of CSP. Species differences between murine and human platelets, microstorage systems, and xeno-transfusion models will be briefly discussed as well.

WP03-L03 | Cold stored platelets—are they safe and effective

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Platelets for transfusion for the past 50 years have routinely been stored at room temperature due to improved survival and recovery compared to cold storage, which was the standard until the early 1970s. It has been known since that time that a cold stored platelet has improved hemostatic function compared to room temperature platelets. Despite RCT data from the 1970's indicating superior hemostatic function with cold storage of platelets, patients who need platelets for bleeding have been receiving room temperature stored platelets. Recent interest in hemostatic resuscitation principles for patients with severe bleeding has caused a reassessment of cold stored platelets for active bleeding. The FDA in the US has recently provided guidance that cold stored platelets up to 14 days of storage were permissible for active bleeding. A definitive trial is also ongoing in the US and Australia to determine the efficacy and safety of cold stored platelets in 1000 actively bleeding cardiac surgery patients (adults and children). This presentation will review the efficacy and safety data of cold stored platelets for patients with severe bleeding and will also summarize the CHIPS trial.

Working party session—Donors and donation WP

The plasma donor

WP04-L01 | How would we decide on a good plasmapheresis frequency?

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Background: Most plasma used for manufacturing plasma-derived medicinal products (PDMPs) such as albumin, immunoglobulins (Ig), and clotting factors is obtained from source plasma collected via plasmapheresis, the majority of which is contributed by the United States

where donors can donate plasma up to twice per week and up to 100 times per year. SUPPLY is a project co-funded by the European Union that aims to strengthen voluntary non-remunerated plasma collection capacity in Europe, to enable a stable and adequate supply of PDMPs.

Aim: To facilitate evidence-based plasma donor protection practices.

Methods: The following tasks were executed in one of the SUPPLY work packages (WP5): Collection of information on current plasma donor protection practices. Evaluation of the best available evidence of plasmapheresis and donor health by conducting. A scoping review and evidence gap analysis. A systematic review to identify, analyse, and critically appraise available studies that assessed the impact of plasmapheresis frequency on donor health. Develop a support tool on standardized donor vigilance data to be collected.

Results: Based on 18 complete survey responses from 17 countries: Annual donation limits vary from 12 donations per year (Luxembourg) to 26 (The Netherlands) to 60 (Germany) to 104 (USA). The minimum donation interval ranged from 2 to 28 days. Donation procedures (equipment, volume limit, flow rates, citrate-based anticoagulants) varied across organizations. Hydration advice (12 organizations), trained staff (7 organizations), and special attention to new donors (5 organizations) were the most common preventive measures. All organizations assessed total proteins (with varying limits). The scoping review identified 94 research articles and 5 registrations of ongoing studies of which 90% were observational studies. Different evidence gaps were identified. The systematic review included 4 cohort studies, 1 non-RCT and 2 RCTs (of which one is ongoing): The 4 included cohort studies were graded as providing very low-certainty evidence (due to methodological limitations and imprecise results because of limited sample sizes). The non-randomized controlled trial showed no statistically significant or clinically meaningful differences in total protein or immunoglobulins (IgG, IgA, IgM) after weekly compared to bi-weekly plasmapheresis during 6 months (low-certainty evidence). The completed RCT found that very high frequent plasmapheresis (2x/week) may result in a large reduction in ferritin and IgG levels (low-certainty evidence). Requirements were described for a support tool on standardized vigilance data to be collected (recording and analyzing plasma donor protection practices, standardized adverse event classification).

Conclusions—recommendations: A maximum of two plasma donations per month, pending sufficient evidence confirming the safety of higher donation frequencies. This recommendation is based on expert opinion and reflects the view of a majority of WP5 members. IgG levels should be monitored. Evidence of optimal IgG algorithms and test intervals is lacking. Urgent initiation of prospective studies to examine the health consequences of plasma donation at varying frequencies. Implementation of a register for standardized haemovigilance data on a mandatory basis. These recommendations are based on the use of the precautionary principle, placing donor safety first while awaiting further evidence.

WP04-L02 | Paid plasmapheresis from a donor point of view**Z Bacsadi¹**¹*Sociology and Social Anthropology, Central European University, Vienna, Austria*

One is not born a donor; one learns to become one. A healthy body is not enough, a donor is forged through bodily learning and communal experiences, adopting lifestyle choices, and absorbing institutional information. Donor bodies are cultivated individually and by social interactions (between donors, with staff and doctors) and by institutional scripts (Healy, 2000)—it also entails the formation of a habitus (Bourdieu, 1990). Becoming a plasma donor involves a certain level of agency, but also serious constraints. Donors' bodies carry therapeutic potential and risk that needs to be regulated. Plasma donation can be seen as a labor of love, gift-giving but also as actual labor, or even self-exploitation. Donors also enter a vast global supply chain which distances them from patients/end users of plasma therapeutics. It takes institutional discursive work to re-connect them through the persisting idea of gift, while being embedded in a global supply chain. The renewable nature of plasma, the frequency of donation provides possibilities and risks for donors; it can exhaust the body but can foster meaningful bonds with plasma center staff and with other donors. Commercial plasma centers also engage in a curious intertwining of medicalization and de-medicalization of the act: emphasizing some medical aspects can promote trust and feeling of safety, while downplaying them in other instances can decrease anxiety and the reluctance of donors. Even though plasma is a component of whole blood, plasma donation as a social practice is different from blood donation. Since plasma donation can involve financial compensation and the harvested material goes through more complex biotechnological treatments and global economic transactions until it reaches its recipients, the Titmussian model of 'blood as gift' and donation as citizen-making (1970) cannot be adopted seamlessly. In Hungary, plasma is procured by commercial organizations which creates tensions and interconnections between altruistic and economic motivations and narratives, both in the case of donors, staff, and plasma centers as institutions themselves. Rather than extinguishing each other, the two work in tandem in the different stages of procurement. Building on personal experiences of being a plasma donor—which can be seen as a form of carnal sociology (Wacquant, 2014) in which the body becomes both the object and means of investigation—I will briefly demonstrate how these tensions are generated and managed in a plasma center in Budapest, how plasma is presented both as gift and commodity, and how the fertile interconnections of the two ideas contribute to the functioning of commercial plasma centers. Several factors enable this blurring of gift and commodity: plasma as a material itself, the technical process of plasmapheresis, donor-staff interactions and the communication of commercial plasma centers as well. Plasma does not become a 'gift' or a 'commodity' merely by material or biotechnological factors: it is human actors and social relations that attach meanings to it. It is not possible to detach paid plasma donation from the socio-

economic context and healthcare system in which it is embedded; therefore, donor management entails social and moral aspects as well.

WP04-L03 | Play to learn—increasing knowledge about plasma donation through a serious game**A Ciausescu^{1,2}, E Merz^{1,2}, R Bekkers², A de Wit²**¹*Donor Medicine Research, Sanquin Research*, ²*Center for Philanthropic Studies, Vrije Universiteit Amsterdam, Amsterdam, Netherlands*

Many European countries, including the Netherlands, are facing plasma shortages. Lack of awareness among people regarding plasma donation is a potential explanation for the low number of donors. This lack might stem from limited learning opportunities for individuals to understand what plasma is and the importance of plasma donation. One approach to increasing awareness about plasma is using informal educational methods. To this end, we developed a serious game about plasma and plasma donation. We examined whether such an intervention can increase knowledge about plasma among children and adolescents in the Netherlands. In collaboration with the science museum NEMO (in Amsterdam), that allows museum visitors to participate in research projects, during July and August 2023 we invited 744 children aged 8-17 to play the game during their museum visit. Participants completed a pen-and-paper questionnaire before and after playing the game. The questionnaire included inquiries about their demographic characteristics, experience during the gameplay session and open questions about plasma and plasma donation. We assessed the effectiveness of the game by measuring the change in knowledge about plasma and plasma donation after the intervention. A paired t-test revealed a significant increase in knowledge post-game, the game being thus highly effective in educating the players about plasma donation. Neither the individual characteristics of the players (age, cultural background, gender, previous game experience, donor network embeddedness) nor the quality of the game experience affected this outcome, suggesting that the game effectively disseminates knowledge across a broad demographic of players. However, it was observed that children who already had some awareness about plasma had less knowledge gain than those who did not. A possible explanation could be that these children already possess some of the basic knowledge that the game was designed to impart. Nonetheless, the study shows that even among those with some prior knowledge serious games are a potentially viable avenue to increase knowledge about the subject among children and adolescents, thereby paving the way for a more sustainable and sufficient plasma donor pool in the future.

WP04-L04 | Blood and plasma donors—incentives and recruitment strategiesJ Georgsen¹¹*Department of Clinical Immunology, Odense University Hospital, South Danish Transfusion Service & Tissue Center, Odense, Denmark*

Background: Although the number of transfusions have been reduced during the last couple of decades, continued recruitment and retention of blood and plasma donors are important. Every year 10%–12% of donors cease to donate and have to be replaced. Furthermore, the increased usage of plasma derived medicinal products (PDMP), foremost immunoglobulin, and demand for self-sufficiency for plasma for PDMP have added to the need for repeat donors.

Aim: To describe the tools used to recruit and retain donors for a massive regional plasmapheresis program.

Methods: Due to a decrease in the amount of recovered plasma, it was decided in 2023 to begin a national plasmapheresis program. Due to supply problems during Covid, the responsible politicians came to a decision about self-sufficiency with plasma for PDMP. By law, the recruitment and retention of donors are the responsibility of an organization of volunteers supported financially by the health care system. However, this responsibility is carried out in strong cooperation with the local/regional blood establishments. To facilitate the need for new donors for plasmapheresis as well as replacement of whole blood donors, the following activities—among many others—have been initiated during the last decade: (1). Procurement of a van with on-line connection to the blood establishment computer system and venipuncture facilities, (2). Cooperation with the local Super League football club, (3). Cooperation with local businesses, (4). Increased cooperation with high schools, colleges, and university, (4). Increased active use of social media, (5). Continuous presence at local fairs and events, (6). Increased use of *donor recruits donor* tools. Furthermore, whole blood donors were recruited to donate plasma as they continued to donate whole blood. For retention, the policy of small tokens and gifts and a yearly event for donors with 50, 100, 200, 300 and so forth. donations was continued.

Results: From 2015 to 2023 the number of whole blood donations decreased in the region from 48,602 to 34,594 and locally from 10,238 to 4,283, whereas the number of plasmapheresis donations increased from 10,689 to 47,898 in the region and locally from 10,689 to 39,512. Thus, the total number of donations including platelet donations increased from 59,291 to 82,722. Regionally, the number of donors who donated in 2015 were 32,005 of which 28,393 donated whole blood and 3529 donated plasma. In 2023 the numbers were 25,839, 18,918 and 8635 respectively. Therefore, 19% fewer donors resulted in 40% more donations due to increased frequency of plasma donation compared to whole blood donation. The number of new donors in 2023 was 4038 regionally and 1794 locally. With the current technology it has not been possible to measure which of the many initiatives that have been most effective in recruiting new donors. The average number of whole blood and plasma donation was 1.8 and 5.4 per year respectively. The amount of plasma

delivered for fractionation increased from 13,473 kg (all recovered) in 2013 to 40,667 kg (of which 7998 kg were recovered) in 2023.

Conclusions: By using different methods the recruitment and retention of the necessary number of donors for implementation of a plasmapheresis program based on unpaid and uncompensated donors has been successful. A tool for measuring of the effect of these methods is urgently needed. For a given number of plasma donations the number of donors needed are much lower than for the same number of whole blood donations.

Working party session

Advancements in blood group genetics in the genomics era

WP05-L01 | The new ISBT database for blood group allelesN Gleadall

Abstract not available

WP05-L02 | Haplotype sequencingE van der Schoot

Abstract not available

WP05-L03 | Blood group phenotypes in times of geneticsN Thornton¹¹*IBGRL, NHS Blood and Transplant, Bristol, United Kingdom*

We are currently in the midst of the genomics era, where the intricate details of our DNA are being revealed at an unprecedented pace. The importance of studying and understanding blood group phenotypes has never been more crucial for ensuring the colossal amount of genetic data generated, can be deciphered and utilised, to make real improvements to health care. The advent of high-throughput genotyping technologies has facilitated comprehensive mapping of blood groups at the molecular level, offering unique insights into the genetic basis of blood group diversity and patterns of inheritance. However, it is imperative to acknowledge that genotyping, whilst instrumental in predicting blood group phenotypes, does not replace the significance of understanding the resulting phenotype. Phenotypes encapsulate the complex interplay between genes, epigenetics and environmental factors, influencing clinical outcomes and shaping the broader context of individual and population health. Acknowledging the limitations of only having genetic information is paramount in avoiding overreliance on genotyping as a sole determinant of blood group characteristics.

Over the past decade, we have witnessed the discovery of the molecular bases of several historical antigens, plus de novo blood group antigens, seeing the formation of 12 new blood group systems, a number which is set to rise again in 2024 and will continue to grow as long as humans evolve. Whilst the advancements in the molecular tools now available, has been fundamental to the discoveries, identifying a candidate gene is only one piece of the puzzle. The work required to prove that a gene is responsible for encoding a blood group antigen is usually difficult and time consuming, often involving many complex techniques, to ensure there is no doubt that the carrier molecule and responsible gene proposed, is indeed correct. The cruciality of this information and effort undertaken to obtain it, is what blood group genotyping is reliant on. If the study of the phenotypic expression is not robust and undeniable, then genotyping cannot be employed with confidence to predict the correct phenotype. The integration of genotyping technologies into the study of blood group phenotypes enhances our ability to predict and understand genetic variations. However, it is essential to appreciate that phenotype data is central to extracting meaningful insights from genetic data. Recognising the significance of blood group phenotypes is not merely about blood types; it is about unlocking the door to a deeper understanding of our genetic blueprint and harnessing the power of genomics. Ultimately, a genotype means nothing without the context of the resultant phenotype.

Working party session—Platelet immunology WP

A focus on platelet immunology

WP06-L01 | Platelet immunology and the Platelet Immunobiology Working Party (PIWP)

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Platelet Immunology and the PIWP

The blood of normal, healthy individuals has 150,000–450,000 platelets (plt)/ μ L. When platelet levels drop below 150,000/ μ L, a condition called thrombocytopenia, the risk of bleeding increases since platelets are important for maintenance of hemostasis. Once thrombocytopenia develops, it is critical to determine the cause so that appropriate therapy can be administered. Determining the cause of thrombocytopenia is difficult. Antibody-mediated thrombocytopenia can be diagnosed by testing patients' blood for platelet-reactive antibodies and typing the patient's platelets for human platelet antigens (HPA). Platelet antibodies react with specific glycoproteins (GPs) on the platelet surface. Antibody-coated platelets are cleared and destroyed by complement activation and/or clearance by

macrophages in the reticuloendothelial system (RES). Platelet immunology reference labs like those represented in the Platelet Immunology Working Party (PIWP) of the ISBT serve a critical role performing tests to detect platelet antibodies and antigens and aid in the diagnosis of thrombocytopenic patients. Five major immune thrombocytopenias diagnosed by labs of the PIWP include: (1) autoimmune thrombocytopenia (ITP), which occurs when platelet autoantibodies form and cause destruction of the patient's own platelets, (2) drug-induced immune thrombocytopenia (DITP) caused by autoantibodies that bind only to platelets in the presence of a drug that induced their production, (3) fetal & neonatal alloimmune thrombocytopenia (FNAIT), in which maternal IgG alloantibodies produced against paternally-inherited HPA on the fetal/infant's platelets cross the placenta and destroy fetal/neonatal platelets, (4) post-transfusion purpura (PTP), a rare, severe thrombocytopenic event that occurs when both platelet allo- and auto-antibodies are produced following a blood transfusion, and (5) platelet transfusion refractoriness (PTR), which develops when patients become immunized against human leukocyte antigens (HLA) and/or HPA following a blood or platelet transfusion, making it difficult to find compatible platelets for transfusion. Techniques used for detection of antibodies and antigens include immunofluorescence by flow cytometry, monoclonal antigen-capture, and HPA genotyping. The PIWP currently supports 31 lab members representing 20 different countries on 5 continents. It aims to enable collaborations in platelet immunology by bringing together members working in a diagnostic, research, and/or clinical laboratory setting. One way to accomplish this is by holding the biennial International Platelet Immunology Workshop. The workshops involve shipping each lab samples selected to challenge their ability to detect platelet antibodies and antigens. Results are reviewed and discussed at the ISBT PIWP business meetings. The PIWP is also responsible for establishing HPA nomenclature. There are currently 41 HPA expressed on 6 different platelet membrane glycoproteins GPIIb/CD41, GPIIIa/CD61, GPIb α /CD42b, GPIX/CD42a, GPIa/CD49b, and CD109. For a current list see: <https://www.versiti.org/products-services/human-platelet-antigen-hpa-database>.

WP06-L02 | Fetal and neonatal alloimmune thrombocytopenia—the Quebec story

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Fetal and neonatal alloimmune thrombocytopenia (FNAIT) is an important cause of death or disabling condition in neonates, sequels being present sometimes for their lifetime. The direct cause of FNAIT is well known; maternal alloantibodies directed against immunologic determinant on glycoproteins (GP) called human platelet antigens (HPA), inherited from the father, induce the destruction of fetal platelets, leading to a high risk for foetal hemorrhage. However, the susceptibility to develop such immunity and the specific prevalence of

antibodies against particular HPA is more nebulous. Distribution of FNAIT and antibody specificities around the world can be different as prevalence of some HPA can differ from one country to another. Individuals from European descent are known to develop mainly anti-HPA-1a antibodies although people from Asian or African descent are more prone to develop anti-HPA-4 and anti-CD36 antibodies. Despite this difference in distribution, it is well accepted that FNAIT induced by an anti-HPA-1a antibody can lead to the most severe outcomes in the foetus and newborn, including intracranial hemorrhage and fetal death. Antibodies to other HPAs are usually considered much less deleterious for the baby, leading to a thrombocytopenia with a much higher platelet count and less risks for hemorrhage to happen. However, many cases of very low platelet count, foetal death, and vital organ hemorrhage due to antibodies directed against HPAs other than HPA-1a have been seen at Hema-Quebec. We report here the distinct prevalence of anti-HPA-5b in FNAIT investigated in the province of Quebec. Our observations go against what has been reported for European descents mothers in many publications. An unexpected higher occurrence of FNAIT caused by anti-HPA-5b antibody raises interrogations concerning the genotyping, screening and antibody identification practices targeting only the HPA-1b. Consequently, the practice described for the province of Quebec includes a screening for all major HPAs, including HPA-1 to 5, and an identification of antibodies against HPA-1 to 5 and HPA-15, as well as genotyping of the mother and father for HPA-1 to 10 and HPA-15. This allowed us to identify disparities between the mother's and the father's genotype at first pregnancy when a FNAIT is identified or suspected, and protect further pregnancies of affected women from dramatic outcomes caused by anti-HPA-5b antibody as well as other anti-HPA antibodies including anti-HPA-1a. Also, the prevalence of HPA-5b in Quebec has been evaluated to explain the observed phenomenon.

WP06-L03 | Drug-induced immune thrombocytopenia

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Drug-induced immune thrombocytopenia (DITP) presents a significant challenge in clinical practice, necessitating a comprehensive grasp of its mechanisms, detection, and treatment. DITP manifests as a sudden decline in platelet counts triggered by pharmacotherapy. While not uncommon, DITP is likely underdiagnosed. Clinical criteria have been established to aid physicians in identifying patients with a high suspicion of DITP. However, diagnosing DITP remains challenging due to the absence of standardized diagnostic tests. Given its potential for severe complications, rapid diagnosis and management of DITP are imperative. Clinicians should consider DITP as a differential diagnosis, particularly in patients presenting with abrupt and severe thrombocytopenia.

WP06-L04 | Immune Thrombocytopenia (ITP)

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Immune thrombocytopenia (ITP) is an autoimmune disease characterized by an isolated thrombocytopenia and variable phenotype as some patients suffer no bleeding whilst others have severe bleeding which may be fatal. This variability probably reflects the disease's complex pathophysiology; a dysregulated hyperreactive immune effector cell response involving the entire adaptive immune system (e.g. B and T cell subsets) that leads to platelet and megakaryocyte (MK) destruction. It appears that these effector responses are due to a breakdown in immune tolerance, and this is characterized by defects in several immunosuppressive cell types. These include defective T regulatory cells (Tregs), B regulatory cells (Bregs), Myeloid-derived suppressor cells (MDSC) and natural killer (NK) cells, all of which are all intimately associated with antigen presenting cells (APC) such as dendritic cells (DC). The loss of this immunosuppressive axis allows for unchecked auto-reactive T cell and B cell activation leading to the development of auto-antibodies and cytotoxic T cells (CTL) which can directly destroy platelets in the periphery and inhibit MK platelet production in the bone marrow (BM). This talk will focus on the effector cell mechanisms in ITP and highlight the defective immunosuppressive axis that appears responsible for this platelet-specific immune hyperreactivity.

WP06-L05 | Antibodies against CD36 cause thrombocytopenia

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CD36 or GPIV (also known as Nak-a) is a molecule expressed on various cell types, including platelets, monocytes, macrophages, microvascular endothelium, adipocytes, cardiac and skeletal myocytes, epithelia of the breast, kidney and gut, erythroblasts and myeloblasts. Its expression is also confirmed on mature red cells (*Blood*, 1992; 180 (8): 2105-2114), and recently it was registered as a new red cell antigen (*Vox Sanguinis*. 2024;1-9). Type I CD36 deficient individuals may produce anti-CD36 antibodies, and if platelets are targeted, clinical conditions of thrombocytopenia, such as fetal-neonatal alloimmune thrombocytopenia (FNAIT), and platelet transfusion refractoriness (PTR), may occur. In case monocytes or endothelial cells are the target, transfusion-related acute lung injury (TRALI) may occur, and in case red cells are the target, fetal anemia may occur. Severe FNAIT cases due to anti-CD36 antibody have been reported, most of the cases associated with severe thrombocytopenia, severe anemia and hydrops fetalis. CD36 deficiency is a rare condition found in a relatively higher incidence in Asian and African population. The genetic background of CD36 deficiency varies, with different SNPs identified in different

populations. Interestingly, the most frequently identified SNP in Japanese is known to not be associated with antibody production, attributed to a weak expression of the antigen. Also, interestingly, it seems that CD36-negative individuals may naturally produce antibodies without an evident immune stimuli. Management of PTR due to anti-CD36 antibody requires transfusion of CD36-negative platelets, which in many countries is not easy to obtain. In Japan, the relatively high incidence of type II deficient individuals, who lack CD36 expression only on platelets, allowed the development of a registry system, which makes supply of CD36-negative platelets for the management of PTR feasible. Since CD36 is also expressed on erythroblasts and myeloblasts, we have followed cases of type I CD36 deficient individuals requiring allogeneic hematopoietic stem cell transplant (HSCT), and observed that in case antibody titers are appropriately managed prior to transplant, it can be successfully conducted irrespective of the CD36 type of the donor. Since CD36 is expressed not only on erythroid and myeloid progenitors, but also on microvascular endothelial cells and various epithelial cells, it is possible that CD36 antibodies may be involved in graft rejection in organ transplant, and this needs to be carefully followed.

WP06-L06 | Anti-CD36 and more—results from the PIWP workshop

N Nogues Galvez

Abstract not available

Working party session—Rare donors WP

The need for rare blood knows no borders

WP07-L01 | The Ibero-American (GCIAMT) rare donor registry—a vision to reality

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Until recently, only two national programmes for donors with rare blood groups existed in Ibero-America, one in Brazil (2014) and the other in Spain (2005). In 2018, the Ibero-American Cooperative Group for Transfusion Medicine (GCIAMT), the scientific society that brings together professionals linked to transfusion medicine in Ibero-America decided to explore the possibility of establishing a register of rare blood donors for those countries where such an initiative has not already been implemented. In order to find out whether we could rely on professionals prepared to carry out this project, we undertook an extensive 30-question

survey among GCIAMT associates. We enquired about the techniques used to investigate irregular antibodies in patients, pregnant women, and donors, and for red cells typing (serological, molecular), the availability of plasma, red blood cells (RBC), or DNA samples of interest, and the capability to cryopreserve RBC units or blood samples. We received 20 responses from people in 12 countries. It was thrilling to confirm that there were already professionals in different countries working on issues of rare blood groups; however, it was needed to help them to bring their efforts and their achievements to light. Finally, 6 people from 5 countries were contacted to start working on this project: Dr Carlos Cotorruelo, Silvia Margineda, Senior MT (Argentina), Dr M^a Antonieta Núñez (Chile), Paula A Gaviria, Senior LT (Colombia), Dr Hector Baptista (Mexico) and Carlos Penalillo, Senior MT (Peru). Recently, Igseda Valdés, LT (Panama), Dr Romi Alcaraz (Paraguay) and Fernanda Bangueses, Senior MT (Uruguay) joined the Working Party, increasing the number of participating countries to 8. After four years of the Working Party establishment and coordinated work, the GCIAMT registry has now incorporated 86 donors with a phenotype of interest that has been serologically and molecularly characterised. All donors have been informed and showed a willingness to cooperate when requested. One hundred and fifty more donors are awaiting to complete their unequivocal characterisation in order to be informed. So far, three alloimmunised patients were transfused with blood from rare donors belonging to our registry (2 k-negative and 1 r'r'). While we were building this registry, some other important things happened: the protocol for freezing rare RBC samples in pearls has been distributed among members and many have implemented it already, the use of molecular techniques is progressively spreading, and IDCBS (Bogotá, Colombia) began to cryopreserve RBC units. In addition, a national WP on Rare Donors has already been set up in 5 of the 8 countries, and a common database has been created to support data entry and management. The ISBT Rare Donors Working Party, ISBT Board of Directors and the ISBT Executive Committee have also supported this project since they became aware of it. Some members of the WP have generously donated various antisera which will allow us to expand the repertoire of blood groups to be typed in our Ibero-American donors. A new survey has recently been carried out to capture new professionals and countries. Sharing knowledge, experience and efforts from different countries is the only way to face the challenge of building the most complete registry of donors with rare blood phenotypes in Ibero-America.

WP07-L02 | Experiences that promote and strengthen the Ibero-American registry of donors with rare blood donors

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In recent years, Latin America has witnessed an increasing number of patients requiring transfusions with rare blood types. Colombia reported a notable case in 2017 when a pediatric patient with the Bombay phenotype required blood imported from Brazil. Other rare

phenotypes, such as Rhnull, D^{-/-}, Kp(b), k⁻, Di(b⁻), and K₀, have also been identified, posing a challenge to the country's blood supply and driving the search for alternative approaches to allogeneic blood transfusion. Several strategies have been implemented in Colombia to address this challenge, such as the Unique Donor Program developed by the District Institute of Science, Biotechnology, and Innovation in Health (IDCBIS). This program has determined the extended blood group profiles of more than 2700 blood donors, has designed loyalty strategies for donors with rare blood phenotypes, and is currently implementing red blood cell cryopreservation. Moreover, it has recently been awarded government funding to expand its activities over four years to screen 9000 donors, conduct the first Bombay phenotype screening in Colombia, and cryopreserve at least 30 red blood cell units. These efforts have not been isolated but synchronized with comparable initiatives in the Rare Donor Working Party of the Ibero-American Cooperative Group on Transfusion Medicine (GCIAMT-WP). This group has consolidated a network of professionals from Colombia, Peru, Mexico, Paraguay, Uruguay, Argentina, Chile, and Panama under the mentoring of Spain and Brazil, who work collaboratively on building an Ibero-American registry of rare blood donors. Additionally, the support and involvement of the Rare Donor Working Party of the ISBT (ISBT-WP) has played an essential role in promoting the GCIAMT-WP activities. During 2023, rare donor programs in the United Kingdom, France, Germany, and Sweden donated rare antisera, such as anti-Vel, anti-PP1Pk, anti-Jra, and anti-Lub. These sera will help screen high-incidence antigens not yet included in Latin America's routine red blood cell phenotyping. Moreover, the ISBT-WP funded the shipment of antisera from Europe to Colombia and from Colombia to different Latin American transfusion medicine centers. The Unique Donor Program and the Advanced Immunohematology Unit of the IDCBIS have played a crucial role in GCIAMT-WP activities by fostering mentorships within the group and carrying out different tasks such as confirming donors from countries lacking molecular immunohematology laboratories, receiving antisera donated by European programs, aliquoting antisera for distribution, designing guidelines for the use and storage of antisera, as well as providing technical, administrative and logistical support for the distribution of antisera to Latin American. Despite the multiple challenges, progress in constructing the Ibero-American registry of rare blood donors has been remarkable thanks to networking, mentoring by international experts, the enthusiasm of new professionals willing to collaborate towards a common goal, and the commitment of Latin American reference transfusion medicine centers. These initiatives are essential to meet the transfusion needs of patients with rare blood phenotypes and represent a significant step forward for immunohematology and transfusion medicine in the region.

WP07-L04 | Experiences that promote and strengthen the Ibero-American registry of donors with rare blood

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Argentina has never had a government or scientific society initiative to establish a Rare Donor Program. Nevertheless, some professionals maintained a list of donors with unusual phenotypes in their laboratories to address potential needs of their own patients. In 2019, two Immunohematology laboratories (one in La Plata city and the other in Rosario city) were invited to join the Working Party on Rare Donors of the Ibero-American Cooperative Group for Transfusion Medicine (GCIAMT). Both laboratories started working collaboratively, performing extended erythrocyte typing in donors from La Plata and confirming the rare phenotypes by molecular biology in Rosario and in 2021 we made the first contributions to the GCIAMT registry while simultaneously building our own database. This involved intensive efforts using the institutions' resources, gradually gaining visibility throughout the country. During 2022 we were contacted on various occasions in a disorderly and confusing manner, requesting compatible blood for two patients carrying anti-Cellano and for a pregnant woman with anti-Di^b. Following numerous phone calls to clarify the requirements, the demands for k(-) blood were successfully managed using our emerging local registry, but we did not have Di(b-) donors registered. Fortunately, the transfusion for the pregnant patient ultimately proved unnecessary. These three clinical scenarios clearly showed the importance of establishing a national registry of rare donors to efficiently address complex and often urgent transfusion needs. Subsequently, we focused our efforts on establishing a local Working Party aimed at promoting the search for rare donors and devising a protocol for requesting rare blood. In collaboration with the Argentine Association of Hemotherapy and Immunohematology, we conducted a nationwide survey to identify enthusiastic and qualified colleagues interested in joining the Working Party. At the end of 2023 the group was formally established, with our primary goal being the creation of the national registry of rare donors. We have arranged online conferences to inspire colleagues from all over the country in the quest for rare donors and offered our laboratories as reference centers for studying complex immunohematological situations. In this regard, the Rosario laboratory verifies rare phenotypes detected in Paraguay and Uruguay, where molecular techniques have not been fully implemented yet. On the other hand, INCUCAI, a national organization that oversees organ donation and transplant activities in Argentina, has expressed interest in our initiative and currently, we are collaborating to streamline the import and export procedures for blood units. In addition, we continue our collaboration between the La Plata and Rosario laboratories performing molecular screening to detect Di(b-) and Co(a-) donors. Recently, following the donation of antisera

targeting high-frequency antigens by the ISBT, we invited blood banks from various regions of the country to engage in exploring of rare phenotypes. We requested them to send donor samples to our laboratories for serological studies. With scarce financial resources, we have implemented various initiatives that started engines of change and attracted the interest of young professionals to delve into the pursuit of rare donors. We currently possess a modest national registry of rare donors, whose phenotype was examined serologically and molecularly, and the donors informed.

WP07-L05 | The global view—ISBT International rare donor panel 2024

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Since the inception of the International Rare Donor Panel (IRDP) in 1965, the core purpose of the panel has been to locate and facilitate the exchange of rare blood between countries, when the blood type cannot be sourced nationally, for those patients in need of it. Though this core purpose has remained the same, the IRDP has undergone significant changes to evolve and adapt to the ever-changing landscape of rare blood provision, driven by scientific advancements in the field of transfusion medicine and information and communication technology. The International Blood Group Reference Laboratory (IBGRL) is responsible for the administration of the IRDP and has been since the panel came into existence. IBGRL works closely with the International Society of Blood Transfusion (ISBT) Working Party on Rare Donors, to ensure the IRDP grows to meet the needs of a progressive international transfusion community. The IRDP database underwent a major upgrade in 2020 to safeguard this precious resource. It is already evident, amidst the increasing use of blood group genotyping platforms, that the IRDP must continue to adapt further. Over the last few years, requests for allele matched donors, most notably rare RH variant alleles, has become apparent and is likely to increase further as advanced technologies and therapies play a more significant role in matching donors and recipients. It is crucial the advancements are embraced and factored into future developments of the IRDP, however, it is essential that the core purpose of the IRDP remains at the forefront of any future iterations of the panel, to ensure it remains relevant for all who may need to use it. Currently the IRDP consists of 14,482 donors plus inventory of 8210 frozen units, from 26 countries, involving 37 contributors, encompassing five continents. It is important to understand that what is considered rare in one region of the world may be more common in another region, highlighting the importance of respecting the unique genetic makeup of different populations and taking a globally informed approach to addressing rare blood needs. The ISBT Working Party on Rare Donors is committed to facilitating any countries, looking to establish a new rare donor program, or build on a nascent program, by fostering collaboration, sharing experiences and offering mentorships. Through this collaboration, the IRDP inventory will grow and become

even more diverse, enabling provision of blood of the rarest types to those equally rare patients who may need it.

Working party session—Clinical transfusion WP

Global variations in transfusion practices and educational approaches

WP08-L01 | Global variation in blood centers practices supporting neonatal transfusions

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Preterm neonates are known to receive high rates of transfusion. Their developmental immaturity, changing physiology, developing hemostasis and small blood volume makes them a special group requiring special care and attention from the blood centers. There are separate guidelines for this age group of patients which have guided many centers worldwide in standardizing the practices. (New et al., 2016). Various surveys/ forums have shown international variations among the blood centers from various countries showing marked differences in policies and practices while supporting neonatal transfusions. (Bruun MT et al., 2019; Reeves et al., 2021; Arora et al., 2023) The majority of variation is seen in the aliquoting of blood components, age of the unit selected, use of anticoagulants/ additive solutions (SAGM), preparation of aliquots, and selection of ABO compatible unit. The specific scenarios requiring phenotype-matched units, CMV-screened units, irradiation units availability, and pathogen reduction of blood have also shown marked variation across the globe. Apart from unit allocation and modification in the blood center, surveys have also reported the variation in indication for transfusion of various blood components, thresholds used, volume for transfusion ordered as well as practices for transfusing these aliquots at the bedside. (Bruun MT et al., 2019; Scrivens A et al., 2023; Arora et al., 2023) Numerous country-specific publications from the USA (Reeves et al., 2021), and India (Arora et al., 2022) also highlighted the variations among the various blood centers within the country. There is a growing need to harmonize these global practices and have consensus at least at the regional level. This is necessary for uniformity in the data reported and analysis and to ensure best practices for this vulnerable group of patients.

WP08-L02 | e-Learning in transfusion education

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The landscape of education and training in healthcare is rapidly changing, with e-learning gaining significant attention in transfusion medicine (TM) and other medical fields. Effective stakeholder management and compliance prioritization are crucial for successful TM e-learning program implementation, particularly in hospital settings. Incorporating program evaluation is essential to ensure learning needs are met. However, there is limited published data on e-learning programs in TM education. The Clinical Transfusion Working Party conducted several studies to explore the use of e-learning in the field of TM. A survey among ISBT members revealed widespread utilization of in-house developed e-learning programs in TM, catering to various audiences and topics (Al-Riyami AZ, Transfusion Medicine. 2022). Many of these programs were developed during COVID19 pandemic. Despite this abundance, there was a gap in demonstrating their effectiveness in providing impactful, cost-effective, and sustainable education. We also performed a scoping review of the literature and found numerous programs incorporating learner knowledge assessment, yet only four publications detailed the impact of e-learning on patient or patient blood management (PBM) specific outcomes (AZ Al-Riyami, Vox Sanguinis. 2023). Moreover, there is insufficient information on development cost, resources required, and methods used to assess the effectiveness, and the impact of implementation on knowledge retention, practice change, and patient outcomes. Finally, we performed a qualitative assessment, interviewing program developers and educators involved in different TM e-learning programs from different countries worldwide, and highlighted limited evaluation of e-learning programs, including their effect on organizational practices and patient outcomes. This study also explored facilitators and barriers for implementing e-learning programs (AZ Al-Riyami, Vox Sanguinis. 2023). In this presentation, we will present the outcomes of these three studies and the recommendations made for future research to assess the impact and the cost-effectiveness of implementing TM e-learning programs on transfusion practices in comparison with other methods of learning.

WP08-L03 | International forum on global patient blood management

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Patient blood management (PBM), a patient-oriented, multi-disciplinary, systematic, and evidence-based approach to improving patient outcomes by reducing unnecessary transfusions, is gaining global recognition. Although most commonly used for elective perioperative patients, PBM has applications in other settings, such as obstetrics and critical care. PBM is usually described in terms of three pillars, which are optimizing patients' blood, minimizing blood loss, and reducing unnecessary allogeneic transfusions. The extent to which these strategies have been implemented differs significantly by jurisdiction and appears contingent on a formal PBM program. Many low and lower-middle-income countries (LICs and LMICs) are struggling with the unmet needs for transfusion while the high-income countries are dealing with rising blood system costs, prompting a greater focus and investment in PBM. LICs and LMICs face the highest burden of anemia globally and the greatest gap between the supply and demand of blood. Hence, PBM in LMIC and LIC should be considered a necessity rather than a luxury. This IF aims to identify the variations in PBM practices across the globe, as well as local challenges and successes. With this knowledge, we may be able to identify key interventions that can be universally applied to facilitate PBM implementation. The IF survey was developed by the PBM sub-group of the Clinical Transfusion Working Party of the International Society of Blood Transfusion (ISBT). The survey was distributed electronically to the ISBT members from 20 countries, covering a wide geographical range as well as low to high-income countries. The members were representatives of the following countries: Australia, Brazil, Canada, China, Congo, Estonia, Germany, India, Israel, Japan, Italy, Mexico, Macedonia, Nepal, Oman, Portugal, South Africa, Sri Lanka, United Kingdom (UK) and United States (US). We collected demographic information, institutional description, details on employed PBM interventions, associated practices, and successes within the PBM pillars, as well as challenges faced during PBM implementation from respondents. The insights gained from this IF will facilitate a comparison of PBM practices in contrast settings and help identify implementation successes and challenges. This, in turn, will contribute to developing effective and universally applicable implementation strategies for the future.

Working party session— TTID WP

Infectious disease panorama

WP09-L01 | Transfusion transmitted infectious diseases WP— update and new initiatives

B Custer

Abstract not available

WP09-L02 | Nucleic acid testing—reflections on blood safety around the world

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Background and Objectives: Blood operators employ nucleic acid amplification testing (NAT) as a valuable tool to detect known pathogens to reduce the risk of transfusion-transmitted infection.

Aim: To review global blood donation NAT.

Materials and Methods: ISBT Transfusion-Transmitted Infectious Diseases Working Party (WP) developed a survey on use of blood donation/donor NAT. The survey was distributed through outreach to WP members and others in as many countries as possible. Content focused on NAT use in 2019 for comparison with data from 2008 (Roth et al, Vox Sanguinis, 2012).

Results: Forty-three responses were received from 32 countries, with 38 respondents performing NAT on donations for at least one virus. NAT adoption was attributable to government mandate for most respondents. Multiplex HIV, HCV, and HBV NAT was performed in most countries, while selective testing was reported for WNV, HEV, and ZIKV. Individual Donation (ID-) NAT was used for HIV, HCV, and HBV by ~50% of respondents, while HEV NAT using mini-pools (MP) was reported by 83% of respondents. Confirmatory testing of NAT-yield samples was generally performed by NAT on a sample from the same donation or by NAT and serology on samples from the same donation or follow-up sample. Of the survey respondents performing NAT for at least one virus, 25/38 were from high-income countries, 10/38 were from upper-middle income countries and 3/38 were from lower-middle income countries. Comparing ID- and MP- NAT for HIV within each country between 2008 and 2019, approaches were maintained, smaller MP size adopted, or changed from MP- to ID-NAT. No country moved from ID- to MP-NAT or increased MP size. NAT-reactive donations were detected for all viruses tested in 2019 (proportion of NAT-reactive donations were 0.0099% for HIV, 0.0063% for HCV, 0.0247% for HBV, 0.0323% for HEV, 0.0014% for WNV,

and 0.00005% for ZIKV). Rates of HIV, HCV, and HBV NAT reactivity varied by country. HEV NAT was performed on donations from many European countries and Japan. Nearly all WNV and ZIKV NAT was performed in the United States and Canada. Multiplexed platforms are increasingly available and being adopted, and expansion of NAT to detection of parasites is occurring. For those countries that have not adopted NAT, cost was the main driver in not performing HIV, HCV, and HBV NAT despite these countries having higher incidence/prevalence of HCV and HBV compared to respondent countries using NAT.

Conclusion: Based on our survey and previous reports, approaches for NAT use are evolving. Increased NAT use with progression to smaller MP sizes/ID-NAT and growing focus on further multiplexing are evident. The survey provides insights into confirmatory testing approaches used for NAT yields. Considerable variation in NAT-reactivity and NAT-yield rates were observed between the different geographical regions. NAT contributes to improving blood transfusion safety globally; however, there is a need to overcome economic barriers for regions/countries not performing NAT.

WP09-L03 | Hepatitis B—international perspectives on application of blood donor data to public health surveillance

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Background: Hepatitis B virus (HBV) is predominantly transmitted from mother to child at birth, by sexual contact and by blood-blood contact including blood transfusion. While acute infection often resolves in infected adults, chronic HBV usually develops if infection occurs in early childhood. Although chronic infection is not curable health consequences such as cirrhosis and liver cancer are reduced by treatment. The World Health Organization has set a goal of eliminating HBV infection by 2030. Strong surveillance is essential to monitor progress towards this goal. Globally, blood donors are tested for markers of HBV infection to ensure the safety of the blood supply. These data may also be valuable for public health surveillance. Blood donors are selected to ensure low risk, and are unlikely to donate if they know they have HBV infection. Thus, blood donors are a population subset very unlikely to be tested in healthcare settings.

Aims: The Surveillance, Risk Assessment and Policy Subgroup (SRAP) of the ISBT Transfusion Transmitted Infectious Diseases Working Party aimed to evaluate public health activities in developed countries and evaluate how blood donor data may contribute to public health surveillance.

Methods: The countries included were Australia, Canada, France, the Netherlands, the United Kingdom and the United States. Information on vaccination and screening policies, public health surveillance and blood service data collection were obtained from web searches, published and grey literature and the study team. This information was compared across countries.

Results: All countries in the study have childhood vaccination programs with variable implementation dates. All countries also have

antenatal testing. All countries have direct acting oral medications available for treatment. Public health HBV surveillance is generally based on case reporting. Past infections (people positive for anti-HBV core total [anti-HBc] in the absence of HBV surface antigen [HBsAg] and HBV deoxyribonucleic acid [DNA]) are not reported. Most countries have carried out prevalence studies and have published modelled HBV prevalence and incidence estimates. In some countries these studies are sporadic, while others have regular surveys. In most cases the sample size is not adequate for regional surveillance. All blood services in the study carry out universal HBsAg and HBV NAT screening of donations, and while all have anti-HBc testing, policies vary from universal, to all donors once, to selective testing. All blood services report HBV positive donors to public health services for health protection as required by law and report surveillance results. Otherwise, collaboration with public health tends to be sporadic.

Conclusions: Blood donor HBV data lend insight into the low-risk population unlikely to seek testing. Strengths of blood donor data include active surveillance, consistent testing methodology across sites, and anti-HBc testing data. Anti-HBc testing provides data on past infections not usually available in public health surveillance and provides insight into occult HBV infection. Some barriers to utilization of blood donor data for public health surveillance include variable integration of blood services into public health, low prevalence detected, limited follow-up testing, lack of information on the outcome of notifying public health services and limited data on donor risk factors.

WP09-L04 | Platelet component safety in the era of advanced bacterial screening and pathogen inactivation

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Platelet component (PC) safety has improved with the implementation of blood donor skin disinfection, first aliquot diversion, and either product screening for bacterial contamination or treatment with pathogen inactivation (PI) technologies (PIT). A large volume delayed sampling (LVDS) algorithm to screen PC with culture methods was first implemented by the NHSBT in the UK and then adopted by other centers in North America, Europe, and Australia. Conversely, facilities with a proactive approach have implemented PC treatment with PIT. The efficacy of these mitigation strategies depends on the timing between donation and PC screening or PI treatment and the growth characteristics of the bacterial contaminants. The remaining safety risk associated with bacterially contaminated PC post-implementation of LVDS or PIT is reviewed herein. Hemovigilance data and published reviews and reports related to septic events resulting from the transfusion of bacterially contaminated PC post-implementation of mitigation strategies were collected and summarized. Contaminated PC units that escaped detection or PI treatment were identified by visual inspection prior to transfusion, during investigation of septic transfusion reactions, or during quality

control testing of expired units. Special focus was given to recent breakthroughs involving PC screened with culture systems using a LVDS algorithm or treated with PIT. Data analysis showed that centers which initially implemented PC testing with one culture bottle and sampling at 24h post-blood collection, and then switched to a LVDS PC testing algorithm, have seen a reduction but not an elimination of false negative screening results (Gilles and Bernier, Ann Blood, 2021). The bacteria most frequently isolated during investigations of false negative screening cases are *Staphylococcus aureus* and coagulase negative staphylococci (Ramirez-Arcos et al, Transfusion, 2019; SHOT report, 2022); however, there are recent reports of missed detection of *Bacillus mobilis* in a center that screens PC with a LVDS algorithm and a new generation culture system. Following a proactive approach, several organizations have implemented PIT to treat PC demonstrating a significant reduction in the incidence of septic transfusion reactions (Pitman et al, Transfusion 2023). Nevertheless, PI-treated PC are not infallible to breakthrough bacterial contamination as exemplified by recent cases involving *Bacillus cereus*, *S. aureus*, or a mix of microorganisms (FDA report, 2021; Richard, Vox Sang, 2023). Overall, hemovigilance data show that PC safety has been enhanced by the adoption of advanced screening methods or treatment with PIT. There is, however, a very low residual risk posed by organisms with slow growth rates in PC units screened with a LVDS protocol. In contrast, fast growing or sporulated bacteria present a challenge for an effective PI treatment. Furthermore, bacteria could be introduced after PI treatment raising awareness to issues with bacterial environmental contamination. Unexplored is also the potential risk posed by endotoxins or exotoxins, bacterial products that could be present in PC and are neither detected with screening methods nor inactivated with PIT. Although no mitigation strategies ensure PC sterility, current approaches are highly effective to increase the safety of transfusion patients.

WP09-L05 | Climate change and parasites—bugs on the move

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Background: Climate change encompasses regional and/or global changes in the average temperature, humidity, and precipitation patterns over seasons-years. This process is often driven by both natural processes and human activities. Outcomes of climate change include not only changes in infectious disease epidemiology but also rapidly rising sea levels, resource scarcity, human refugee crises, and human conflict. Blood operators are not only responsible for ensuring the safety of the blood supply but also play an important role in supporting public health surveillance impacting public health, human health, and animal health (e.g., OneHealth). In 2022 the ISBT parasitology subgroup reviewed the peer reviewed and gray literature for how climate might impact the epidemiology of parasitic diseases and the downstream effects on the safety of

the blood supply. This exercise led to the publication of a manuscript entitled “Climate change and parasitic risk to the blood supply” (Transfusion. 2023 Mar;63(3):638-645. doi: 10.1111/trf.17234). At that time no major studies were identified that linked the impact of climate change on vector-borne parasites to the practices of blood operators. The group determined that although it was too early to provide specific recommendations to blood operators, the blood community should remain vigilant to climate change, its associated effects on the epidemiology of vector-borne parasites, and how this might impact blood transfusion safety.

Aims: This session will introduce concepts and generate discussion on three topics. How will climate change potentially impact on parasitic disease risk to the blood supply as well as the general population? How these “bugs on the move” might have broader impacts on the general activities of blood operators? How can blood operators begin to plan, prepare, and respond to “bugs on the move” and changing risks to the blood supply?

Methods and approaches: As a case study, this session will present how climate change might be changing risks of transfusion-transmitted malaria in non-endemic countries. This will include an emphasis on how these changing risks might impact health systems in these regions. A matrix of potential blood operator medical/scientific and operational activities impacted by climate change and changing malaria risk will be presented to the session participants. In the discussion period, participants will be asked to contemplate and comment on these changing activities and offer alternative viewpoints or thoughts on these risks. Knowledge “buckets” introduced to participants will include laboratory testing technology, surveillance, research and development, donor engagement, operational resource allocation, donor recruitment/deferral strategies, general communication strategies, and non-transfusion medicine health care practitioner engagement.

Expected outcomes of this session: Opportunities for knowledge translation: This information discussed in this session will be used to formulate a white paper focussed on the how blood operators can begin to plan, prepare, and respond to “bugs on the move” and changing risks to the blood supply. Participants will be challenged to provide ideas on how ISBT can lead further activities to generate.

Posters

Management and organisation—organisational issues

P001 | Reducing patient's wait time for blood transfusion in an outpatient setting - a multidisciplinary approach in a local tertiary hospital

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Background: National University Hospital Singapore is a local tertiary hospital which offers tertiary services including outpatient oncology clinic which runs outpatient transfusion services for selected patients such as Thalassemia patients. This service allows patient to leave the hospital post transfusion, thus saving patient's money and time from overnight hospital stay. Despite best efforts to allow patient to leave for home, the average wait time was 3 h from blood ordering to start transfusion, leading to an increased in complains due to prolonged wait time.

Aims: To reduce wait time, a multidisciplinary team including Blood Transfusion Service (BTS) scientists, nurses, clinicians, and ancillary staff performed a Root Cause Analysis (RCA) to identify areas for improvement. Long wait times were due to delay in blood ordering leading to delay in orders given to BTS thus and delaying the release of blood products by BTS for transfusion. Further delays were expected for patients with multiple red cell alloantibodies, due to longer turnaround time for Group and Crossmatch (GXM) results as well as logistic delay in supply as the blood will need to be ordered and delivered from the supplier. The lack of process in place on how and what to do for patients who needed transfusion also led to delays due to miscommunication. Nurses very often became the main point of contact for any form of communication and multiple phone calls were required to communicate between parties to get the blood ready for patient as soon as possible.

Methods: Delay in placing blood orders that lead to long wait time was mitigated by scheduling patients to come in one day prior of the scheduled transfusion for consultation and blood tests. This allowed patients to leave immediately after blood taking and consultation, and allowed clinician to decide if transfusion was required the next day. If not necessary, transfusion slots were cancelled, freeing up space for acute cases. Long wait time was also attributed by logistic delays and was mitigated by allowing clinicians to place advance order in the electronic system. This gives BTS ample time to match blood or to arrange antigen specific blood with supplier, if needed, immediately after GXM results were released. With this advance arrangement, logistic delays were avoided. A standardized workflow also ensures that practices were not deviated. It detailed the actions required by

each staff involved ultimately leading to better communication between different teams.

Results: We monitored for 6 months after implementation. It was found that patient's wait time was reduced from 3 h to 20 min. 50% of unused transfusion slots were cancelled appropriately allowing free slots for acute cases. Communication between BTS and clinic also improved with a significant reduction of phone calls of up to 80% was observed from an average of 10 calls for blood ordering to less than 2 calls daily.

Summary / Conclusions: Singapore is a small island nation with commute time of an hour from end to end thus making this workflow feasible despite patient require to visit hospital twice as wait time in the hospital can be longer than commute time from hospital to home. This initiative reduced patient's wait time, improved efficiency in transfusion slot booking as well as communication between clinic staff and BTS. The workflow also reduced BTS pressure to provide blood urgently as blood will be ready before patients come in for transfusion the next day.

P002 | Optimising occupational safety in blood services—the successful Australian experience of decreasing biological exposures and improving hepatitis B vaccination

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Background: From May 2018, Australian Red Cross Lifeblood (Lifeblood) began enhanced reporting of biological exposures in staff (including definite and possible parenteral exposures) with cumulative data from January 2018 onwards including incidents by division, hepatitis B immunity status, source outcome and contributing events. Early reporting documented donor-facing staff were at risk, with incomplete needle retraction contributing to the residual risk and facial exposure from splash incidents contributing to possible parenteral exposure. Procedural controls were recommended. Monitoring demonstrated plasma needles with a different retraction system were responsible for more parenteral exposures than whole blood. Despite work safety being paramount, there is little international surveillance data on biological exposures in blood services for benchmarking. We describe our national surveillance and successful strategies to reduce risk from biological exposures.

Aims: To use surveillance data of biological exposures to decrease the risk to staff to as low as reasonably achievable through a risk reduction program including engineering control changes, training and education and staff vaccination policy updates.

Methods: All biological exposures were evaluated from January 2018 until November 2023, with exposures from donation type (apheresis and whole blood) from September 2018. Contributing events were evaluated and focussed on. Rates were compared to the first 6 months of monitoring to the last 6 months of monitoring to demonstrate the impact of our program.

Results: The 6-month average definite parenteral biological exposures for the first 6 months of monitoring was 6.98 per 100,000 donations (48/687,622) and the last 6-month period was 2.21 per 100,000 donations (19/813,205), the relative risk reduction being 0.33 ($p < 0.000$, 95% CI 0.19-0.56). From September 2018 when donation type was monitored, there was initially a significant difference in needlestick incidents comparing apheresis 5.12 per 100,000 (20/383,672) to whole blood 0.89 per 100,000 (3/338,198), with the rate ratio being 5.88 times ($p < 0.001$, 96% CI 1.91-24.84). At the end of the evaluation period (6 months) there was no longer a significant difference with the apheresis rate being 1.42 per 100,000 (6/427,242) and whole blood being 2.33 per 100,000 (9/386,063) rate ratio 0.6 ($p = 0.37$, 95% CI 0.20-1.75). This is attributed to new sample and apheresis plasma needles implemented gradually nationally in August to October 2019 and August 2020- June 2021 respectively with improved safety enhancements, optimal phlebotomy set up and discard policies and training and multiple needlestick awareness campaigns. Hepatitis B vaccination for all new Category A staff was made mandatory in 2023, with active follow-up of all existing staff enabled by national vaccination status reporting and immunity status reporting.

Summary / Conclusions: Lifeblood has demonstrated a significant reduction in biological exposures through a comprehensive surveillance system and actions in response. Hepatitis B, because it has the highest prevalence in blood donors and transmissibility is higher compared to other blood-borne viruses, is the biggest risk of biological exposures. Fortunately, hepatitis B is vaccine preventable, and our vaccination policy has increased protection to decrease risk to staff.

P003 | Reorganization and informatization of transfusion service in Croatia experiences of regional centre for transfusion medicine, University Hospital Centre Osijek

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Background: Transfusion medicine is part of modern health care that covers a wide range of activities, from promotion of voluntary blood donation through chain of activities as collection, production, testing, storage and distribution of blood products up to bedside transfusion to patient. Reorganization is a set of measures motivated by weaknesses of the existing structure and requisite for improvement. The main problem of the transfusion service in Croatia was the excessive number of centers for transfusion medicine where blood products were produced, so equipment, finance and experts were used irrationally. At the same time, small centers could not meet the quality assurance and quality control standards for blood products. A major step forward in the reorganization of the transfusion service was made by the establishment of the legal basis (implementation of EU directives to Croatian legislation). The preconditions for the

reorganization of the transfusion service were realization of financial support, computerization of the transfusion service, reorganization of work in the transfusion services and the IT connection of the transfusion services with the national IT program.

Aims: Aim of reorganisation was to increase production uniformity and process standardization, sufficient supply and self-sufficiency of safe and high-quality blood products in region, cost reduction and connection to the national IT program which ultimately leads to increased quality and safety of transfusion treatment for patients.

Methods: Management started a systematic analysis of the current situation. All strengths and weaknesses of the system were considered and strategic and organizational plans were drawn up.

Results: In Croatia, with the adoption of the Basic Network of Transfusion Services for blood and blood products, the transfusion service was rationalized, and from the existing 32 transfusion units the following blood establishments were established: Croatian Institute of Transfusion Medicine, three regional and three subregional centers. All other transfusion units were reorganized for clinical transfusion activity. University Hospital Centre Osijek, Clinical institute for transfusion medicine (CITM) as regional center took over blood establishment activities from 6 General Hospitals. CITM took over the collection, production, testing, storage and distribution of blood products for seven health institutions in four counties (6000 km² and population cca 3 Croatia) and increase production from cca 10,000 to more than 30,000 collections per year. In order to do so, CITM reorganized the existing organizational structure, ensured a sufficient number of educated staffs, purchased new equipment, computerized the service with staff education and implemented robust quality system. At the same time, we gain more favourable purchase of higher quality consumables and achieve higher standards for produced blood component. connection with Informatisation and implementation of national IT program "e-Delphyn" (Hemasoft) enabled that sorting, searching and data processing are done by device which reduced possibility of mistake caused by human error.

Summary / Conclusions: In less than two and a half years, CITM, the University Hospital Centre Osijek, took over the tasks of collecting, testing, storing and distributing blood products for seven health institutions in four counties. By consolidating the businesses, the quality of the product was increased, costs were reduced and the safety of transfusion treatment was increased.

P004 | Abstract withdrawn

P005 | National register of bone marrow donors of Kazakhstan—search and activation of donors

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Background: Since 2012, the National Registry of Hematopoietic Stem Cell Donors has been established on the basis of the Scientific and Production Center of Transfusiology. One of the main functions of the Register is the timely processing of requests for the search of unrelated donors coming from transplant and search centers in Kazakhstan and Russia, the search and activation of a HLA-phenotype compatible HSC donor in the database.

Aims: Analysis of the work of the National Registry of Bone Marrow Donors in Kazakhstan.

Methods: A retrospective analysis of requests received in the Register for potential donors from clinics and centers for 2021-2022 was carried out. Statistical data was analysed in the Microsoft Excel.

Results: As of the end of 2023, the Register included more 10 900 donors. All donors were sampled at 5 loci at high resolution in accordance with the recommendations of the European Federation of Immunogenetics. The majority of those recruited (63.5%) are males. The largest proportion among donors were the age groups from 25 to 35 years (44%) and from 35 to 45 years (33%). There are more than 30 nationalities in the Register. The ethnic structure of the Register is polymorphic and corresponds to the population of Kazakhstan. During the period from 2012 to 2023, the Register received 216 requests from the National Scientific Oncology Center (NSOC), the National Scientific Center for Motherhood and Childhood (NSCMC), the Scientific Center for Pediatrics and Pediatric Surgery (SCPPS), oncohematology clinics of Russia. Over the period from 2021 to 2022, the number of requests increased from 35 to 43 per year. According to the results of 2021-2022, the majority of requests came from Russian clinics - 34.6% (27), SCPPS - 32.1% (25), NSCMC - 19.2% (15) and NSOC - 14.1% (11). These requests were related to the following nosologies for patients aged from "up to 1 year" to 61 years: acute lymphoblastic leukemia - 29.5% (23), aplastic anemia - 20.5% (16), acute myeloid leukemia - 15.4% (12), primary immunodeficiency - 12.8% (10), myelodysplastic syndrome - 7.7% (6), Hodgkin's lymphoma - 3.8% (3), chronic myeloid leukemia - 3.8% (3), chronic lymphocytic leukemia and multiple myeloma - 1.3% (1), including diagnoses were not established in 3.8% (3) cases. The patient and donor HLA typing matches ranged from 5/10 (50%) to 10/10 (100%). The majority of HSC donors were aged 26-30 years - 24% (18) and 31-35 years - 22.7% (17), the smallest number of donors from 41 to 45 years - 4% (3). Upon activation of the donor, confirmatory typing of the potential donor was performed at high resolution by NGS in the HLA-laboratory. 37.1% (13) of donors were activated in 2021, 41.9% (18) in 2022. According to the results of donor activation, 2 and 3 allogeneic bone marrow transplantations were successfully

performed in 2021 and 2022, respectively (matching HLA typing 9/10 and 10/10).

Summary / Conclusions: To sum up, despite the young age of existence, the National Register has active operation, including continuous work with oncohematological clinics. Since 2020, the Register has access to search and comparison of results in WMDA. In August 2022, the WMDA Board approved the Register's Membership, which allows the Register to list data on potential donors in an international database and use it in a secure mode.

P006 | Current situation of the blood service in the Republic of Kazakhstan

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Background: The Kazakhstan Blood Service is state-owned; blood centers are engaged in the procurement, processing and sale of donor blood and provide blood and its components to medical organizations within their region. Transfusiology departments in hospitals deal with clinical issues. Funding for blood service organizations is carried out from the state social health insurance fund, at uniform tariffs throughout the country, according to the actual number of blood components issued. In the republic, for many years, the consumption of blood components in medical organizations has remained at the same level; only the consumption structure is changing: the need for plasma is decreasing, the need for cells is growing.

Aims: Conducting an analysis of statistical indicators for the activities of the blood service of Kazakhstan for a 10-year period.

Methods: Monitoring of performance indicators of the blood service of the Republic of Kazakhstan for 2013-2023.

Results: In 2023, compared to 2013, the number of donations of blood and its components in the republic decreased by 14.4% and amounted to 248.7 thousand (284.6 thousand in 2013). The structure of donations has changed: the number of blood donations in the total volume of donations increased from 81.3% in 2013 to 90.5% in 2023, the number of plasma donations decreased from 15.7% to 0.4% (from 44.5 thousand to 1 thousand). Cell donations in the structure increased from 3.0% to 9.1% (from 8.6 thousand to 22.7 thousand). There is an increase in gratuitous donations in the republic: 97.0% in 2023 (85.3% in 2013). There is an increase in the volume of consumption of blood components transferred for clinical use by 20.5% compared to 2013. It is worth noting that the ratio of procurement and distribution has changed throughout the republic - with a negative increase in donations, there is a positive increase in the volume of distribution; the number of blood components issued to clinics from one donation was 1.7 in 2023 versus 1.2 in 2013. In Kazakhstan, the pathogen inactivation rate of platelets issued for use reached 100% in 2018 (versus 61% in 2013) and remains at this level. The leukofiltration rate of erythrocytes also amounted to 100% since 2018 (61.4% in 2013). In 2023, the share of plasma that underwent quarantine and pathogen inactivation was 99.4% compared to 70.4% in 2013. Since

2013, screening of donor blood has been carried out in two stages - immunological screening and NAT testing, carried out using fully automated closed-type analyzers, the use of which has reached 100%. Since 2022, new markers of hepatitis B virus have been added to the mandatory screening of donor blood - anti-HBcore and anti-HBs.

Summary / Conclusions: The main trends in the activities of the blood service of Kazakhstan over 10 years are the increase in indicators of free voluntary blood donation, changes in the structure of donations in favor of donations of whole blood and cells, an increase in the consumption of erythrocytes and platelets, an increase in indicators of infectious safety of transfusion therapy due to the introduction of additional markers of transfusion infections and an increase in volumes of application of technologies for pathogen inactivation of fresh frozen plasma.

P007 | Driving change leadership—lessons from SANBS' implementation of the blood establishment computer system

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Background: The South African National Blood Service (SANBS) plays a crucial role in ensuring a safe and adequate blood supply for patients in South Africa. To enhance operational efficiency and embrace new technology, SANBS initiated the implementation of the Blood Establishment Computer System (BECS), signifying a substantive departure from their existing system. This software, tailored for blood banks and collection sites, plays a vital role in ensuring the safety and traceability of blood products throughout the transfusion process. Recognizing the importance of effective change management, SANBS aimed to integrate BECS into its existing blood management procedures. Digitizing the entire donation value chain was a key objective to contribute to a safe and sufficient blood supply. The initiation of the BECS project occurred in 2020 and directly impacted 1563 individuals across various departments within SANBS, out of a total workforce of approximately 2729 employees.

Aims: The aim of this study was to describe the change management strategy followed in the implementation of BECS in SANBS.

Methods: The change management approach used in the study was the ADKAR model, which emphasizes Awareness, Desire, Knowledge, Ability, and Reinforcement. Strategies included introducing the mascot Betsy, conducting workshops, and internal communication to promote awareness of BECS. Moreover, the team trained super-users, adopted a train-the-trainer method, implemented performance assessment tests, and offered refresher training to encourage knowledge retention. The number of tasks logged with the Information and Communication Technology (ICT) division will be evaluated over 7-week post implementation of BECS. A thematic analysis was performed to determine the effectiveness of the change management strategy.

Results: The change management efforts enhanced data management and accessibility, increased operational efficiency, and resulted in positive user adoption of the new system. The BECS System

implementation on November 7, 2023, marked a significant achievement for SANBS. However, challenges surfaced, notably during the first two weeks post-launch, with 6797 tasks logged with the ICT division by January 24, 2024, highlighting areas needing improvement. Key themes included Access and Authentication Challenges, Network and Hardware Disruptions, and Processing and Inventory errors. SANBS's ICT department addressed these issues by upgrading bandwidth across 167 of 174 sites and configuring routers for about 105 sites. While many issues were resolved, some system-related defects persist, requiring ongoing attention. Post-go-live plans include optimizing Donation Testing algorithms, refining training content, and scheduling refresher training for donor staff. Efforts are ongoing to stabilize the system and address visibility issues with Donor Recruitment. A post-implementation assessment in mid-April 2024 will evaluate adaptation and utilization rates, integral to assessing BECS implementation success.

Summary / Conclusions: In summary, even with favorable results, unforeseen difficulties surfaced throughout execution. User preparedness was not appropriately assessed by the competency assessments or the readiness assessment. The upcoming post-implementation assessment will highlight the vital role that change management plays in boosting user adoption and guaranteeing successful system integration, and it will offer insights for ongoing improvement.

P008 | Establishing the first HLA laboratory in Republika Srpska—a landmark for regional transfusion medicine and patient care

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Background: The absence of a dedicated human leukocyte antigen (HLA) laboratory in the Republika Srpska entity of Bosnia and Herzegovina has significantly impacted the local healthcare system, especially for hematological and oncological patients in need of hematopoietic stem cell (HSC) transplants. Historically, these patients have had no alternative except to seek diagnostics and treatment abroad, incurring substantial costs and facing delays in receiving critical care. The initiative by the Institute for Transfusion Medicine of Republika Srpska to establish the first regional HLA laboratory is a significant advancement, building on our previous successes in molecular diagnostics, notably in molecular immunohematology (blood group genotyping). **Aims:** This study outlines the journey and strategic planning involved in founding the region's first HLA laboratory, emphasizing its role in enhancing management and overall organization at the Institute for Transfusion Medicine. Focusing on the HSC donor registry and providing diagnostic support for cadaveric transplantation, the laboratory aims to transform local patient care. The highlighted anticipated regional

healthcare improvements minimize the need to travel abroad, hence enhancing accessibility, lowering costs, and improving transplant outcomes.

Methods: Establishing the HLA laboratory involves a comprehensive approach, starting with sophisticated equipment procurement. A major emphasis is placed on team education and training for technical operations and assay results evaluations to ensure high standards of diagnostic accuracy. In addition, we are developing an extensive HSC donor registry, adopting best practices through collaboration and knowledge exchange with established registries. The laboratory will implement diverse well-established methods, such as sequence-specific oligonucleotide (SSO) typing for donor registry and rapid qPCR typing for emergency cadaveric transplantation. A key component of our laboratory's innovative approach is the adoption of next-generation sequencing (NGS) based on nanopore sequencing for fast donor-recipient matching. This novel application in our regional context surpasses traditional assays for high-resolution HLA matching of donors and recipients in both speed and accuracy. Additionally, advanced HLA antibody testing by solid-phase assays, crossmatching and HSC chimerism evaluation will ensure thorough transplant compatibility assessments.

Results: Completed phases include the furnishing of the required laboratory space as well as instrumentation procurement and team recruitment, all aligned with the highest applicable international standards for HLA testing. Upcoming phases entail comprehensive staff training, now already under discussion and agreement, to ensure operational proficiency and accuracy in HLA analysis. This initial work lays the groundwork to provide critical services that simplify the local transplant process.

Summary / Conclusions: Our initiative for the first local HLA laboratory marks a milestone in transfusion medicine and patient care in Bosnia and Herzegovina. Through advanced molecular diagnostics and regional collaboration encouragements, the Institute aims to significantly impact patient outcomes and healthcare efficiency and contribute to global transplant medicine and immunogenetics knowledge.

P009 | Technical direction of transfusion

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Background: Catalonia with a population of 7.6 million (17% of the Spanish population) is served by the Banc de Sang i Teixits as its unique Blood Establishment. Our mission is to issue blood components to whatever hospital in our area where, by law, they must have in charge a haematologist as its hospital blood bank's responsible. Due to the lack of this professional profile, our organization offers the possibility, through an agreement, to cover this need. We currently provide the technical direction to more than fifty public or private hospitals throughout Catalonia that could need blood components for transfusion.

Aims: Review the work done by these technical directions in the last four years to standardize transfusion practices in all our hospitals. The main goal is to ensure safe, quality, and efficient blood components use, creating a transfusion network under the supervision of our organization.

Methods: Our organization conducted a risk analysis in all those hospital blood banks managed by this technical agreement in our countryside. Procedures were drafted to define their processes and the working method, determining the minimum frequency of follow-up visits for each hospital and allocating responsibilities among the parties. Each hospital blood bank has a haematologist assigned by our organization to oversee their daily work. We established a skilled nursing team that collaborates to conduct systematic follow-up visits to these centres using a checklist to supervise their activities.

Results: After 4 years, this management model is well-established, currently employing a team of five full-time dedicated nurses that cover 53 technical directions. Follow-up visits by this staff provide a real-time view of transfusion activities, the training of professionals in each centre and quality controls for equipment and critical materials. Corrective and preventive measures have been implemented, leading to increased reporting of incidents to the hemovigilance system. Standard operating procedures have been written, reviewed, and updated by each hospital, adding to consistent safety and quality guidelines, which are integrated into each centre's quality management system. Annual training plans ensure the continuous education and development of nursing professionals and laboratory staff involved in the transfusion process. The first training day for hospital blood banks in 2023, brought together over 70 participants from different centres covered by these technical directions from our organization. Beyond

updating knowledge in transfusion medicine and promoting transfusion safety, the event encouraged active participation and the exchange of ideas and experiences among participants.

Summary / Conclusions: Our technical direction model facilitates the standardization of different hospital blood banks while maintaining their individual identities and addressing their specific needs. The exclusive dedication of nursing professionals specialized in transfusion to the oversight of these duties provides a comprehensive view of these centres, strength in their unification, ensuring consistent safety and quality in the transfusions done in Catalonia under our transfusion network. Internal team and cross-departmental meetings with related Banc de Sang i Teixits departments are crucial for maintaining homogeneity.

P010 | Organisational impact of a middleware for managing the hub and spoke network of immunohaematology laboratories of the transfusion medicine department of Venice district

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Background: The Transfusion Medicine Department of "dell'Angelo" General Hospital operates in a large metropolitan hospital which acts as a hub for a spoke network made up of seven acute care hospitals, acting as a reference for the entire district of Venice, which has approximately 900,000 inhabitants. All second level immunohaematology is centralized in our laboratory, we have three different platforms for serological diagnoses; tube test, gel test and solid phase tests and a micro-arrays-based platform for genotyping.

Aims: we present our experience in the evaluation of the Blood Typing Manager middleware proposed by Grifols in the departmental management of immunohaematological diagnostics

Methods: The study was performed to evaluate the Blood Typing Manager in an operational setting, characterized by routine immunohaematological diagnostics with medium-high volumes of activity, possibility of access to / alternative instruments, presenting complex diagnostics. In this study, the management of analytical sessions with the possibility of importing work lists and exporting results, the ability to manage different instruments and integrate the results obtained, the self-validation and result blocking algorithms were evaluated first. Subsequently, we evaluated the software to assist in antibody identification, the management of reflex tests, and the interface with the IT management system. Particular attention was then dedicated to the management of both internal (IQC) and external quality controls (EQA).

Results: We believe that in a multi-site Transfusion Medicine characterized by a hub and spoke organisational model, the use of a middleware for the management of immunohaematological testing is essential for a series of reasons. Firstly, in the hub lab, to symmetrically distribute the analytical load on different analyzers to optimize activities and response time. A further relevant fact has been demonstrated: the possibility of setting customizable rules for the management of re-runs, reflex tests, second level tests, which can also be carried out with different methods (for example the selection of

samples to be subjected to genotyping). Rules which, once set, will be used automatically in the management of the analytical routine. Another relevant matter is the possibility to import samples and requests from peripheral labs and to export results. This also involves the possibility of moving the entire routine from one lab to another in the event of an instrumental malfunction that cannot be resolved in a short time. In our organization, characterized by centralization of second level diagnostics, a relevant question is about operators skill's maintaining, an objective that can be achieved using the Blood Typing Manager which allows an exchange of expertise, by sharing the tests performed and the related results, including images, allowing remote interactions on both clinical cases and EQA. We particularly appreciated the presence of customizable validation algorithms. For example auto validation of analytical sessions on the basis of results obtained processing IQC or samples self validation after evaluation of congruity with available historical results.

Summary / Conclusions: We therefore believe that the adoption of a middleware as Blood Typing Manager, that allows the management of immunohaematological tests at a large area level is an indispensable premise in the departmental organization.

P011 | Founding of permanent blood collection branches based on donor centric approaches

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Background: Blood transfusions are integral parts of both emergency and elective healthcare systems without supportive potential of which the modern multidisciplinary areas would be nonsense. The precise functionality of the blood industry is a crucial objective of Blood Service facilities. This involves constant monitoring of blood supply and replenishment of blood products, ensuring ongoing blood safety interventions. It is of utmost importance to have a comprehensive understanding of blood donor drives, as they are solely carriers of invaluable biotherapeutic resource. The blood collection process in Kazakhstan is made at fixed sites and mobile sites. Fixed sites represent blood centers collecting both whole blood and apheresis platelets separately or in combination with concurrent plasma while mobile sites are scheduled for whole blood collection only. The supply of donor blood and the replenishment of blood products are influenced by factors that stay far beyond the motivational behavior of donors. They also depend on the population density in each region of the country. Because the distance to blood donation sites and the diversity of available transportation infrastructure are the primary factors directly affecting donor attendance. That's why implementing donor-centric approaches by establishing networks of permanent blood collection sites would resolve donor blood deficiency issues.

Aims: To prove the founding of permanent blood collection branches based on donor centric approaches.

Methods: The study includes: Analysis of blood donation statistics observed during the last three years in Astana. Review of donor data, including residential addresses, to map them with the blood center location. Identification of urban bus lines passing through stops nearby the blood center. One-day survey of donors regarding the conditions of their attendance at blood donation sites.

Results: The blood collection statistics from the Astana Blood Center over the last three years have not shown significant trends, except for a slight increase in mobile session settings. Mapping the residential points of donors reveals that the vast majority of them reside at a large distance from the blood center. Moreover, there are limitations in the public transportation system, which includes five bus lines stopping nearby. The situation has aggravated by the fact that these lines do not cover approximately 20% of the city's donor-most-populous area. During one of the routine blood collection days, 129 donors were surveyed with their informed consent where 67% of them were male, and 64% were regular. Almost all donors are habitants of Astana, the majority of whom donate blood at a blood center moving to there in their own vehicles and finding it as quite convenience way while 1/3 of them use public bus transport. Interestingly, that 52% of respondents wants small blood collection networks to be close to the donor-most-populous areas.

Summary / Conclusions: The forecast of blood donation trends for 15 coming years in Astana makes the establishment of permanent blood collection branches inevitable, which indeed may become a new reality in resolving donation issues based on the donor-centrism concept.

P012 | Blood banking system in Bosnia and Herzegovina

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Background: The blood bank service is vital in the health care system. According to the WHO, developing national blood policies in one country is considered one of the main achievements. According to European Council Directive No. 2002/98/EC, a competent regulatory body that regularly controls the quality system and issues licenses for transfusion services is required. Bosnia and Herzegovina has experienced essential changes in the past thirty years. The move from centralized to market-based economies affected the health system. In Bosnia and Herzegovina, there is no national blood policy. The federal government, with ten cantonal ministers of health and one federal, has no strength to deliver a national policy.

Aims: To evaluate the current status of blood banking in Bosnia and Herzegovina, according to the last report of the EU Commission for Bosnia and Herzegovina. To highlight weaknesses and threats, but also obligations and opportunities of the country on the way to the European Union.

Methods: The different data sources were used in the retrospective analysis: the internal reports from blood banks, the reports from Ministries of Health, the data from the Agency for Statistics of Bosnia and

Herzegovina, and the analytical report from the EU Commission 2019 and 2022 for Bosnia and Herzegovina.

Results: Bosnia and Herzegovina is a relatively small economy with a population of approximately 3.5 million, living on a territory of around 51,000 km². The blood donation system in Bosnia and Herzegovina is voluntary based on 100%, particularly regular voluntary donation, which is self-sufficient in one manner. During 2022 ca 100.000 blood units were collected. Thus, blood supply is partly based on replacement/family donation (around 70%–75%) and cannot meet WHO recommendations. The crucial message from the EU Commission in Bosnia and Herzegovina was: "Bosnia and Herzegovina is at an early stage of preparation in consumer protection and public health. No progress was made in the area during the reporting period; last year's recommendations were not implemented and remain valid. In the coming year, Bosnia and Herzegovina should, in particular, further align its legislation with the EU acquis on substances of human origin and on medicines for human use and establish an oversight system in this field to ensure efficient coordination in the whole country".

Summary / Conclusions: The main challenge is that there is no national mandate for health care financing and provision. Consequently, there is no blood banking strategy on the national level, and there is no Ministry of Health on the country level. In Bosnia and Herzegovina, there are two different models of the organization of blood service: a centralized one in the Entity of Republika Srpska and a decentralized one in the Federation, with one independent hospital-based transfusion department in the District of Brčko. Considering the current situation and working conditions, it can be said that the present system of transfusion services in Bosnia and Herzegovina is inadequate and unreasonable. The country has respectable potential: excellent healthcare professionals, a long tradition of transfusion medicine and voluntary blood donation, and solid infrastructure. The future progress in the field of blood banking is uncertain and depends exclusively on the government's commitment to achieving the highest standard in transfusion medicine.

P013 | Abstract withdrawn

P014 | Building resilience—navigating the volatility, uncertainty, complexity, and ambiguity (VUCA) environment in low-medium countries blood services

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Background: The VUCA describes the features of a rapidly changing and unpredictable business environment. The VUCA environment have a significant impact to all sectors of the economy in Zimbabwe and a not-for-profit healthcare entity like the National Blood Service Zimbabwe (NBSZ) bear the consequences of the economic meltdown. VUCA de-stabilize operating environment leaving both employees

and organisations feeling overwhelmed. Addressing the challenges posed by the VUCA environment demands substantial investments, time and resources to navigate its complexities effectively.

Aims: To share how NBSZ has responded to the VUCA environment for sustainability.

Methods: NBSZ identified the VUCA environment apt responses that included robust risk management. The process involves conducting regular risk assessments, reviewing of Strategic plan, developing contingency plans, and establishment crisis management protocol on a quarterly basis. The established strategic partnerships with schools, profit making entities, faith based organisations and pledge 25 club improves blood collections and revenue lines. Furthermore, NBSZ fostered strategic partnership through participation in competitive grant writing and establishment of the cross-functional project teams, fostering a culture of innovation and continuous learning. The NBSZ uses a commercial blood management system to monitor blood product inventory levels, expiries, and demand patterns.

Results: NBSZ managed to proactively identified potential risks, their impact, and contingency plans to mitigate them. NBSZ regularly forecast demand and assess the long-term viability of providing blood products and services. NBSZ managed to meet 60-80% of the demand in the past three years. Several strategic partnership had been instituted that leverage complementary strengths, shared resources, and access to new markets. Strong flexible leadership helped NBSZ to be resilient and adaptable to the VUCA conditions. The right technologies enabled NBSZ to enhance operational efficiency ensuring a reliable supply of blood products. Strategic partnerships provided access to additional resources. Continuous learning and development enables employees to stay updated with emerging trends by participating at international and regional conferences. The key performance indicators were largely satisfactory averaging above 70% on HPMS, Corporate strategic plan, BSMS and risk tracker. A staff incentive programme was established to relieve employees working under excessive pressure, stress and anxiety associated with working under the VUCA environment.

Summary / Conclusions: Despite the VUCA environment in Zimbabwe, the resilience by NBSZ provides evidence that the mitigation measures identified and implemented were effective. It is important for blood services to assess their specific context and challenges to identify appropriate strategies that align with their goals and values. For sustainability, it is recommended that blood services focus on building resilience, fostering a culture of continuous learning and adaptability, investing in technological capabilities, and developing strong networks and partnerships.

P015 | Abstract withdrawn

P016 | Abstract withdrawn

Management and organisation— information technology

P017 | ISBT 128 goes 2D

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Background: Traceability is of outmost importance when handling Substances of Human Origin (SoHO), both between donor and patient, but also to equipment and consumables. To facilitate collection of data during the processes from donation to transfusion or transplantation, bar codes have been widely used, first using the American Blood Commission's ABC Codabar. The ABC Codabar was not maintained and disintegrated. The Codabar symbology had many misreads compared to newer symbologies. ABC Codabar and other national systems were replaced by the ISBT 128 standard developed by ISBT's Working Party on Automation and Data Processing (WPADP, now WPIT). To secure development and maintenance ISBT 128 the International Council for Commonality in Blood Bank Automation (ICCBBA) was established. The name ISBT 128 is derived from the symbology used (Code 128).

Aims: To reduce the number of scans 2D symbology (Data Matrix) recommended by the ISBT 128 standard was implemented on labels of all handled blood components, cells and tissues.

Methods: In 2006 ICCBBA began investigating which 2D symbology was optimal for labeling of SoHOs. The choice fell on Data Matrix. After profound discussions in ICCBBA's Technical Advisory Groups and Standard Committee, the first version of ICCBBA's implementation guide *Use of Data Matrix Symbols with ISBT 128* was published in 2011. In Scandinavia, there has been a consensus regarding labeling and use of flags in the Donation Identification Number (DIN) for process control. A group of customers was established by the major blood bank computer system (BECS) vendor to accommodate the labeling with Data Matrix symbology with the consensus standard. The group comprising members from all Scandinavian countries worked from 2018-20 where after the BECS provider reprogrammed the blood component handling module of the software and delivered it for testing and validation in 2023.

Results: The new software version was implemented January 2024. As the ISBT 128 uniquely identify a SoHO unit within a century by combining the DIN and the product code, two 1D (Code 128) barcodes always has to be scanned when handling a unit. An example of the string of characters in a Data Matrix symbol on a Red Blood Cell unit:

= +06000 = V00422440841642
= %9500 ≤ E3846V00& × 0240340000& > 0240692359
= \930330005500002399

which codes for a compound message with the donation number, the blood group, the product code, the collection date & time, the expiry date & time and special testing (phenotypes). Some examples of the reduction in number of scans:

P017 Table 1

Procedure—number of scans	Code 128	Data Matrix
Release, moving blood component	2	1
Buying/selling from /to other blood center	6	1
Platelets from 4 buffy-coats	16	8
Electronic Identification of patient, blood component and staff bed-side	3	2

Summary / Conclusions: Labelling with Data Matrix was successful implemented in the blood center, the tissue bank and all hospitals in a region with 1.2 million inhabitants. This labelling will during the next couple of years be disseminated to most of Scandinavia. 2D symbology halves the number of scans and since hundreds of thousands of scans are performed each year, it will have an impact on effectivity and performance. Furthermore, it secures that all relevant information (data structures) are scanned from one and only one unit, thus reducing the risk for mix-ups. The changes in the BECS means, that should change from 2D symbology to RFID-tags be relevant, you only have to change the scanner.

P018 | Implementation of the radio frequency identification system for packed red blood cell units in a hospital based blood centre—process and quality parameters evolved

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Background: Bedside administration of blood components still remains the weakest link in the transfusion chain even today. We implemented the RFID system from Biolog-ID for RBC units in a phased manner with the intent to enhance inventory management, limit storage related hemolysis and improve blood administration process in recipients.

Aims: Implementation and validation of the RFID system and its objective evaluation based on measurable quality parameters.

Methods: The Biolog-ID RFID system was implemented along with exhaustive training of the blood centre and nursing staff for the use of the patient safety device as well as the software system. Total 2000 PRBC units were tagged with RFID label. The time taken for bedside checking prior to transfusion, expiry of PRBC units in inventory and the percentage of hemolysis in the PRBC units as measured with a Plasma low hemoglobinometer from Hemocue were considered as objective parameters for the analysis of effectiveness of this system. Data related to implementation of the process was analysed separately for various clinical areas. Any adverse incident related to the new system was noted.

Results: Initial validation of the system in the pilot phase was completed using 150 RFID tags. 14 patient safety devices were handed over to the team leaders in clinical areas after successful completion of training by authorized company personnel. Average time required for pre-transfusion bedside verification process reduced from 15 to

5 min after implementation of the RFID system. Expiry of PRBC units was prevented due to alerts provided by the RFID system. Extent of hemolysis in PRBC units stored for various time periods in refrigerated storage reduced from a mean of 0.39% to 0.22%. This difference was found to be statistically significant ($p < 0.05$). One error related to wrong blood transfusion could be prevented with this system. Three incidents of leakage of PRBC units stored in the RFID cabinets due to limited space between the storage racks were noted.

Summary / Conclusions: The RFID system could be implemented successfully at our blood centre. This has significantly reduced the time required for bedside identification and documentation before transfusion. The blood centre has also been benefitted by virtue of prevention of undue expiry of PRBC units and maintenance of better quality and minimal hemolysis during their storage.

P019 | Development of an interactive PBM dashboard for monitoring implementation of strategies among the stakeholders in a tertiary hospital

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Background: Patient Blood Management (PBM) programs contribute to decrease the number of transfusions and improve patient outcomes. However, hospital implementation is many times challenging due to the complexity of healthcare data and the lack of streamlined analytical tools, which can hinder the assessment of the current state of PBM practices. Traditional manual methods for data analysis are time-consuming and prone to errors, making it difficult for the physicians to identify trends, assess performance metrics, and implement timely corrective actions to improve their processes. The absence of automated data analysis tools compromises the ability of healthcare providers to proactively address issues and optimize PBM practices. Without comprehensive insights into transfusion practices and blood component utilization, healthcare institutions may struggle to identify areas for improvement, leading to suboptimal patient care outcomes and inefficient resource allocation.

Aims: This project aims to introduce a robust dashboard tool designed specifically for PBM. The primary objectives include facilitating data-driven decision-making, enhancing operational efficiency, and improving overall patient outcomes through informed resource allocation and strategic planning.

Methods: The development of the dashboard involved a multidisciplinary approach, leveraging expertise in data analytics, healthcare management, and process optimization. Extensive consultations with stakeholders and end-users were conducted to ensure the tool's relevance and usability in real-world healthcare settings. The dashboard

was created using PowerBI, a powerful business intelligence tool used by many organizations across a variety of sectors. It integrates diverse data sources, including laboratory results and transfusion and pharmaceutical data to provide a comprehensive overview of patient care operations. Hospital information services supplied the necessary data for conducting the study and developing the dashboard.

Results: The dashboard system offers a user-friendly interface with customizable dashboards and intuitive data visualization tools. It consists of four principal tabs focused on different aspects of PBM objectives: Prevention of perioperative anemia. Minimization of blood loss. Optimization of blood component transfusion and transfusion safety. Impact on perioperative morbidity and mortality. Additionally, the system includes two summary tabs: one presenting the focal key performance indicators for each goal and another containing the metrics detailed in the annual PBM results report from the Transfusion Service. The tool allows filtering by date and surgical procedure, enabling an effortless provision of results corresponding to each specialty and the assessment of annual trends. This structure facilitates convenient and early access to relevant information for stakeholders and specialists. Early feedback from pilot implementations has been promising, highlighting the tool's potential to identify current practice gaps and orchestrate improvement plans for the different stakeholders.

Summary / Conclusions: The development of this data analytics tool represents a significant step forward in Patient Blood Management strategies implementation in our center. By providing specialists with timely access to actionable insights derived from comprehensive data analysis, the dashboard empowers the Transfusion Service to optimize resource allocation, improve operational efficiency, and ultimately enhance patient care delivery.

P020 | Correct matching process donor/blood unit/test tube—5 years of experience at Brescia Blood Bank (BBB)

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Background: Transfusion safety involves the implementation of an operational flow to transfuse the right blood component to the right patient. In 2018, following Regional indications, the BBB proceeded to implement the blood collection process through the use of Radio Frequency Identification technology (Rfid), achieving fully computerized control between donor identification and correct matching with donation/ test tube. The following is our experience of 5 years of operation.

Aims: To have a complete automation in donation-transfusion traceability and an automation in the process of production and handling of blood components.

Methods: Certification pathway: 1- Labeling the units (Toshiba printer provided by Fresenius Kabi) with Rfid tags (Ceracarta labels provided by Fresenius Kabi.) in which a chip and antenna are integrated to enable radio frequency data storage and transmission. Each Rfid is identified with a unique Unified Information Devices (UID) code. 2- Automatic labeling of all tubes required for biological qualification examinations (HEN Mini labeler Menarini). 3- Automatic reading of the 4 Rfid affixed to the 4 units of the collection set by the scales (Vasini Mod E051P.B.TC.RF.ID). Presence of a bidirectional information flow to ensure control of outgoing and incoming data between BBB computer system/scales software. 4- Correlation with donor identification data, donation initiation, and reading of validation tube codes, 5- Confirmation of tube barcode reading by Blood Group (BG) at the end of the donation with recording in the scales software and sending all donation data (operator and collection time) to our LIS management system. Current applications: 1- Loading of incoming units to the BBB via gate (Emopath gate 03 Infosolution), 2- Unique determination of BG by the use of the correctly matched tube to the unit (Neolris Immucor). Future applications: 1- Matching of blood component type and Rfid with R/W on fractionators (CompoMat G5 Plus with integrated antenna by Fresenius Kabi). 2-Outgoing unit handling from BBB versus collection centers of Brescia departement (CCBD), 3- Discharge plasma to the plasma-derived industry 4- Checking unit assigned to the patient's bedside.

Results: In 5 years, about 180,000/300,000 (60%) units collected in the BBB were managed with this technology. For 120,000 units the management was traditional (barcode loading and blood group control from unit segment). Of the units managed with Rfid technology, 0.1% failed the Rfid read step at point 3- of the Certification pathway, 100% passed reading the Rfid tag at the gate in acceptance at BBB, while 1.5% failed the certification path hesitating in blood group control from unit segment.

Summary / Conclusions: The implementation of this technology allowed us to handle 60% of the units received in a fully computerized manner. The BG tube correctly matched to the unit has made the process more smooth and automatic. The future goal is to be able to complete the entire operational flow by allowing us to automate the traceability of the donation and transfusion processes.

P021 | The knowledge portal—a tool that incorporates the capabilities of big data and artificial intelligence into management

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Background: The digital transformation of processes generates a large amount of data that increases the complexity of efficient management and exploitation by the information systems currently available in

transfusion centers. On the other hand, there are currently new tools -Big Data (BD) and Artificial Intelligence (AI)-, which can contribute to fulfil current needs.

Aims: To design a new tool that responds to the requirements and new challenges derived from this situation in our transfusion center.

Methods: The use of No-SQL databases oriented to Big-Data allow managing large volumes of information and more complex data models. For algorithms integration as third-party services, the predictive models MSTL (Multiple Seasonal-Trend decomposition), ARIMA (Autoregressive Integrated Moving Average), CROSTRON (algorithm proposed by Costron, J.D., forecasting and stock control for intermittent demands) and LSTM (Long Short-Term Memory neural network) have been included in a first phase. The development of the new tool was carried out in the following phases: Definition of requirements and functionalities. Web solution accessible from any workstation and capable of handling large volumes of information. Management of access rights that protects access to personal data and guarantees the traceability of who accesses the available information. Interoperable and able to manage federated data from different sources, after homogenizing them without the need to dump them into a single system. Dynamic and simple analysis of data without the need to know query or programming languages. Attractive visualization of analysis results in tables, graphs, or geo-referenced maps. It should allow the development and implementation of artificial intelligence algorithms (temporal series, pattern search, neural networks, ...).

1. Working sessions with the managers from different areas.
2. Tool development and parametrization.
3. Validation in a test environment.
4. Go-live
5. Evaluation of results
6. Scaling up to more working areas and process.

Results: From September 2023 to January 2024, processes in the areas of blood donation, apheresis, fractionation and distribution of components have been addressed with this new tool. As a result, a "Knowledge Portal" has been deployed. By accessing it, users can instantly obtain all the information related to the different processes and visualize them in tables, graphs and explanatory maps.

Summary / Conclusions: Beyond the significant savings in time and human resources, this novel and scalable "Knowledge Portal" adds great power and agility in the exploitation of the available information, generating, in a fast, effective and efficient way, very useful knowledge for processes management and decision making of our organization, meeting the requirements and expected functionalities.

P022 | Digital transformation of blood transfusion management systemY Huang¹, W Su¹, C Chung¹, L Hsing¹¹Medical Laboratory, Pingtung christian hospital, Pingtung, Taiwan, Republic of China

Background: It has been said, "To err is human." However, in the blood transfusion operation process, it was intolerable to make any mistakes. Every step during the blood transfusion process should be precise and free of errors. Therefore, preventing human error or any mistakes is critical for the improvement of blood transfusion safety. In this project, we identified possible defects during the blood transfusion process and also tried to simplify the end-to-end steps during the whole blood transfusion workflow. A total solution for an electronic blood transfusion management system was established to improve patient safety.

Aims: The aim of this project is to develop a complete solution for an electronic blood transfusion management system.

Methods: To avoid mistakes or near miss events during the blood transfusion process, it is critical to monitor all steps and procedures, including patient identification, blood drawing, specimen transportation, sample receiving, laboratory testing, blood cross-matching, issue blood, patient blood transfusion, and transfusion feedback reporting. Our quality improvement team thoroughly discussed all essential steps and identified probable failures or near miss events. Three major actions were implemented throughout the whole project. First, a novel barcode system was implemented to not only identify patients during the blood-drawing process, but also to retain information that is directly linked to the LIS system, such as patient medical data and laboratory test requests. Second, a real-time dynamic laboratory system was utilized to track the progress of the blood transfusion process, from physician ordering to blood transfusion feedback reporting. Third, the paper-based blood transfusion reporting was replaced with a computerized blood transfusion feedback report system which patient's vital sign data was directly uploaded into cloud LIS system.

Results: The following are the major advantages of utilizing an electronic blood transfusion management system: (1) the fewer phone calls between blood bank laboratory staff and clinical care nurses (approximately 5-10 calls and 15-30 min); (2) the percentage of emergency blood issued within 30 min was achieved at 99.15% in 2023; (3) the near-miss event of blood issued is reduced to 0% after barcode recheck and approval mechanism; (4) time savings from manual paper-based feedback reporting to patient identification and computerization electronic blood transfusion feedback reporting (reduced from 120 min to 0 min); (5) reduction in the amount of paper used, including blood transfusion request forms and blood transfusion paper feedback reports (reduced from more than 28,021 sheets of A4 paper from January to December 2023).

Summary / Conclusions: After the implementation of the total solution electronic blood transfusion management system, there have been no more near misses of specimen collection or blood issue errors. The advantage of implementing this digital blood bank system

is that it can timely monitoring of the entire transfusion process and also reduce the work stress of both blood bank staff and bedside clinical nurses. Moreover, the whole improvement project also eliminated paper use. It was concluded that the utilization of electronic blood transfusion management systems can prevent human mistakes and near-miss events and, most importantly, guarantee blood transfusion safety for patients.

P023 | New roster planning tool in enhancing efficiency for roster planning at blood collection sitesK Wong¹¹Blood Services Group, Health Sciences Authority Singapore, Singapore, Singapore

Background: The Blood Services Group at the Health Sciences Authority (BSG, HSA) used to create staff rosters twice a month. This involved ensuring sufficient coverage for all blood bank stations, fair duty rotation, and managing staff leave and time-off. Each roster for a collection site took approximately 1.5 h to prepare.

Aims: BSG, HSA aims to utilise advanced Excel functions to develop a roster planning tool to facilitate blood bank roster preparation.

Methods: The project team collaborated closely with managers and supervisors from various collection sites to gather requirements before developing the Excel-based tool. The tool was meticulously designed to automate tasks, replacing manual processes and enhancing the efficiency of roster planning. In late 2023, the roster planning tool was trialed at three collection sites namely Bloodbank@Woodlands (BB@WDL), Bloodbank@Dhoby Ghaut (BB@DG), and Bloodbank@One Punggol (BB@OP).

Results: The roster tool's effectiveness was evaluated through a poll among roster planners, comparing the time needed to create a roster using the new tool versus the previous manual process. Planners also rated the tool's features and ease of use on a scale of 1 to 5, with 1 being the lowest and 5 being the highest. The poll feedback is compiled in the table below.

Summary / Conclusions: The poll feedback revealed a significant 25%–50% reduction in roster planning time, freeing up time for staff to focus more on donors. Additionally, the tool received positive ratings for its useful features, which were unavailable in the previous

P023 Table 1

Survey	BB@WDL	BB@DG	BB@OP
Time taken to make roster using new tool	15 min	30 min	45 min
Time taken to make roster using manual process	30 min	45 min	1 h
Percentage of time saved	50%	33%	25%
*Rating for usefulness	5	4	3
*Rating for user friendliness	5	4	3

* Scale of 1 to 5, with 1 being the lowest and 5 being the highest.

manual planning method, and for being easy to use. Continuous monitoring and analysis of time saved, as well as feedback given by roster planners, are important in ensuring the long-term sustainability and effectiveness of the new Roster Planning Tool. BSG, HSA would continue to gather feedback from users periodically and enhance the tool as required.

P024 | Do operators in the processing lab recognize the value of IT connectivity?

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Background: The Blood and Tissues Bank of Aragón (BTBA) gradually created an ecosystem of devices interconnected and managed by software, increasing the level of automation and introducing the use of automated decision-making in its blood processing laboratory. Some operators were present since the beginning of this journey and experienced different levels of data connectivity.

Aims: To evaluate the level of satisfaction of operators in the processing lab with increasing levels of automation and software decision-making procedures.

Methods: Blood is processed with the Reveos[®] Automated Blood Processing System. TOMEs (Terumo Operational Medical Equipment Software) support, monitor and documents procedure data through its bi-connectivity to the blood bank information system or BBIS (eDelphyn), and guide the selection of Interim Platelet Units (IPUs) to compose a pool of 4 to 5 IPUs via T-Pool Select (a TOMEs application). TOMEs then control the pooling process in the TSCD[®] II stations (Terumo Sterile Connection Device) and document the treatment of pools with the Mirasol[®] PRT system. Information about operators, lot numbers, PRT treatments, and pool compositions are transferred from TOMEs to the BBIS. Further, BTSA implemented the Lhema software which relies on a production algorithm that computes the optimal number of PCs to be manufactured, and the optimal number of WB donations to be fractionated by Reveos following two different protocols. A survey was conducted where operators ($n = 18$) were asked to give grades from 1 (bad) to 5 (excellent) for the improvements provided by the new technologies.

Results: Fourteen full-time equivalent and four half-time equivalent (FTE) employees operate in the processing lab. Seventeen operators (94.4%) have completed the survey; seven of them (38.8%) were present from the start of full automation in 2013. None of these employees would like to return to the semi-automated system. Overall, operators are very satisfied with the Reveos system (4.7/5) and with the T-pool-select software (4.6/5). They consider Reveos a useful tool (5/5), intuitive, and easy to use (5/5), which decreases the workload (4.4/5) and reduces the processing time (4.7/5) as compared to semi-automation. Operators consider the T-Pool Select application useful (5/5), intuitive and easy to handle (4.7/5), decreasing workload

(5/5) and reducing processing time (5/5). They also acknowledge that it minimizes interpersonal variations (4.3/5) and errors (4/5). Twelve operators have worked without TOMEs in the past and only one (8.3%) would work again without this software. The satisfaction level of operators ($n = 16$) with TOMEs is 4.6/5. They value its features, such as traceability, equipment control (5/5), and reduction of errors (5/5). They recognize its usefulness (4.7/5) and ease of use (4.9/5). Operators graded its capacity to reduce workload by reducing manual data input with 4/5. Concerning the Lhema software, operators ($n = 17$) consider it a useful technology and easy to use with 4.3/5 and 4.6/5, respectively.

Summary / Conclusions: In conclusion, operators consider that automation, increased software-based process control and electronic data transfer are very useful in their daily work. They appreciated mostly the improvement in ergonomics and safety of their working place, followed by the error-reducing feature of the new ecosystem.

P025 | Practical implementation of a donor survey computer application

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Background: We describe the development of a computer application to conduct surveys among our population of altruistic blood donors.

Aims: To analyse the impact of a new computer application (APP) to capitalize data from surveys taken from donors that the Transfusion, Tissue and Cells of Seville.

Methods: The quality assurance department defined the surveys to be completed by donors whose donations took place during the third quarter of 2023. Participation was stimulated via staff in charge of admission and processing of donors. The survey involved data involving: age, sex and place of collection plus a series of ten additional questions. They were related to: application form layout, assistance received from the staff during medical interview and extraction, information provided by the doctor during de interview, privacy, recommendations to follow after extraction, extraction room conditions, waiting time and global assessment of care received. The person responsible for the center's IT service designed an APP based on the standardized HTLM5 language. The novelty is that the APP is deployed in Docker containers inside servers at the local Data Processing Centre. Survey results are displayed via Java Script Canvas in the fashion of an interactive circular graph. Only fully completed surveys can be uploaded in the APP. Administration staff adapted surveys to the APP format.

Results: 700 surveys were analysed that represent 4.8% of the donations made from October up to December 2023. Each data entry by administrative staff was completed in less than a minute. Data exploitation results of the 13 analysed items are displayed on the different screens. Additional data exploitation was performed on some specific surveys.

Summary / Conclusions: The APP permits fast assessment and evaluation of the donation process. It fosters upgrading of the procedure so as to improve care quality and data exploitation. We believe that it might become a powerful tool in the near future to streamline time-consuming administrative paperwork by allowing donor direct on-line interaction with the app.

P026 | Development of a national dashboard and evidencing effective anaemia management on patient outcomes—the journey so far

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Background: In 2021, the Blood Health National Oversight Group (BHNOG) issued the All-Wales Perioperative Anaemia Pathway, to embed patient blood management (PBM), increase optimisation, improve outcomes, and reduce avoidable transfusions for patients undergoing major elective surgery. A dedicated team was established using Value Based Health Care funding to ensure implementation of the pathway into preoperative assessment clinics. Data is key to support ongoing benchmarking, and compliance to NICE quality standard QS138, and is vital for evidencing the impact of effective anaemia management on patient outcomes, including targeted treatment prior to surgery.

Aims: Develop a data dashboard to evidence the standards of the All-Wales Perioperative pathway: screening for anaemia, assessment for iron deficiency and subsequent treatment and link to patient outcomes such as length of stay, mortality, and readmission rates.

Methods: Digital Health and Care Wales have centralised access to various national data systems and the dashboard in development is built upon: Utilised Commissioning for Quality and Innovation (CQUIN) definitions to identify relevant surgeries performed in Admitted Patients Care (APC) dataset. NHS numbers used to link APC data to Welsh Reporting Services pathology dataset. Scoping opportunities to access treatment data – IV iron and transfusion data.

Results: - Dashboard allows visualisation of anaemia prevalence and iron deficiency to provide perspective and allow benchmarking by health boards. Treatment data was identified as a gap, approval has been agreed for development of a new data stream to capture this. To facilitate all new data streams, new data standard approval sought from DHCW's Welsh Information Standards Board (WISB), and Welsh Information Development Group (WIDG). Dual approach agreed: Short-term, includes direct feed for transfusion legacy data and IV iron CSV files into DHCW direct from HB. For longer term, digitisation across NHS Wales allowed opportunity to link in with these and ensure incorporation into workplans to ensure a long-term digital solution for the data feed and linked to dashboard. Also, activity to

ensure data protection impact assessments to comply with information governance, data sharing agreements.

Summary / Conclusions: The All-Wales pathway aims to improve optimisation and outcomes of patients undergoing major elective surgery using PBM, which is an evidenced-based approach to patient care. Development of a national dashboard demonstrating post-operative outcomes for anaemic patients, along with the benefits of treatment pre-operatively, is crucial for driving change and evidencing ongoing improvements. The NHS is renowned for being data rich, but information poor. As we experienced, access to robust data remains an ongoing issue. However, the resourcing for this programme has enabled the time needed to focus on the data requirements.

P027 | Construction and application of information system for quality control and quality assurance in a blood service

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Background: The quality management of blood services can be divided into quality control and quality assurance. Quality control in blood services includes quality analysis for whole blood and blood components, monitoring of equipment, materials, process environment, personnel training and operational processes. Quality assurance in blood services focuses on the construction of quality system and document issuance, document review intervals and internal audit. At present, the documents of quality system in the most blood services are recorded on paper, and the experimental operations in the quality control process are manually determined, which to some extent affects the quality management of blood services.

Aims: Build an efficient information system for quality control and quality management of blood services, which will achieve the documents work of quality system to electronize and automate the data entry or conversion in the experimental testing process of the quality control procedure, thereby improving the level of blood quality management.

Methods: Propose the requirements for information technology was based on current state of quality control and quality assurance in our blood center. Quality analysis for whole blood and blood components according to national guidelines in China. The experiment procedure for quality control is designed according to the instructions of the equipment and reagent manufacturers. The documents for the quality management system shall be digitized according to the paper content and shall be able to achieve process traceability.

Results: (1) The electronize of quality system documents has been achieved. The process of drafting, modifying, reviewing, approving, and distributing documents has been fully implemented, and employees can read the electronic files through their accounts by password, which can achieve traceability of the quality system documents. (2) Detection and recording were automated in quality analysis for whole blood and blood components, including specimen reception,

barcode printing, parameter conversion, transmission of test results, report generation, electronic signature of reports, report distribution, storage of original test records. (3) It is capable of networking the detection devices with data interfaces, including blood cell counters, coagulation analyzers, biochemical analyzers, bacterial culture analyzers and residual white blood cell counters. (4) It can automatic import/export of data in monitoring the equipment calibration, personnel operation standardization and process environment inspection by mobile platform, also can do the generation of report, storage of raw data, printing and receiving of specimen barcodes.

Summary / Conclusions: An information system for quality control and quality assurance has been established in our blood center, which can achieve the digitization of system documents and automation detection processes for quality control, and then will enhance the level of quality management of blood service.

Management and organisation—cost/effectiveness

P028 | Reducing the cost of medical waste processing with pre-donation Transfusion Transmitted Infection (TTI) screening

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Background: Blood bags with screening reactive could become one of the largest sources of medical waste in hospital services, because the volume of waste from destroying these bags is quite large. One unit of blood bag is equivalent with 100 disposable tubes from laboratory waste. In Dr. Agoesdjam Hospital, the destroying process of reactive blood bags considered as significant source in increasing of hospital cost. Various innovations are needed to reduce the amount of medical waste from donor service. One of the innovation to reduce the cost is implementation of pre blood donation transfusion transmitted infection (TTI) screening.

Aims: To evaluate the effectiveness of pre-donation transfusion transmitted infection (TTI) screening in reducing the cost of hospital's medical waste processing.

Methods: The research was conducted for 2 years (2019 and 2023) on whole blood donors. In the first year (2019), TTI screening was carried out post-donation. Blood bags with reactive screening tests were documented and the cost of destroying the blood bags for one year was calculated. In the second year (2023), pre-donation TTI screening was carried out, if TTI was found reactive, the process would not continue to phlebotomy step. The number of reactive donors and the estimated number of bags destroyed in that year were calculated. An evaluation of the cost and effectiveness of pre- and post-donation TTI screening services were carried out.

Results: In the first year (2019) TTI screening was carried out post-donation, there were 166 blood bags with reactive screening test and it was equivalent to 74.7 kg of medical waste. The cost of destroying 1 kg of medical waste is IDR 50,000 (3.20 USD), in a year required cost of IDR 3,735,000 (239.2 USD). In the second year (2023), TTI screening was carried out pre - donation, there were 142 reactive donors and it was equivalent to 63.9 kg of medical waste which costed of IDR 3,195,000 (204.7 USD). Hospital could save of Rp. 3,195,000;- (204.7 USD) per year if pre-donation TTI screening was carried out. Apart from reducing the cost of destroying medical waste, it could also reduce the cost of consumables such as blood bags, disinfectants and save the service time. With a blood bag cost of Rp. 94,000,- (6.02 USD) then the blood bag that could be saved in 2019 during post-donation TTI screening was IDR 15,604,000,- (999.5 USD) while in 2023, during pre-donation TTI screening was IDR. 13,348,000 (855.0 USD). There was an efficiency of service time in 2023 around 4260 min or 71 h per year, assuming the average length of the donor blood collection process was 30 min per donor.

Summary / Conclusions: Pre-donation TTI screening is really helps reducing the cost of hospital medical waste processing and increasing the efficiency of the use of consumables in blood donor services. It is necessary to evaluate the weaknesses of pre-donation TTI screening, especially from the donors aspect because they will waiting for the TTI screening results first to be sure whether they can donate blood or not.

P029 | When assay miniaturization is in favour of lower carbon footprint—an example with Rh-Kell phenotyping

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Background: IVD industry has a strong impact on the planet. Literature estimates that medical diagnostics industry generates 5.4 million tonnes of waste each year, which are essentially plastic based. In 2019, European commission adopted the European green deal to accelerate the transition. In this respect, we have been working on various solutions to push into this direction without affecting the patient safety.

Aims: Green deal and ESG (environmental, social and governance) are a big challenge for blood banks so we wanted to develop a multiplex phenotyping assay in order to improve the throughput, reduce reagents, consumables & waste. The idea is to help laboratories like blood banks in their ecological transition.

Methods: The developed method allows the detection of different antigens at the surface of the red blood cells with a simple multiplex test. Picovolume printed antibodies catch any red blood cells with corresponding antigens on the surface. Low blood volume (5 µL) and one well are enough to test complete Rh-Kell phenotyping in less than 5 min

Results: Our different evaluations on hundreds of samples gave us close results compare to existing technology. However their ecological footprint is lower. We quantified the weight of devices, reagents, consumables needed and even automate weight to compare the technical results in regards with ecological footprint.

Summary / Conclusions: Miniaturization and combination of blood grouping tests will help laboratories and blood banks to decrease their environmental impact, simplify their process and answer ESG expectations. The same approach can be used to other blood tests like Direct & Back typing, irregular antibodies detection and extended phenotyping.

Management and organisation—training and education

P030 | A bibliometric analysis of research publications in hemotherapy—publish and perish

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Background: Bibliometrics is the use of statistical methods to analyze books and other media of communication. Publication of research in hemotherapy journals, which are included in the "hematology" category, let the possibility of "publish and perish" because the use of classical indicators, such as journal impact factor (JIF), decreases visibility of hemotherapy journals. However, new bibliometric indicators are available.

Aims: To present a retrospection of the performance and science of research in hemotherapy.

Methods: The number of hemotherapy articles published was obtained with PubMed using Medical Subject Headings (MeSH terms [MH]): "Journal Title" [TA] AND "2018-2022" [DP] AND ("blood transfusion" [MH] OR "blood component removal" [MH] OR "transfusion" [TI] OR "apheresis" [TI] OR "donation" [TI]). The list of hematology journals in

the 2018-2022 Web of Science (WoS) as well as the 2022 JIF of each journal was obtained via Journal Citation Reports. Journals, whose main focus was publishing hemotherapy research, were identified by reviewing the aim and scope of the journal. Data from WoS Core Collection database was obtained with the following strategy: "[blood donation (Topic) OR blood transfusion (Topic) OR transfusion (Topic) OR apheresis (All Fields)] AND [2022 (Year Published)]". Regarding performance analysis, the quantity of hemotherapy articles published in scientific journals were retrieved. Regarding science mapping, a citation analysis was performed. VOSviewer was used as network visualization software. Qualitative data were reported as number (frequencies) and quantitative data were reported as median (range). Comparisons between two groups of quantitative data were performed with non-parametric Mann-Whitney U test. A *p* value of less than 0.05 was considered significant.

Results: WoS hematology category included 73 journals in 2018, 76 journals in 2019-2020, and 79 journals in 2021-2022. In 2018-2022, 11 journals were identified as focused in hemotherapy publications. Table 1 shows the number of hemotherapy articles published in hematology, hemotherapy, and other journals in 2018–2022. The median 2022 JIF of hemotherapy journals was statistically lower than the median 2022 JIF of hematology journals [3.2 (range: 0.3-28.5) vs. 1.9 (range: 0.9-4.5); *p* = 0.04]. No hemotherapy journal positioned in quartile Q1 of the 2022 WoS hematology list. In 2022, science mapping showed that 1477 (70%) out of 2111 journals published at least 1 hemotherapy article and at least received 1 citation. Only 567 (27%) out of 2111 journals were connected to each other. Using VOSviewer software, Transfusion and Vox Sanguinis were the two most connected journals with total link strength of 193 and 114, respectively.

Summary / Conclusions: The majority of hemotherapy articles were published in non-hematology journals. The use of classical indicators, such JIF, decreases the impact of research in hemotherapy journals which are included in the WoS category "hematology". However, the use of new bibliometric indicators, such as citation analysis, showed that Transfusion and Vox Sanguinis were the two most influential journals.

P030 Table 1

Year	Hemotherapy articles published in hematology journals <i>n</i> (%)	Hemotherapy articles published in hemotherapy journals <i>n</i> (%)	Hemotherapy articles published in other journals <i>n</i> (%)	Total number of hemotherapy articles <i>n</i>
2018	324 (8%)	708 (17%)	3027 (75%)	4059
2019	347 (8%)	757 (17%)	3247 (75%)	4351
2020	390 (9%)	960 (21%)	3217 (70%)	4467
2021	344 (8%)	854 (21%)	2899 (71%)	4097
2022	288 (8%)	701 (20%)	2445 (72%)	3434

P031 | To investigate the attitudes and knowledge of healthcare professionals regarding the utilization of leukocyte-reduced blood components at a medical center in Northern Taiwan

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Background: Leukocyte reduction of blood products effectively eliminates a significant number of leukocytes, cytokines, and other antigens, thereby mitigating adverse reactions following blood transfusion. Presently, many developed nations have widely adopted the utilization of leukocyte-reduced blood components to prevent febrile transfusion reactions.

Aims: Since July 2017, our hospital has instituted a comprehensive policy advocating for the use of pre-storage leukocyte-reduced RBCs (LR-RBCs) for transfusions. The aims to assess the level of comprehension among medical staff regarding the implementation of this policy.

Methods: Anonymously, a cross-sectional survey was administered to doctors and nurses in our hospital, medical technologists performing blood bank operations in different hospitals in Taiwan, and medical personnel from the Supply and Composition Sections of Blood Donation Centers located in Taipei, Hsinchu, Taichung, Tainan, and Kaohsiung. The survey covers the knowledge, implementation, and attitudes of medical staff towards various blood products, including leukocyte-reduced blood products

Results: This study included 110 physicians and 355 nurses from our hospital, along with 74 blood bank medical technologists and 97 medical staff from the Blood Donation Centers. The purpose was to conduct questionnaire surveys. The results revealed significant disparities between doctors and nurses in their comprehension of various aspects of blood transfusion medicine. They showed unfamiliarity with indications for washed RBCs and cryoprecipitate products. In emergencies with unknown blood types, 61.8% of doctors preferred type O LR-RBCs, while 22.7% opted for type O whole blood (WB). Conversely, only 45.6% of nurses chose type O LR-RBCs, with 31.6% selecting O WB. This discrepancy was statistically significant ($p = 0.017$). Moreover, only 1/5 of doctors and less than 1/10 of nurses could accurately address health insurance payment requirements for pre-storage LR-RBCs. Before the implementation of a comprehensive leukoreduction policy, only 22.8% of physicians prescribed LR-RBCs; this rose to 40.9% afterward, with a significant statistical difference ($p < 0.0001$). Following the policy, 80.1% of doctors and 70.7% of nurses observed reduced fever reactions, while 74.6% of doctors and 67.6% of nurses noted decreased wheezing or dyspnea symptoms, indicating improvement. Furthermore, 58.1% of medical technologists believed pre-storage LR-RBCs could "reduce work burden," and 40.5% believed they could "ease blood supply time pressure." However, only 25% of frontline nurses and blood bank staff received education and training on leukoreduced blood, and just

13.2% of bedside leukoreduced blood-performing nurses could correctly adhere to quality control standards. Overall, medical staff perceive using pre-storage leukoreduced products as beneficial, reducing workload and stress, and proving more advantageous for patients than post-storage leukoreduction products.

Summary / Conclusions: This study indicates that medical staff believe that utilizing pre-storage LR-RBCs can alleviate workload and stress. Furthermore, medical staff observed a decrease in transfusion reactions and highly recognized the efficacy of this policy. Therefore, the comprehensive utilization of leukoreduced RBCs proves to be a superior option for transfusion patients and frontline medical personnel.

P032 | Is it important to add transfusion training in mandatory induction training list of junior doctors and nurses?

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Background: Junior doctors and registered nurses should work within their level of competence and this applies to blood and blood product administration; they must also be proficient in monitoring for potential adverse reactions. Despite this, a lack of fundamental skill and knowledge relating to blood transfusion practice is putting patients at significant risk. The majority of errors can be attributed to inadequate patient identification, blood being stored incorrectly and, lastly, wrong blood group being transfused. To prevent this type of errors it is crucial to involve junior doctors and registered nurses at transfusion teaching especially at the time of joining the organization so that their knowledge and skills are UpToDate.

Aims: To evaluate the impact of the induction teaching of transfusion medicine among newly joined junior doctors and nurses and if the skill lab based sessions were effective in increasing confidence and interest among the participants.

Methods: The junior doctors and nurses who joined the hospital during the study period (March 2021 to March 2022) had to attend the mandatory sessions of transfusion medicine induction programme. The participants were given a pre-assessment questionnaire related to the entire transfusion chain followed by interactive training of the participants and post-training re-assessment. The training was delivered in 2 sessions—1st brief theory discussion and 2nd skill lab based demonstration and laboratory visit and escalation method in case of transfusion reaction. The data, thus collected, was recorded on a pre-designed and pretested Performa, and was then arranged in an excel spreadsheet. Statistical analysis was then carried out using SPSS Inc. According to type of data and distribution, parametric test like t test or non-parametric test like Wilcoxon signed rank test was applied.

P032 Table 1

Topic of questions	Pre-induction training score	Post-induction training score	p value
Decision making on platelet transfusion	62	97	0.000042
Decision making on packed red blood cells transfusion	84	104	
Storage of platelets in interval between issue and transfusion	21	82	
Storage of packed red cell in interval between issue and transfusion	54	97	
Thawing of fresh frozen plasma and its indication	37	102	
Pre-transfusion checklist	72	101	
Vitals monitoring during transfusion	88	103	
Rate of transfusion	82	94	
Proper consent & documentation	67	91	
Identification of transfusion reaction	62	93	
Immediate management of transfusion reaction	74	89	

Results: The mean score in the pre-training assessment was 63.9 while in the post-training assessment the mean score was 95.7; the difference was statistically significant (t -value is -4.9112; p value = 0.000042). There were significant differences in knowledge pertaining to storage temperature, shelf life of red cells and platelets, identification of transfusion reaction. All participants mentioned in the feedback skill lab based sessions boosted their confidence and interest in the training session. Details of the scores are here –

Summary / Conclusions: Education and training are fundamental to ensuring health professionals have the knowledge and skills to provide high-quality, safe and effective patient care. Our study suggests that educational session on blood transfusion practices should be included in induction training of junior doctors and staff nurses of all healthcare organization and if possible skill lab based session should be added in addition to the lecture session. This gives them confidence to work in the new clinical environment.

P033 | Abstract withdrawn

P034 | How to meet the educational needs of immunohematology laboratories in a diverse and shifting environment

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Background: One of the main missions of the transfusion medicine laboratory is to resolve immunohematology cases and guide physicians to ensure the best care for patients. This requires a specific understanding of the methods used and a good knowledge of the antigen and antibody characteristics. Additionally, the patient's history and clinical condition often contribute to the resolution of the case when available. However, transfusion medicine laboratories are facing new challenges such as staff shortages, increasing workload combined with ever

stricter financial constraints. In this evolving environment, providing easily accessible educational materials on the latest advances in immunohematology is essential for laboratories to meet these new challenges.

Aims: In 2020, a new online educational program was launched called Transfusion Science Webinar. It was initially planned to replace the in-person trainings which were all cancelled due to the pandemic, but it turned out that these webinars met educational needs that were not met by traditional in-person courses. It provides access to educational programs to a wider audience and makes them available anywhere and anytime at a lower cost. Additionally, research suggests that online learning increases information retention while saving time and resources.

Methods: An online webinar series was created in 2020 with various objectives. The first was to provide high-quality educational materials on advanced immunohematology topics. It was also important to set up surveys and collect feedback to ensure that the selected topics were sufficiently relevant to participants and allow suggestions for new topics. Additionally, it was essential to make it universally accessible by covering all time zones, free of charge, and then hosting these sessions in a location where they would be readily available on demand. Interactions between attendees and the speaker were key via a live Q&A session and awarding continuing education credits was an important goal.

Results: At the end of 2023, 16 webinars were produced and are all available on demand. Various immunohematology topics were covered such as "Hemolytic anemia and why patient history matters", "Resolution of complex immunohematology antibody problems", "D" solution: when serology meets genotyping", "Hemolytic disease of the foetus and newborn", "Sickle Cell Disease and Hemolytic Transfusion Reactions" and much more. In total, the 16 webinars were viewed by 19,280 participants (live and on-demand views), representing an average of 1205 views per webinar and a conversion rate of 52% (attended vs registered). On average, 56 questions were answered per webinar and the sessions were well received: 73% rated them excellent, 27% rated them good, and 1% rated them fair. "Resolving complex antibody cases" is the most requested topic

followed by “reviewing new molecular technologies for immunohematology laboratories” on a list of 41 requested topics. The program has been recognized as educational in several countries where continuing education credits can be awarded.

Summary / Conclusions: In this shifting era, providing online educational materials on the latest advances in immunohematology is essential for laboratories to meet their new challenges. This webinar program was initially intended to replace in-person training during the pandemic but was quickly realized to meet a critical need for easily accessible educational materials. So, this program is here to stay with new topics coming in 2024.

P035 | Abstract withdrawn

P036 | Establishing a new training system for employees of the Korean Red Cross Blood Services

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Background: Continuous education and evaluation of competency of each employee is essential to provide knowledge-based skillful donor management and to ensure the quality and safety of blood products. In Korea, blood services employees must receive job training at least twice a year in accordance with the Blood Management Act of the Ministry of Health and Welfare. Job training has to include education about (1). related laws such as the Blood Management Act, (2). guidelines and work procedures related to blood management, (3). matters related to quality control, and (4). matters related to blood safety management.

Aims: To establish an effective education and training system for employees of the Korean Red Cross Blood Services (KRCBS).

Methods: A survey of employees of different skill levels and professional background about their training need was conducted to determine what kind of training needs to be developed to improve work skills and performance. A working committee, comprised of employees with expertise in each job area, determined the method of training, curriculum, and subjects for each training target. The result of the survey served as baseline data to design the training courses.

Results: According to the survey, training to acquire specialized and detailed knowledge and skills for unit jobs, and education on basic knowledge for performing work, such as laws, standards, and guidelines were identified as the most requested areas. As in-depth discussions rather than one-sided knowledge transfer was found to be more effective for professional training about unit jobs, a face-to-face training method was chosen. This method would also allow for hands-on training, if necessary. A curriculum, comprised of 25 professional training courses, was designed to cover updates on related laws, guidelines, and educational subjects reflecting new knowledge trends. Face-to-face training will be conducted 43 times per year covering about 850 employees (35% of total employees). For basic education about laws, standards, and guidelines, education using an on-line platform

was found to be more adequate. KRCBS employees are located throughout the nation, working at various workplaces including mobile sites. To have location flexibility, this on-line platform can also be accessed on mobile phones. Educational materials for on-line education were developed and the scripts for the lectures were narrated using AI voices. Currently 37 lectures are uploaded on the on-line platform of the KRC. After completion of each lecture, trainees are subjected to evaluation and receive a certificate of completion. These educational materials are open to all employees enabling a self-study system so that employees can actively train themselves on their own.

Summary / Conclusions: A training system encompassing all areas of the blood services operations and employing both face-to-face and on-line education methods has been established. This system will ensure that skills and knowledge of employees are kept relevant which is of utmost importance to safeguard the quality and safety of donor and blood product management.

P037 | The value of digital programming in transfusion medicine continuing education

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Background: Healthcare professionals (HCPs), including those working in transfusion medicine (TM), must remain up-to-date on the newest clinical data, treatment guidelines and technological advances in their field. Continuing education (CE) accomplishes this goal and helps ensure that HCPs deliver quality care and maintain professional certification. Digital delivery models, now an established platform, have been shown to deliver high-quality continuing education.

Aims: CE programs in TM were provided via digital platforms that maximize outreach and participant engagement.

Methods: Digital delivery of CE materials was accomplished through podcasts and webinars. Podcasts offered a wide range of topics on science and technology that were relevant to laboratory professionals, presented in an easily accessible format. Webinars, on the other hand, provided comprehensive knowledge about laboratory procedures and emerging concepts in a more structured format. Featuring defined learning objectives and traditional speaker presentations, the webinars were presented to live attendees and were also available for on-demand viewing. Some webinars were accredited through professional agencies, allowing attendees to receive professional CE credits. Additionally, metrics were periodically gathered to measure audience interaction and engagement.

Results: Twelve educational podcasts were released from January through December 2023, with downloads most frequently occurring in the United States, Canada and the United Kingdom. Podcasts were downloaded over 7200 times in 2023, representing a 20.4% increase in downloads from the previous year (January to December 2022). Three CE webinars were offered in association with top scientific associations and key industry experts, covering the topics of sickle cell disease (SCD), pediatric practices in TM, and publishing in a peer-reviewed

journal. Webinars were presented via live broadcast and on-demand formats in English, with subtitles available in English, Spanish and Portuguese. The pediatric practices webinar, which sought to explore blood type options for neonatal transfusion, complications associated with red blood cell and plasma transfusion in neonates, and laboratory options for transfusion decision support when passively acquired maternal antibodies are present, drew 1011 registrations and nearly 500 attendees worldwide, with Algeria, Argentina and Australia representing the highest attendance. The SCD virtual event drove an incredible number of viewers from 55 different countries around the world. Registration surpassed 1054 attendees, with over 380 attending live. SCD topics covered the challenges associated with treatment, the research landscape of emerging genetic and cellular therapies, and the case-based experiences of a Sickle Cell Warrior. The how-to webinar on publishing one's work in a peer-reviewed journal garnered extensive interest, with registrations from 92 countries. Of 881 registrants, 237 attended the webinar in the live format, viewing the program for an average duration of 72.6 minutes and offering 78 questions/comments.

Summary / Conclusions: Digital platforms are highly effective for the worldwide delivery of CE in transfusion medicine. Podcasts are flexible and accessible, while webinars offer benefits similar to attending an in-person conference. Digital CE is a convenient, cost-effective way for TM professionals to stay updated while avoiding the stress of valuable time spent traveling and away from one's job or family.

P038 | Use of multiple language patient resources in clinical transfusion practice

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Background: In 2016–2017, the South Australian BloodSafe clinical transfusion practice quality improvement program developed a range of fact sheets for patients/consumers on blood transfusion, iron deficiency anaemia (IDA), and oral and intravenous (IV) treatment of IDA. These were targeted at patients/consumers and were intended to be used by medical and nursing staff during discussions with patients regarding informed consent and treatment options. In Australia, health

P038 Table 1

Table 1: Languages

English	Greek	Polish
Arabic	Hindi	Punjabi
Burmese	Indonesian	Serbian
Chinese	Italian	Swahili
Croatian	Khmer	Tamil
Dari	Macedonian	Turkish
Dinka	Persian	Vietnamese

care is largely delivered in English. However, as an increasingly multicultural society, there was a need for these resources to be translated into languages other than English that are relevant to Australia's migrant population (Table 1). The translation was undertaken by accredited government translators at an approximate cost of AUD\$ 40,000. In 2023, a review was undertaken of these documents to ensure clinical relevancy and accuracy, and to determine whether these resources should be maintained.

Aims: To evaluate the usage of multiple language fact sheets for clinical transfusion and iron deficiency anaemia.

Methods: Medical and nursing review of the English language versions of these fact sheets to ensure clinical currency and accuracy, and analysis of download data for the last 12 months using Google Analytics (GA) to determine the usage rates.

Results: A review of the contents showed that the fact sheets were current and clinically accurate. Analysis of the download data for 1 January to 31 December 2023 showed that the multiple language versions of the 'Having a Blood Transfusion' and 'Intravenous Iron Transfusion' fact sheets were used extensively. Analysis of country of origin showed usage was significantly higher outside of Australia than was anticipated. Selected data is shown in Table 2.

Summary / Conclusions: Results of this analysis showed that these multiple language fact sheets were used extensively. While the resources were designed and intended for an Australian audience, the analysis showed that they had a greater usage outside of Australia, particularly in Arabic-speaking countries and Italy. This was an unexpected finding. This analysis demonstrated the utility and reach of web-based resources and how a country such as Australia, which creates information resources in languages other than English, can make these resources

P038 Table 2: Download data of multiple language resources

Having a blood transfusion	Downloads	Intravenous Iron Infusions	Downloads	Download countries
Arabic	10,087	Arabic	57,710	Saudi Arabia (16,869), Jordan (7666), Egypt (4700), Syria (3697), Kuwait (3657), UAE (3248), Iraq (2635)
Hindi	5570	Italian	31,749	Italy (28,926), Switzerland (1954)
Greek	2642	Tamil	7996	India (7918)
Turkish	864	Turkish	5818	Turkiye (5335)
English	741	English	4503	Australia (3886)
Italian	586	Croatian	3233	Croatia (1920)

available to other countries and potentially contribute to improved patient care. The needs of non-Australian users will now be considered when reviewing online resources. An area for future development is to explore whether translated versions of information can be created using artificial intelligence such as ChatGP and Google Translate and whether these are clinically accurate and appropriate for native speakers. This could reduce the costs and increase the number of languages available.

P039 | Access to continuing education credit and translated content may be crucial to online user

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Background: Global access to high-quality transfusion medicine (TM) education continues to be a driver of advancement in this specialty. Internal survey responses from health care show respondents seek Continuing Education (CE) credits and improvements in quality and depth of training. We applied this to the rollout of a global, online, free TM education platform. Here we present user comparison data related to CE credit and language translation access.

Aims: The aims of this analysis were to evaluate the respective impacts of 1) CE credit availability and 2) language translation implementation on user content completion, defined here as rate of completion of an online lecture and subsequent post-lecture knowledge check (PLKC).

Methods: Registered platform users were placed into 4 Areas: Asia-Pacific (APAC), Europe-Middle East-Africa (EMEA), Latin America-Canada (LAC), United States (US). Standard platform user adoption and retention initiatives were employed across all Areas except for the interventions described in Aims 1 and 2. For Aim 1 the platform certifies 0.5 CE credit equivalents upon completion of a PLKC consisting of 5 questions. The PLKC can only be accessed after completion of the associated lecture. Rates of PLKC completion for registered US and non-US users were compiled for 18 months (1.7.2022 to 31.12.2023). For Aim 2 the platform released Spanish and Portuguese subtitled and primary content during an 8-month evaluation period (1.5.23 – 31.12.23). Completion rates for PLKC in registered Latin America-Canada users and a deidentified comparator Area with no translation investment during this period were compiled. Statistical analysis on deidentified data was performed for both Aims.

Results: These results demonstrate notable, significant differences between users of an online transfusion medicine education platform. In Aim 1 the PLKC completion rate in users who could obtain CE credits by completing the PLKCs was significantly higher than those who could not. The significant difference between US users and a theoretical user group with a benchmark PLKC completion rate of 40% is also notable. In Aim 2 PLKC completion rates were significantly higher in LAC users, who had access to material translated into and in some

cases recorded in Spanish and Portuguese. The findings of these Aims analyses could help focus future deployment of these and other targeted global educational initiatives. Taken together, wider data analysis over a longer period may more fully assess.

Summary / Conclusions: These results demonstrate significant differences between users of an online transfusion medicine education platform. In Aim 1 there was significant difference in PLKC completion rates between US users, who could obtain CE credits by completing the PLKCs, and non-US users, who had no access to CE credits during the analysis period. The significant difference between US users and a theoretical user group with a benchmark PLKC completion rate of 40% is also notable. In Aim 2 there was a significant difference in PLKC completion rates between LAC users, who had access to material translated into and in some cases recorded in Spanish and Portuguese, and users in a comparator Area where translation did not take place. The findings of these Aims analyses could help focus future deployment of these and other targeted global educational initiatives. Taken together, wider data analysis over a longer period may more fully assess.

P040 | Development of a model system to address disparities between High-Income Countries (HICs) and Lower- and Middle-Income Countries (LMICs) in transfusion medicine education

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Background: Our team was tasked in a global health event with creating an initiative for advancing transfusion medicine education and providing an equitable environment that elevates peer voices in LMICs. Challenges in local regulations and resource availability, language hegemony, education distribution, sustainability, and outreach were discussed.

Aims: Investigate benefits of an open-source platform consisting of a resource database, physician forum, and graphic design suite on reducing barriers in transfusion medicine education and collaboration. Apply linguistic theory and interpretation models to allow for accurate translation of content into other languages; reduce the impediments of English language hegemony toward medical education and international collaboration. Identify strategies for iterative improvement to database contents and translation fidelity using feedback from users, field testing, and conferences.

Methods: Objectives were divided according to expertise and frequent team meetings were held to maintain coherence and assess need for changes in direction. Literature was reviewed to avoid common pitfalls and determine linguistic models that would fulfill the needs of a multilingual collaborative environment; these were extrapolated to a protocol for implementation and iterative improvement. Current international education and cooperation initiatives were researched; areas where information exchange could be more equitable and sustainable were identified. Principles of active learning, for

example, primary frameworks of stepwise refinement and inductive reasoning, were explored. A prototype platform was drafted.

Results: Our solution was presented as an online, open-source, free-to-use platform. Accessible and dynamic, it targets areas of need and addresses diverse situations and specific problems; practitioners will be able to direct their learning towards relevant concerns and share their wisdom with international colleagues across languages and time zones. To address cultural influence on language structure, we found that professional translation and field testing were effective at improving quality, so a hybrid neural and statistical machine translation approach was selected for its fidelity and ability to iteratively improve through quality ranking. The timeline for short to long term is designed so that leadership and editorial roles will pass from HIC colleagues to LMIC colleagues, becoming more equitable and representative over time. Concerns were raised about loss of service during crises and regional lack of internet availability. While local infrastructure limits accessibility, our approach was nonetheless determined to be highly practical given its benefits. Furthermore, given the increase in internet availability in the global south over the past two decades, it is expected that an increasing number of regions will have access to the platform over time. This solution was awarded first place due to its engaging educational model, equitable and bidirectional learning system, and realistic vision for sustainability and growth over the short, mid, and long term.

Summary / Conclusions: The objective and true power of our solution is the design of a system that supports bidirectional learning between colleagues in HICs and LMICs. By working to elevate the voices of our colleagues in LMICs, we hope it sets the stage for equitable sharing of knowledge, experience, and perspective; and thereby, the advancement and flourishing of a global community in transfusion medicine.

P041 | Building a culture of voluntary blood donation in Latin America—the LUDS initiative

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Background: In most countries in the Latin American region, blood services face challenges in making sufficient blood available while ensuring its quality and safety. In 2020, only 47.3% of the collected blood came from voluntary donors, (the remainder originated from replacement donations). Additionally, in three countries, over 90% of the blood came from regular altruistic donors, while in others, this percentage did not surpass 10%. The Grupo Cooperativo Iberoamericano de Medicina Transfusional (GCIAMT) is a non-profit scientific society founded in 1994 and has more than 1250 individual members from all Ibero-American countries. GCIAMT's leaders promoted an

P041 Table 1

	December 16, 2021	December 15, 2022	December 14, 2023
Countries	17	15	16
Blood Services	137	101	271
Collection Sites	151	201	321
Voluntary Donors	13,287	13,525	20,864

intervention to improve voluntary non-remunerated blood donation (VNRBD) in the region which remained a major weakness.

Aims: To describe the Latinoamérica Unida Dona Sangre initiative (LUDS, as per its acronym in Spanish), which has two main objectives: (1). Develop an educational activity targeting blood service professionals (medical, technical, and nursing staff), social organizations, community leaders, and government officials from Ministries of Health (2). To conduct a simultaneous and regional blood donation event to validate the educational process, raise awareness of the need for VNRBD, and unify and amplify the message across the entire region.

Methods: Three editions of LUDS were conducted from 2021 to 2023. Support was provided by the Pan American Health Organization. Virtual meetings were held from June to December each year, where the participants had the opportunity to learn from expert professors, share experiences, and collaborate on donor care procedures for the simultaneous annual blood donation event held every December. Participants suggested topics through a survey (donor, eligibility, organization of blood drives, marketing, community promoter role, regulations, human resources training, equipment management). A coordinated plan was implemented to convey specific educational messages for the donation event through various channels and social media. A dedicated LUDS space was designed on the GCIAMT official website, providing access to all training sessions: <https://gciamt.org/latinoamerica-unida/#>. Participants completed self-administered surveys and were awarded a Diploma in recognition of their participation.

Results: A. Educational meetings: Five educational workshops were conducted each year, with an average attendance of 220 participants (2021), 230 (2022), and 243 (2023) participants. **B. LUDS Donation Events**

Summary / Conclusions: The situation of VNRBD in the region is far from optimal, indicating an urgent need for improvement. To our knowledge, LUDS is the world's first initiative of this kind. It is an experience that, through organizing the blood donation event, translates academic content into action, raises awareness of blood donation, and fosters cooperation among Latin American countries. Through a comprehensive communication strategy, we successfully unified messages, and channels across 17 countries in Latin America. With the integrated communication strategy implemented in LUDS, we surpassed geographical boundaries, bringing together blood services and community leaders in the region for a common purpose: to enhance the quality, safety, and sufficiency of blood transfusions.

P042 | Enhancing blood transfusion safety—assessing knowledge and training impact among paramedical staff in a multidisciplinary laboratory

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Background: Blood transfusion is a life-saving therapeutic procedure, not devoid of risks. Mastery of transfusion safety rules is, hence, crucial. This highlights the importance of maintaining a high level of knowledge regarding rules and best professional practices among practitioners particularly multidisciplinary laboratory technicians,

Aims: The objective of our study was assessing the knowledge of transfusion safety among the technicians of the multidisciplinary laboratory of Ben Arous regional hospital, as well as the impact of a training action on this thematic.

Methods: We conducted a cross-sectional, quasi-experimental, pre-post comparative and evaluative study of an educational action between January and May 2023. All members of the paramedical staff working as technicians were included. The assessment tool used was a survey consisting of 19 multiple choice questions (MCQ) focusing on professional data, as well as knowledge and practices in transfusion safety. A training session was organized using a PowerPoint presentation, followed by a final evaluation using the same questionnaire immediately after the session.

Results: Our study showed that the knowledge and attitudes of paramedical staff regarding immunological safety in blood transfusion were generally acceptable. The overall rate of correct answers was 82%. The average score was 7.4/13. Seven items had a rate of correct answers above 80%, including the one relating to the antigenic and serum characteristics of the AB group, which achieved a 100% correct response rate. Six technicians obtained at least 9 correct MCQs, and none answered all questions correctly. There was a notable improvement in responses after the training action. The overall rate of correct answers improved significantly ($p < 0.001$) from 82% to 98%, with an overall progression percentage of 16%.

Summary / Conclusions: In light of our results, we proposed a series of recommendations summarized as follows: Continuous training, periodic evaluation of practices, and monitoring of the application of national reference texts. This could be achieved by activating hospital blood transfusion committees and committing appropriate human and material resources.

P043 | Audit of blood request forms—a district headquarter hospital experience

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Background: Transfusion audits are essential for evaluating and educating individuals in need of blood components, offering valuable insights into the appropriateness and safety of transfusion practices. Through meticulous analysis of transfusion data, audits identify areas for improvement and contribute to enhanced patient care. Moreover, they facilitate the dissemination of evidence-based guidelines and best practices among healthcare professionals engaged in requesting blood components. Overall, transfusion audits play a pivotal role in promoting optimal transfusion practices and safeguarding patient safety.

Aims: The objective of this study was to determine the frequency of incomplete or inadequate laboratory request forms prescribed at a district headquarters hospital in Mandi Bahaulddin.

Methods: A comprehensive examination was conducted on 5000 request forms, with meticulous recording and analysis of relevant information on a separate sheet. The recorded data included essential details such as demographic information, required investigations, patient location, sample collection date and time, sample type, clinical history, as well as the name and department of the clinician involved.

Results: Upon analyzing 5000 request forms, it was found that none of them were fully completed. Notably, only the patient's hospital number and the required investigation fields were consistently filled in all request forms, achieving 100% completion. However, completion rates for other crucial fields were notably lower, with the patient's name, age, gender, and location being complete in only 41.46%, 9%, 9.75%, and 44.56% of the forms, respectively. The clinical history or diagnosis section was present in a mere 2.57% of the request forms. Additionally, the date and time of sample collection were available in 20.18% and 19% of the forms, respectively.

Summary / Conclusions: The substandard completion of laboratory request forms has led to inadequate utilization of laboratory services. Continuous efforts are essential to raise awareness among physicians regarding the importance of accurately completing these forms. This sensitization is necessary to ensure optimal utilization of laboratory services and enhance overall patient care.

P044 | Assessment of blood transfusion knowledge and practices among junior doctors

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Background: Establishing a robust knowledge foundation is imperative for the successful implementation of effective procedures, particularly in the intricate domain of blood transfusion. Within the framework of the institution's quality assurance protocols, the competence of junior medical practitioners, including interns and residents, undergoes evaluation at the initiation of their training.

Aims: This study aims to assess the knowledge levels of junior doctors and discern any potential requirements for additional training.

Methods: A prospective, cross-sectional evaluation spanning one month was conducted, encompassing junior physicians from diverse medical and surgical departments at La Rabta University Hospital. The evaluation comprised 24 questions and was administered at the commencement of their training. It addressed three critical domains: pre-transfusion work-up, transfusion procedure, and adverse reactions in recipients. All participants provided informed consent, and data collection occurred through a Google Forms link.

Results: The study included 122 participants (70.5% women), with 74.6% being residents. The Overall Rate of Correct Answers (RCA) was 33.3%, ranging from 0.8% to 99.2%. Notably, the lowest RCA was observed for post-transfusion prescription. The median score was 8/24, with scores ranging from 2 to 14. Only two questions achieved an RCA higher than 80%, while 17 questions scored below 50%. The predominant score was 9/24 (17.2%). Specific domain scores were as follows: pre-transfusion work-up 34.9%, transfusion procedure 43.9%, and adverse reactions in recipients 25.6%.

Summary / Conclusions: This study underscores the significance of a qualitative process and the preparation of junior physicians at the commencement of their training. It emphasizes the need for tailored training programs and regular assessments to identify and address knowledge gaps.

P045 | Abstract withdrawn

P046 | Abstract withdrawn

P047 | Laboratory aspects of transfusion training for haematology postgraduate doctors in training (PGDiTs)—results of a UK-wide survey

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Background: Transfusion knowledge and experience is essential for all haematologists, and this is reflected in the UK haematology training curriculum and examinations. Incidents reported to the UK haemovigilance scheme (SHOT) repeatedly highlight the importance of transfusion training for patient safety. Haematology PGDiTs must undertake training in haemato-oncology, coagulation, red cell disorders, immuno-haematology, paediatrics and transfusion. Transfusion training, especially laboratory aspects, is often reduced due to competition from more obviously clinically focussed subspecialties. Previous UK-wide surveys in 2008 and a smaller snapshot survey in 2012 found widespread dissatisfaction and variability in how transfusion training was delivered.

Aims: The Royal College of Pathologists (RCPATH) UK Transfusion Medicine Standing Advisory Committee (SAC) led this survey to understand the current status of transfusion training across the UK from a PGDiT perspective and facilitate improvements.

Methods: A 30-question online survey was distributed to UK haematology specialty PGDiTs in June-July 2023.

Results: 150 responses (response rate 24%) were received from PGDiTs at different stages of training (England 82.7%, Northern Ireland 6.0%, Scotland 6.0%, Wales 5.3%; year 1 8.7%, year 2 22.2%, year 3 22.0%, year 4 29.3%, year 5 17.3%). 48 (32.0%) PGDiTs had undertaken a dedicated transfusion post, in 13 different training regions. The posts undertaken were at Blood Services (31.3%), hospitals (33.3%) or both (35.4%). Deficiencies in laboratory aspects of transfusion training were highlighted frequently. The most common barrier to training identified by PGDiTs was lack of exposure to the transfusion laboratory (75%), followed by other clinical commitments taking priority (74%) and lack of provision of dedicated transfusion training (70%). The most frequent suggested solution to improve training was more laboratory-based teaching (74%), followed by more teaching at local hospital level (67%) and a dedicated transfusion post (61%). Of those PGDiTs who had received teaching in hospital outside of a transfusion post, 85.4% (88/103) reported being taught laboratory aspects of transfusion. Less than half of these PGDiTs (42/103, 40.8%) had spent time in the hospital transfusion laboratory during these sessions. The most common transfusion queries handled by PGDiTs (transfusion reactions, emergency transfusion and special transfusion requirements)

require a good understanding of laboratory processes. In terms of overall satisfaction with transfusion training, 45 (30.0%) PGDiTs were very satisfied/satisfied, 46 (30.7%) were neutral and 59 (39.3%) were dissatisfied/very dissatisfied. 48 (32.0%) did not think their training programme would equip them to support a hospital transfusion laboratory at consultant level and 30 (20.0%) were unsure.

Summary / Conclusions: This survey continues to highlight ongoing widespread dissatisfaction and inconsistent approaches to transfusion training for UK haematology PGDiTs. Laboratory aspects of transfusion training in particular need improvement. The Transfusion SAC is working with other stakeholder organisations and trainees on resources to signpost training opportunities, and highlight the importance of dedicated training time in the hospital laboratory for all haematology PGDiTs to ensure safe patient care. The RCPATH is developing a Pathology Portal online education resource and laboratory management webinars.

P048 | Pathology portal—developing a novel, interactive education portal for transfusion medicine in the UK

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Background: Transfusion is essential for patient care with ~2million units of blood and components per annum administered across the UK. There are significant concerns around current transfusion education with a lack of integration between clinical and laboratory services that can lead to ineffective blood management and patient safety concerns as highlighted by the Serious Hazards of Transfusion (SHOT) UK Haemovigilance scheme. Recent national feedback from trainees has highlighted key gaps including decreased exposure of trainees to laboratory transfusion. There is a pressing need for improved education in transfusion medicine not only in the UK but also internationally. The Pathology Portal is an innovative award-winning learning resource for pathology trainees and consultants, launched in Aug 2022 by The Royal College of Pathologists (RCPATH) and Health Education England (HEE). Published content includes > 4000 modules with phased roll-out covering 17 pathology specialties, that have been collectively accessed >100,000 times since the Portal launch (www.rcpath.org/profession/pathology-Portal.html).

Aims: We are developing educational resources for transfusion medicine to promote greater consistency and efficiency in training. We aim to develop a sustainable learning platform which is well-structured, up to date, aligned to the relevant curricula and freely available, to support training in transfusion medicine for healthcare professionals.

Methods: Key transfusion learning objectives were identified from the curricula for haematology trainees and higher specialist scientists in training (HSST). This has been coupled with a comprehensive review of a wide range of educational resources available, identifying key gaps where development is needed. We have recruited clinical and scientific representatives in transfusion medicine from professional organisations across the 4 UK nations and the International Society of Blood Transfusion (ISBT) to support content development and provide editorial oversight. This collaboration will also allow access and engagement for international trainees.

Results: We have collated >400 transfusion learning resources from several sources including BSH Guidelines, SHOT, British Blood Transfusion Society, UK Blood services and ISBT, aiming to cover all aspects of curricular, management and additional topics. Hosting links with permissions to high-quality material from trusted resources will aid learners to access content easily. We are now also developing novel material with a focus on case-based teaching greatly valued by learners. Management training has been identified as a key learning gap and we are recording webinars addressing real-world examples of lab management issues. User engagement including new and return users will be tracked. Quantitative and qualitative feedback, via a diverse focus-group of volunteers, will inform modifications to support more effective learning.

Summary / Conclusions: We have designed transfusion medicine educational resources for the Pathology Portal as a sustainable learning platform, freely available to trainees and other healthcare professionals. A particular challenge is updating and ensuring continuity of resources to maintain accuracy, which will require ongoing input.

Management and organisation— Risk models, standards and regulation

P049 | Abstract withdrawn

Management and organisation— blood supply management and utilization

P050 | Discard rate of red blood cells and their management in Japan

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Background: Efforts to reduce the red blood cell (RBC) discard rate are a general part of broader patient blood management initiatives that focus on minimising unnecessary transfusions, improving patient care, and reducing the overall demand for blood products.

Aims: This study aimed to determine the extent of the RBC discard rate in Japan and evaluate the factors contributing to wastage.

Methods: The blood discard reports from the nationwide questionnaire survey on transfusion medicine, conducted in fiscal year 2022 as a project commissioned by the Ministry of Health, Labour and Welfare of Japan, was retrospectively reviewed. The RBC discard rate was calculated as the ratio of the annual number of discarded bags to the annual number of purchased bags at each institution.

Results: Survey responses were received from 4824 (52%) of 9277 institutions that received blood products from the Japanese Red Cross Society. The annual RBC purchases and discards for the entire survey were 5,147,405 and 71,794 units, respectively (the rate of discard was 1.4%). The most prominent reasons for RBC discard were expired blood (85%), inability to reissue to another patient (8%), and poor storage management (3%) (Table 1). The annual units of purchased RBC increased in proportion to the number of hospital beds, but the annual units of discarded RBC did not significantly change from hospitals with 200–299 beds. RBC discard rates were almost zero in clinics, increased in hospitals with 200–299 beds, and decreased thereafter (median [range]: 0% [0–100], 1.6% [0–100], 0.98% [0–31.6], 0.36% [0–100] in clinics and hospitals with 0–99 [*n* = 3,007], 100–299 [*n* = 1,104], 300–499 [*n* = 447], and >500 beds [*n* = 266], respectively). Increased general anesthesia surgery, history of massive bleeding, and medical specialty (cardiovascular surgery, obstetrics and gynecology, and emergency medicine) were associated with increased RBC discard rate in hospitals with 100–299 beds (median: 2.3% vs. 0.9%, 2.8% vs. 1.3%, 2.6% vs. 1.4%, 2.6% vs. 1.1%, and 2.2% vs. 1.4%, respectively).

Summary / Conclusions: The 21-day storage period for RBCs in Japan until March 2023 may have led to the discard of expired RBCs. The increased number of surgery and preparedness for major bleeding may have increased the hospital's RBC inventory and discard rate because of fewer patients to divert in smaller hospitals. Additionally, smaller hospitals that require large-volume transfusions should encourage collaboration with large hospitals, consolidate medical services, and consider implementing blood rotation so that near-expiring RBCs can be collected and reused to reduce waste.

P050 Table1 The annual number of RBC discards for each reason

Bed capacity (# of beds)	0-99	100-299	300-499	>500
# of institutions participated	3007	1104	447	266
Expired blood	4279 (81%)	15301 (91%)	7182 (87%)	3907 (69%)
Damage	15 (0.3%)	46 (0.3%)	107 (1%)	162 (3%)
Poor storage management	43 (0.8%)	170 (1%)	295 (4%)	688 (12%)
Inability to reissue	817 (15%)	1110 (7%)	529 (6%)	509 (9%)
Others	145 (3%)	119 (0.7%)	140 (2%)	361 (6%)

P051 | Blood transfusion trends in Taiwan—national inpatient data, 2018 to 2021T Chiueh¹, Y Tu²¹Laboratory Medicine, ChangGung Memorial Hospital, Taoyuan,²Laboratory Medicine, New Taipei Municipal TuCheng Hospital, New Taipei City, Taiwan, Republic of China

Background: Patient blood management (PBM) is essential for the proper use of limited blood products. Restrictive transfusion is mostly preferred in terms of clinical outcomes (Blanca, Eur J Intern Med, 2023). After introducing the PBM concept, publishing the national guidance of blood transfusion, and providing continuing education programs to health care providers, calls for blood shortage are still frequently made even though the highest donation rate remains in Taiwan. Inappropriately excessive utilization of blood products is generally believed and worried, but lack of supportive evidence statistically. In the US, the analysis of national trends in red blood cell (RBC), plasma, platelet, and cryoprecipitate transfusions showed a decline in RBC and plasma transfusions, stable platelet transfusions, and increased cryoprecipitate utilization from 2015 to 2018 (Goel, Blood Advances, 2021).

Aims: This study investigated the annual proportion of discharges transfused with a given blood component stratified by the hospital level from 2018 to 2021 in Taiwan.

Methods: Taiwan's National Health Insurance Research Database (NHIRD) between 2018 and 2021, was applied for calculating the annual proportion of discharges transfused with a given blood component for each hospital. A total of 413 hospitals of 5 levels were included for analysis. Three blood components were analyzed including RBC, FFP, and platelet. Annual transfusion proportions of hospitals were illustrated by the box-and-whisker plot stratified by the hospital level.

Results: The percentages of hospitalizations with a given blood component varied greatly among hospitals even those belonging to the same hospital level (Table 1). Generally, the percentage of hospitalizations with an RBC transfusion was about 12% on average for most hospitals irrespective of their levels except small local hospitals. Slightly increasing trends in RBC transfusion percentage were noted from 2018 to 2021. The percentages of hospitalizations with an FFP transfusion were hospital-level dependent. The highest 3% on average was observed for medical centers, 2% and 1% for regional and local hospitals, respectively. Slightly increasing trends of FFP transfusion percentage were also noted. Platelet transfusions were also high and nearly remained stable through 4 years, around 3% for medical centers, 1.4% for regional and local teaching hospitals, and 0.8% for regional hospitals. Small local hospitals presented widely variable data on all transfusion percentages.

Summary / Conclusions: Compared with US data, folds higher percentages of 3 blood component transfusions were observed. Emphasizing “less is more”, the concept of PBM, is always a great challenge in Taiwan which maintains a superior high blood donation rate.

P051 Table 1. Hospital's average annual proportion of discharges transfused with a given blood component in Taiwan

	RBC	FFP	Platelet
Medical center	11.95 ± 1.94% (12.71 ± 1.89%)	3.01 ± 0.98% (3.33 ± 1.18%)	2.93 ± 0.86% (3.17 ± 0.89%)
Regional teaching hospital	10.94 ± 2.74% (13.16 ± 3.02%)	2.03 ± 1.10% (2.26 ± 0.98%)	1.34 ± 0.58% (1.54 ± 0.52%)
Regional hospital	11.38 ± 4.02% (12.73 ± 3.96%)	1.37 ± 1.02% (1.41 ± 0.84%)	0.83 ± 0.46% (0.98 ± 0.66%)
Local teaching hospital	9.72 ± 2.81% (12.33 ± 3.14%)	1.76 ± 0.97% (2.36 ± 1.30%)	1.38 ± 0.85% (1.64 ± 0.86%)
Local hospital	22.07 ± 35.37% (20.98 ± 27.81%)	4.22 ± 12.4% (3.23 ± 7.43%)	1.62 ± 3.24% (1.94 ± 3.69%)

Note: 2018 data on the upper row, 2021 data on the lower row.

P052 | Adverse events in pediatric large volume leukapheresis for peripheral blood stem cells collection—critical variables to take into account for a safe procedure in children, a single site experience

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Background: Pediatric peripheral blood stem cell collections (PBSCC) have become a routine medical procedure. Different medical and technical difficulties derived from the patient size must be considered before and during the apheresis. Vascular access, extracorporeal blood volume and anticoagulation management are the main concerning factors. The bigger challenge are very low weight children.

Aims: To analyze adverse events (AE) during large volume leukapheresis (LVL) in cytokine-mobilized patients and to evaluate variables affecting the PBSCC.

Methods: It is a cross sectional retrospective study. The LVL procedures analyzed were performed using Spectra Optia CMNC protocol. Parameters studied included type of donor, diagnosis, age, gender, blood volume, pre and post procedure complete blood and CD34+ counts. The details regarding blood priming, venous access, flow rates, volumes processed, duration of procedure, adverse reactions, volume collected, stem cell yield expressed as CD34+/kg, were evaluated. Categorical data was expressed as frequencies and percentages, continuous data as mean and ranges. Descriptive statistics was used to determine factors associated with adverse events LVL, and other variables of PBSCC.

Results: Between January 2017 to April 2023, 66 LVL were programmed on 50 children (male 30; female 20) including 6 healthy donors and 44 patients, 9 had hematologic tumors and 35 had solid tumors. The age range was 7 months to 16 years. Venous access was established using a dual lumen central catheter in 49 (98%) children, inserted in the femoral vein. Three LVL were canceled because of obstruction of vascular access. A total of 63 LVL were performed, 53 (84%) needed priming of the apheresis circuit. The average blood volume was 1815 (561–4865) The flow rate ranged between 15 and 77 mL/min depending on body weight and tolerability. A median of 3.8 (2.1–6.2) blood volumes were processed. The average duration was 212 (104–391) The median cell dose collected per procedure was 5.96 (0.35–33.08) CD34+ × 10⁶/kg. Adverse events were recorded in 26/63 (36.5%) LVL: 18 (28.6%) hypotension, 12 managed with volume expansion and 6 needed red blood cells transfusion, 4 (6.34%) measure hypocalcemia requiring calcium administration, 2 (3.17%) hypokalemia, 2 (3.17%) thrombocytopenia with platelet transfusion requirement. No LVL needed to be interrupted by any of these adverse events. The pattern of AE was shown to vary according to the age of the donor. Older donors had higher incidence of AE related to hypocalcemia than the youngest donors who suffered mainly cardiovascular complications due to hypovolemia and anemia.

Summary / Conclusions: In the present study, AE related to LVL in pediatric patients/donors were higher than other reports in the literature, where the most common AE reported was thrombocytopenia. The prevalence of hypotension is striking in this study and it should force a review of procedures

P053 | A single-center blood wastage reduction initiative in a tertiary academic center

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Background: Blood products are a precious resource, and concerns about supply-demand mismatch highlight the importance of conserving blood. Blood wastage is a pertinent issue, and past quality improvement (QI) projects have demonstrated success in reducing wastage through root-cause analyses and feedback. At our tertiary academic center, we found a high baseline blood wastage of 739 products (2.2% of 34,225) annually, costing CAD \$173,961 per year.

Aims: Achieve an average annual blood wastage cost of less than \$100,000.

Methods: A retrospective audit of hospital wastage data revealed various causes of wastage, including error-prone red blood cell (RBC) and plasma storage containers, inappropriate albumin use, failure to return products within required time limits, preventable wastage of home-use products, and overactivation of the massive hemorrhage protocol (MHP) leading to plasma wastage. A multidisciplinary QI team was assembled to address wastage. In 2023, the following interventions were deployed: Online training, newsletters, and meetings for hospital staff on proper blood handling and reporting of wastage events to the hospital incident system. Close monitoring with a tracking dashboard and online incident reports, enabling investigation into all wastage events. New institutional guidelines for albumin use and extending return time from 1 to 24 h. Design and validation of a new transport container to address transport-related RBC wastage. Personalizing the volume of home-use products prescribed by immunology physicians.

Results: We observed a numerical decrease in overall blood wastage after the interventions were implemented. In 2023, 369 products (1.4% of 26,165) were wasted, costing \$106,658, compared to the baseline wastage of 739 units costing \$173,961 annually from 2019 to 2022. Notably, there was a significant reduction in the wastage of plasma ($p = 0.002$), intravenous immunoglobulin ($p = 0.047$), platelets ($p = 0.02$), and 25% albumin ($p = 0.02$) post-intervention. Although we nearly reduced annual wastage costs to less than \$100,000, the reduction in costs did not reach statistical significance or the QI initiative target ($p = 0.37$).

P053 Table 1: Pre- and post-intervention annual wastage of blood components.

Blood Product	Pre-Intervention Annual Wastage (%)	Post-Intervention Annual Wastage (%)
RBCs	72 (1.18%)	47 (1.04%)
Platelets	14 (1.14%)	2* (0.46%)
Plasma	181 (6.06%)	142* (18.73%)
Fractionated products	478 (2.13%)	224* (1.31%)

Note: * $p \leq 0.05$.

Summary / Conclusions: Our QI project demonstrated the importance of root-cause analyses and monthly feedback, leading to \$67,303 in annual cost savings. Uniquely, we monitored the wastage of all blood products, enabling us to identify and address catastrophic losses of home-use products. We nearly reached our goal of reducing wastage costs to less than \$100,000 in 2023, achieving an overall blood wastage rate of 1.4%, down from a baseline of 2.2%. Previous studies have demonstrated that an RBC wastage rate of less than 1% was achievable in a large academic center. Thus, further interventions based on ongoing audits will be implemented in 2024 to target an annual wastage cost below \$100,000 and a wastage rate of less than 1% for all products: Transition to preparing lyophilized coagulation products at the bedside. Development and validation of a cooler to store warm thawed plasma for MHPs at appropriate temperatures to facilitate the return of unused units to blood bank inventory. Education highlighting the impact of unnecessary MHP activation on blood wastage.

P054 | Plasma self-sufficiency for plasma derived medical products (PDMP) from unpaid and uncompensated donors is feasible in countries with a high usage of immunoglobulin

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Background: The European Union is not self-sufficient with plasma for PDMP but relies on import of plasma from paid donors. This makes the supply, especially of immunoglobulin (IG), vulnerable as seen during the Covid-19 pandemic, where IG treatment for European patients was restricted in several countries. In some European countries, the introduction of paid plasma donations by commercial operators have been disruptive to collection of blood for transfusion. Payment for donations is not in accordance with Art 21 of the Convention on Human Rights and Biomedicine, which states: The human body and its parts shall not, as such, give rise to financial gain.

Aims: To establish national self-sufficiency with plasma for PDMP through a plasmapheresis scheme with unpaid and uncompensated donors in a country with a very high usage of IG and within the public transfusion services.

Methods: Until 2007 recovered plasma was adequate to self-sufficiency with IG. Decreasing usage of RBC resulted in a reduced amount of plasma from whole blood (WB). Simultaneously, the usage of IG increased. In 2013 only 75% of the amount of plasma stipulated in the national contract on fractionation was delivered. It was decided to initiate a plasmapheresis program corresponding to the contractual requirements. During the Covid-19 pandemic only 80% of the necessary IG was available, which resulted in a political decision of achieving self-sufficiency with plasma for PDMP. As the WB donations had decreased, there was facilities and donor resources for the initial part of the program, whereas

the goal of 100% self-sufficiency demanded new centers and increased donor recruitment. Two new centers with 25-30 beds are in operation and construction of two additional centers have been announced opening in 2025 and 2026. In sparsely populated areas, collection sites have been expanded to 10 beds. Further expansion will be decided and in operation in 2030 where full self-sufficiency should be achieved.

Results: After the decision of initiating the plasmapheresis program in 2013, the first deliveries of source plasma took place in 2015 (21 t) and have increased since then to 86 t in 2023. In the same period recovered plasma has dropped to 39 t in 2023 compared to 85 t in 2004. Thus, the total amount of plasma delivered for fractionation in 2023 was 125 t. In 2022 the usage of IG was 1031 kg or 174 g/1,000 inhabitants. Considering a yield of 5 g IG/L plasma, the needed plasma for this amount is 206 t or 35 kg plasma/1000 inhabitants. The current deficit thus is 81 t of plasma. As the two planned centers has a capacity of 35 t and 17 t respectively, the deficit in 2028 will be 29 t. The expectation is that more plasma can be collected in existing centers and that more will be planned. On the other hand, usage of IG may increase again after a drop during and after the Covid-19 pandemic. In 2022 the total amount of plasma collected varied between 11 and 33 kg/1000 inhabitants among the five blood centers. This was caused by different pace of the political decisions and shows that is a potential for more collections.

Summary / Conclusions: (1). Self-sufficiency with unpaid and uncompensated donors is possible even with a very high usage of IG, (2). Keeping WB and plasma donations from the same pool of donors in public blood services prevents disruption of supply for transfusions in hospitals, (3). The increased donor pool contributes to the overall robustness during holidays, pandemics, crisis, conflict and war.

P055 | Wastage of blood and blood components in 21 Polish regional blood transfusion centers (BEs) in 2011-2022—retrospective analysis, a pilot study

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Background: Over the past 30 years, numerous procedures have been implemented in the Polish BEs which have markedly contributed to the safety of transfused blood and blood components. Alongside procedures with direct impact on the risk of transmission of infectious agents, there were also procedures which indirectly affected the risk associated with transfusion. One such procedure is discard and wastage of blood and blood components. The procedure covers all areas of the BE performance related to blood component preparation. Blood component wastage is supervised by the Quality Assurance Department. Each process of blood component preparation and storage is burdened with the risk of all sorts of errors, which may lead to discard and wastage of a blood component. Despite the implemented procedures and quality monitoring, errors sometimes do occur and eliminate donations from further processing. The task of the BE is to monitor blood

component wastage and regulat analysis of the causes which not only allows to respond immediately with corrective actions, but also prevents the occurrence of such problems in the future. BE inspections by the Institute of Hematology and Transfusion Medicine (IHTM) revealed that systematic analysis of wastage is not always properly performed.

Aims: The study aim was a retrospective analysis of discard and wastage of blood components in 21 Polish BEs in the period 2011-2022.

Methods: The analysis of wastage was based on the data from reports of 21 BEs forwarded annually to IHTM. Pilot data on the number of blood components prepared and wasted (whole blood, red blood cell concentrate, platelet cell concentrate and fresh frozen plasma) and the causes of their wastage were subjected to statistical analysis using Microsoft Excel and the Python programming language. Tables and graphs were generated, and the pilot analysis of wastage was performed.

Results: In the period under analysis, in 21 BEs, the total percentage of wastage for all types of blood components increased between 2011 and 2013 (from 4.74 to 5.46%), and then decreased gradually to 3.31% in 2022. The lowest percentage of wastage was for whole blood (from 1.57% in 2012 to 0.99% in 2022). The percentage of wastage for RBCs was 4.20% in 2011 and 2.53% in 2022. The highest percentage of FFP wastage was in 2013–29.23% and 2015–28.82%. The percentage of wastage for both apheresis PC and pooled PC decreased (from 3.53 to 3.09% and from 9.08 to 4.03%, respectively), however, no clear trend was observed over the analyzed period. WB was mainly wasted due to incorrect volume during donation. The highest percentage of RBCs and PC was wasted due to expiry while the main reason for FFP wastage was incorrect visual control. A significant percentage of all blood components was wasted due to reasons marked as “other”.

Summary / Conclusions: The implementation of numerous corrective measures translated into a downward trend for WB and RBC. Cause for concern is lack of trend for the wastage of PC and a high percentage of wastage due to unspecified “other” reasons. Data on the wastage of all blood components require a detailed, in-depth analysis to identify the causes. Such analysis would help identify the critical stages in the processes of blood component preparation and storage which could be the starting point for suggestions addressed to the management of the organizational units of the public blood transfusion service. This may help to improve the functioning of the quality assurance system and cut the costs of utilization.

P056 | Short-term effectiveness of a patient blood management program in patients undergoing radical cystectomy (2017–2023)—an observational cohort study

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Background: Around 50% of patients undergoing radical cystectomy (RC) present anemia and approximately 67% receive a blood transfusion during the perioperative phase. Preoperative anemia and blood

transfusions are often associated with poor patient outcomes. The Association for the Advancement of Blood & Biotherapies (AABB) recommends implementation of Patient Blood Management (PBM) programs for optimizing hemoglobin levels improving patient safety and outcomes.

Aims: The objective is to evaluate the short-term effectiveness of a PBM program on patients undergoing RC by analyzing the improvement in hemoglobin levels and the proportion of perioperative blood transfusion.

Methods: Medical records from 229 patients undergoing RC between 2017 and 2023 were reviewed. Demographic data, hemoglobin levels and prevalence of blood transfusions were collected. For statistical analysis, Pearson's chisquare, Welch's test and Student's t test were used; $p < 0.05$ has been considered significant. Statistical tests were carried out using the IBM SPSS Statistics version 28.0.1.1 and Epidat version 3.1 programs.

Results: The average age of patients undergoing RC was 68.64 years. 52 patients (23,96%) received blood transfusion. From the 206 patients with available laboratory data and who were evaluated in anesthesia consultation, 113 patients (54,85%) had anemia. Preoperative anemia (assessed within the 72 hours prior to the surgical procedure) was found to be correlated with blood transfusions (OR = 17.33, 95% CI 5.95–50.5). Patients over 74 years of age presented preoperative anemia in a significantly higher proportion (76.92%) in comparison to the general population (53.88%). Among patients with anemia, evaluation and treatment in PBM consultation constituted a protective factor for blood transfusions (OR = 0.31, 95% CI 0.13–0.78) and led to a higher improvement in hemoglobin levels: 1.26 g/dL (95% CI % 0.57–1.95) versus 0.22 (95% CI -0.11–0.55). No significant differences have been found in the preoperative hemoglobin levels between the two groups, probably due to selection bias.

Summary / Conclusions: Preoperative anemia is a risk factor for blood transfusion and can be modifiable through optimization programs. Any patient with anemia should be optimized, but greater attention must be required in elderly patients. The PBM program implemented was found to be effective for anemia treatment and reduction of blood transfusion.

P057 | Restricted platelet transfusion for intractable gastrointestinal bleedings

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Background: Apheresis platelet is precious for preventing spontaneous hemorrhage or stop bleeding. Target platelet concentration ($50 \sim 100 \times 10^3/\text{cumm}$) orientated platelet transfusion is recommended for managing acute bleeding events. To transfuse platelets effectively and efficiently, the massive transfusion protocol (MTP) is issued for processing traumatic acute bleeding in the emergency room. Several attempts were also made to extend the MTP concept for other acute bleedings of inpatients with gastrointestinal (GI) bleeding, postpartum hemorrhage, and so forth. Some severe GI

bleeding was however intractable due to unavailable or unsuccessful preceding vasoconstriction procedures. Platelet transfusions are barely effective to stop bleeding for cases of intractable GI bleeding. Our hospital therefore initiated the single unit policy to avoid excessive utilization of apheresis platelets while managing intractable GI bleeding inpatients since October 2020.

Aims: Although no complain or medical legal issues after initiating the single unit apheresis transfusion policy for intractable GI bleeding patients, its clinical impact was closely followed. This study further reviewed and presented several parameters of clinical presentation to demonstrate appropriateness of the policy.

Methods: A retrospective before and after analysis was conducted using the Chang Gung Medical Research Database to assess the clinical impacts of the single-unit transfusion policy on patients with gastrointestinal bleeding due to either peptic ulcer or esophageal varices rupture. The study compared clinical outcomes such as the length of hospital stay, changes in blood oxygen saturation and hemoglobin levels, total usage of platelet components, and mortality rate in terms of implementing the single-unit policy.

Results: Length of Hospital Stay: The average lengths of hospital stay for patients with gastrointestinal bleeding were 13.3 and 12.5 days before and after starting the single-unit transfusion policy, respectively. Blood Oxygen Saturation and Hemoglobin Levels: Occurrences of low blood oxygen saturation (<95%) or hemoglobin (<7 gm/dL) events represented critical conditions of those intractable GI bleeding patients. Before and after the policy, occurrences of low O₂ saturation were 0.068% and 0.063%; occurrences of low hemoglobin were 24% and 22%, respectively. Total Usage of Platelets: After initiating the policy, the average units of platelet transfusions per admission decreased from 8.07 to 2.87. Mortality Rate: The mortality rate of patients after starting the policy was 48%, and the mortality rate of patients before starting the policy was 69%.

Summary / Conclusions: In summary, the single-unit apheresis platelet transfusion policy for patients with intractable GI bleeding demonstrated no adverse clinical impact. Our finding may provide support for extending the MTP concept for non-traumatic bleeding of inpatients.

P058 | Abstract withdrawn

P059 | Blood reservation patterns for live donor liver transplants at a quaternary care setting in South India

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Background: Blood Centre play a vital role in major surgeries like liver transplantations, where massive blood loss is anticipated due to various factors associated with liver dysfunction. The preoperative assessment of blood requirements is often an overassumption thus overburdening the blood centre personnel, & depletion of blood

centre resources, and wastage of time. Over the past few years, higher number of blood units are reserved for transplant surgeries inspite of the advancements in anesthetic and surgical techniques.

Aims: Our primary goal was to audit the blood utilization in liver transplant surgeries in our hospital over a 5-year period and review the blood ordering schedule(BOS).

Methods: A retrospective analysis of adult patients who underwent liver transplant surgeries over a period of 5 years from 2019 to 2023 at our centre was done. All the pediatric liver transplants, cadaveric and combined transplants, retransplant surgeries were excluded from the study. At our centre, current blood reservation pattern include reserving 12 units of LDPRC, 12 units of FFP, 4 units of SDP, 4 pooled cryoprecipitate. The data was analyzed for average consumption of all blood components along with the transfusion indices (C/T Ratio, transfusion index and transfusion probability) and expressed as mean \pm SD or median (interquartile range) as appropriate.

Results: A total of 753 live adult transplants were analyzed in the study period. Mean utilization introspectively along with range were: LDPRC 4.23 ± 3.5 (0-30); FFP 2.43 ± 2.2 (0-24); SDP 0.4 ± 0.7 (0-6) and cryoprecipitate 6.6 ± 6.4 (0-36). 100 patients did not require transfusions at all, indicating the transfusion probability being 86.7% (653/753). In transfused patients C/T ratio had a mean of 3.9 ± 3.12 (1-12), transfusion index 0.4 ± 0.24 (0.08-1.3); Out of 9114 units cross matched, 65% of the total cross matched units were not transfused leading to wastage of resources and working hours for the staff.

Summary / Conclusions: The current blood ordering schedule can be safely brought down in a staged manner keeping in mind the specific needs of unexpected scenarios in coherence and communication with liver transplant team with a balanced blood component inventory to manage the unforeseen situations.

P060 | Is it possible to foresee the usage of red blood cells in coming years?

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Background: More than 80% of red blood cells (RBC) are transfused to patients elder than sixty. In Western Europe this population is growing fast whereas the population eligible for blood donation is decreasing. Tools for forecasting the need for RBC and thereby the need for blood donors in due time may be helpful for regional and national transfusion services.

Aims: The aim has been to look at an earlier prediction model for the usage of RBC, and to pinpoint which parameters are necessary for optimizing the model.

Methods: Fifteen years ago we tried to predict the need for RBC in 2028 (Transfusion 2009;S3:39A) taking into account the development in the national demography. Today the usage of RBC in 2023 is known. As in 2009 the usage of RBC can be compared with that of other Western European countries published by Council of Europe

and other sources. National differences among regions are known from an annual report by the competent authorities.

Results: Due to the demographics a 52% increase of blood usage in 2028 was projected in 2009. As the blood usage varied between 30 and 70 units per 1000 inhabitants among countries, and as our country was the top-user, this prediction was frightening, since the eligible donor population would decrease in the same period with 3% and have to provide more than 100 units per 1000 inhabitants. However, the predicted increase in usage of RBC was not seen. Instead, the consumption was more than halved. In Western Europe the interval in 2023 was reduced to 15 to 40 units per 1000 inhabitants. Thus, most countries have seen a considerable reduction in RBC usage. Most countries are now in the range 30-35 units per 1000 inhabitants although a considerable number are in the range of 20-25. Among the five Danish Regions, the range is 26-32.

Summary / Conclusions: The development has not been consistent with the prediction made in 2009 for 2028. Instead of a RBC usage of more than 100 units per 1000 inhabitants, we have seen 26 per 1000 inhabitants, or only a quarter. Clearly, demographic data is not adequate to foresee the demand. Introduction of evidence based transfusion medicine, Patient Blood Management combined with modern operation technology and medicinal treatments all have had impact on the usage of RBC. Further developments such as CAR-T treatment of hematological diseases may further reduce the usage. Therefore, demographic data should be supplemented with trends of usage in different diagnoses groups. Restoring of regional and national databases (Transf Med 2002;12:34) combining blood usage and diagnoses is therefore pivotal for better predictions.

P061 | Abstract withdrawn

P062 | Abstract withdrawn

P063 | Utility of bloodmobiles in blood collection during Covid-19 pandemic

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Background: The blood collection agencies, employ two major logistical modes for the conduct of outdoor blood collection activities—blood donation drives. In the first, the blood collection agency use a permanent structural space or a similar makeshift tentage arrangement for carrying out the blood collection activities. The phlebotomy is performed with the use of, either the foldable blood donor couches or tables with bedding material and upholstery put over it—referred to as routine blood donation drives. In this the team for blood donation as well as all the equipment and material are driven in a vehicle to the site of the blood collection. In the second, the blood collection agencies use heavy duty vehicle incorporating a mini blood donation centre in itself; specifically fixed blood donor couches—referred to as bloodmobiles.

Aims: The aim of the present study was to compare the two logistical modes employed for the conduct of the outdoor blood collection operations by the blood collection agencies during and before the covid-19 pandemic.

Methods: A retrospective analysis of data with respect to the blood collection at the outdoor blood donation drives over a period of 9 months (March–November) in 2020 (referred to as "during") and 2019 (referred to as "before"—the referent data) covid-19 pandemic. The number of drives, blood donations in the outdoor blood donation drives (total outdoor, routine vs. bloodmobiles) was reviewed for the purpose of better operational preparedness in future. Online software by VassarStats was used for statistical comparison.

Results: Overall, there was a decrease of 8.3% and 42.1% in the number of total outdoor blood donation drives and blood donations in the total outdoor voluntary blood donation drives during the pandemic respectively. On a similar trend, but on a higher note, there was a decrease of 19.2% and 46.6% in the number of routine blood donation drives and blood donations in the routine blood donation drives during the pandemic respectively. However, contrary to the expectations, there was an increase of 11.6% in the number of bloodmobiles drives and only a decrease of 5.2% in the number of blood donations in the bloodmobiles drives during the pandemic respectively. On an average there were 24, 13.6 and 10.4 total outdoor, routine and bloodmobiles donation drives per month in 2020 against 26.2, 16.9 and 9.3 in 2019 respectively. There was a trend in the increase bloodmobiles donation drives (43.3% in 2020 vs. 35.6% in 2019), with odds of 1.38 (CI: 0.99-1.9), $p = 0.05$, during the pandemic in comparison to the referent data. On an average there were 1673.6, 1242.1 and 431.5 blood donations in total outdoor, routine and bloodmobiles donation drives per month in 2020 against 2889, 2331.8 and 577.2 in 2019 respectively. There was a significant increase in contribution in terms of blood donations from bloodmobiles donation drives (25.8% in 2020 vs. 19.5% in 2019), with odds of 1.43 (CI: 1.4-1.5), $p < 0.05$ during the pandemic in comparison to the referent data.

Summary / Conclusions: The bloodmobiles compliment the public outreach efforts of the blood collection agencies during the covid-19 pandemic, both, in terms of the organization of the outdoor blood donation drives and the blood donations obtained thereof. The bloodmobiles are effective vehicles in bridging the gap in the blood collection supply chain logistics during pandemic of respiratory origin, such as the covid-19 pandemic.

P064 | Abstract withdrawn

P065 | The five-year descriptive analysis of blood demand, reserved and unserved in a regional blood center in the Philippines

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Background: Blood transfusion is a life-saving intervention that has an essential role in the total patient management within the health care delivery system, Dophu et.al. The ability to provide the needed

blood to transfuse requires initiatives for the target number of blood donated obtained through ample recruitment of donors. Moreover, the need to serve patients and cater with the needed blood component is part of the blood banking services.

Aims: General: This study was done to generally assess blood demand of a Regional Blood Center in the Philippines. Specific: (1). It specifically aimed to determine the most requested blood component, specific blood type requested per component and type of request (i.e. per demand, blood stocks, partner agencies or for sharing) over the recent 2 year data (2022 to 2023). (2). In addition to the demand or requested blood, the study aimed to show the number with the percentage of reserved and unserved units over the past 6 years (2019 to 2023).

Methods: A comprehensive descriptive analysis of blood demand was performed including the specific blood component, blood type, type of request (i.e. per demand, blood stocks, fellow agencies or sharing). In parallel, with the use of a newly created dashboard that provided a monthly and yearly analysis of the blood demand in this regional center.

Results: The study referred to all blood requests received from 2019 to 2023 that spanned over 5 years within the Philippine Blood Center. The researchers determined that a total of 304,662 blood requests were received over the past 5 years. The researchers noted from the general data, the important impact of the COVID pandemic in relation with the number blood requests received from the trend of blood requests. In addition, due to the creation of a detailed dashboard that started last March 2022 a 2-year data (from 2022 to 2023) showed a more specific analysis of blood requests received that totaled to 103,438 blood requests. The most requested blood unit was the packed red blood cells (PRBC) at 58,163 or 57.9% of the total blood requests over the past 2 years, while this is followed by the platelet concentrate (PC) at 30,938 or 30.6%. The remaining blood requests were for the Fresh frozen plasma (8%) and cryoprecipitate (3.5%). The specific blood type that is most requested was the type O Rh-positive blood group. Likewise for the Rh-negative blood, the most requested was the O blood group. Overall, the type of blood requests received were used as blood stocks with a total of 63.9% for the past 2 years.

Summary / Conclusions: The results obtained from our descriptive analysis will help to further optimize the recruitment of blood donors and the timing of blood donation. This will entail the scheduling of blood drives within the blood center and outside with the aid the available mobile blood donation unit.

P066 | Extending red cell shelf life from 35 to 42 days—would waste reduce?

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Background: Blood services may face shortage of red cells at a blood group or overall level caused by factors influencing donation; hospital demand, or both. At times of stock pressure, blood services will consider a range of interventions at both the donor, supply, and hospital,

demand, ends of the supply chain. One such intervention is to consider extending shelf life as a possible means to minimise waste by the blood service and hospitals. In the UK, the shelf life of red blood cells is 35 days.

Aims: To consider the value, or otherwise, of extending shelf life of red blood cells from 35 to 42 days at times of shortage as a means to reduce wastage through increased time to issue and use red cells for transfusion. To identify the operational actions required to move to 42 day shelf life as a contingency to low stocks and how quickly this could be achieved. To identify the risks and any mitigations

Methods: NHS Blood and Transplant (NHSBT) supply and stock management specialists investigated practical and regulatory considerations for the blood service and hospital transfusion teams of moving from 35 to 42 days shelf life for red blood cells. The group also reviewed data evidencing the age of stock at issue by the blood service and hospital wastage, volumes and causes, at times of shortage. In addition, a previously published ISBT study (Yazer et al 2015) was considered in light of the data.

Results: NHSBT red cell wastage reduces during periods of lower stock levels as the relative age of the stock is reduced. Units are issued fresher and far fewer expire within NHSBT possession, thus the extension would have little impact on NHSBT wastage. Hospital red cell wastage is lower when NHSBT stock is reduced as stock is issued fresher, giving hospitals longer to use the units, again with little impact on wastage. NHSBT encourages hospitals to reduce stock holding, within safe levels, during shortage which reduces the time units are stored before use. Hospital time expiry wastage is most influenced by 'institutional' and operational factors rather than shelf life, with units likely to expire just as often even with an extension. A number of problematic operational issues both with the blood service and hospitals would be required to be overcome and there was insufficient confidence that all issues could be overcome quickly or effectively.

Summary / Conclusions: At times of stock shortage, red cells are both issued and used early in their shelf life meaning extension to shelf life is unlikely to benefit supply through waste reduction. Hospital wastage is most often caused by factors that are not contingent on shelf life. There is insufficient benefit, when weighed against the complexity of implementation, to move to 42-day expiry for red blood cells as a waste minimisation intervention.

P067 | Abstract withdrawn

P068 | The efficiency of blood product utilization in one referral hospital in Indonesia

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Background: Evaluation of blood product utilization is very important to be done continuously. In addition to evaluating the rational use of blood, it is also very helpful in evaluating the service efficiency and

preparing products in sufficient, safe and timely quantities. Prof. Dr. I.-G.N.G. Ngoerah Hospital is one of the referral hospitals in Indonesia for Bali, West Nusa Tenggara and East Nusa Tenggara. Besides being a referral hospital, Prof. Dr. I.G.N.G. Ngoerah hospital is also a teaching hospital. Research on the efficiency of blood product utilization has never been conducted.

Aims: Evaluation of the utilization of blood products at the Prof. Dr. I.-G.N.G. Ngoerah Hospital.

Methods: The research was conducted at the Blood Transfusion Service of Prof. Dr. I.G.N.G. Ngoerah Hospital in January 2024 by taking data reports from the Blood Service Management during 2023. The data was then analyzed using various formulas to obtain the Transfusion Probability (%T), Transfusion Index (TI), Crossmatch to Transfusion Ratio (C/T Ratio), Maximum Surgical Blood Order Schedule (MSBOS) and Wastage Rate (WR).

Results: The evaluation results during 2023 showed an average Transfusion Probability (%T) of 83.97% (target >30%). The overall Transfusion Index (TI) was 2.52 (target >0.5), specifically the Transfusion Index of the Surgical Department reached 1.57. The average monthly Crossmatch to Transfusion (C/T) ratio was good at 1.19 (target <2). From the crossmatch data, the number of bags that were cross-matched but not used most (53%) came from the Department of Surgery, 21.2% from the Department of Internal Medicine, 10.1% from the Department of Pediatrics, 9.7% from the Department of Obstetrics and Gynecology, and 6% from other Departments. The Maximum Surgical Blood Order Schedule rate obtained for all surgical procedures with bleeding risk was 2.38. The average waste rate (WR) of PRC was 2.52 (target 2-3%), TC was 7.68 (target 12-18%), FFP was 10.08 (target 10%), and cryoprecipitate was 5.13. Most blood products destroyed were TC components, followed by PRC and FFP. The cause of most blood product destruction was expired products, followed by PRC manufacturing remnants with pediatric bags and bag tubing that had run out and could not be used for crossmatch.

Summary / Conclusions: Blood product utilization efficiency based on Transfusion Probability (%T), Transfusion Index (TI), and Crossmatch to Transfusion ratio (C/T ratio) met established targets. The Maximum Surgical Blood Order Schedule (MSBOS) rate for bleeding risk procedures is 2.38. The waste rate for PRC and TC products has met the target. Further evaluation and innovation is needed to reduce the WR of FFP components that did not meet the target.

P069 | Functional assessment of lyophilized sterilized platelet concentrate

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Background: Storage of platelets (PLTs) remains a major problem for blood banks. In this study we describe the development of a lyophilization protocol for platelet concentrates obtained from a blood bank.

Aims: We report here that platelets can be preserved by freeze-drying them with trehalose, a sugar found at high concentrations in organisms that naturally survive drying in comparison with sucrose commonly used for proteins lyophilization

Methods: In both human fresh and lyophilized PLTs, aggregation response and PLT surface marker P selectin, were measured. To evaluate the efficacy of gamma irradiation as Pathogen Inactivation (PI) method, the reduction factor (RF) of titer of model viruses and bacteria were examined.

Results: Trehalose and sucrose-loaded platelets were freeze-dried. The response of these platelets to the agonists thrombin (1 U/mL), collagen (2 µg/mL), and ADP (20 µM) was evaluated. All trehalose, sucrose and trehalose/sucrose groups had decreased in thrombin, collagen and ADP agonists aggregation but this reduction for trehalose 60 mM was not statistically significant and this group had function about 40% for these agonists. Also, the CD62p expression for trehalose group was similar to fresh PRP and was significantly lower than sucrose and trehalose/sucrose groups. Freeze-drying reduced 2 logs the HSV and Polio viruses titration. Moreover, the dose of 30 KGy and 40 KGy gamma irradiation demonstrated a favorable effect on lyophilized PLTs.

Summary / Conclusions: Our in vitro characterization studies showed maintenance of hemostatic functions of lyophilized and sterilized PLTs. In vivo studies are needed to determine the survival and function of lyophilized PLTs.

P070 | A study on whole blood and blood products usage and wastage in a tertiary hospital in Cebu, Philippines

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Background: About 112.5 million blood donations are collected worldwide. Blood products contribute to the saving of millions of lives every year, improve dramatically life expectancy and the quality of life of patients suffering from life-threatening conditions, and support complex medical and surgical procedures. In many countries, demand outstrips supply, and blood services throughout the world face the daunting challenge of making sufficient supplies of blood products available, while also ensuring the quality and safety of these products in the face of known and emerging threats to public health. The issues of sufficiency, availability, and access cannot be considered in isolation from the use of blood. Blood wastage may occur for several reasons, including time expiry, wasted imports, blood medically or surgically ordered but not used, stock time expired, hemolysis, or miscellaneous reasons.

Aims: The study aims to determine the usage and wastage of whole blood and blood products in a tertiary hospital in Cebu, Philippines from January 2018 to December 2018.

Methods: The research utilized a retrospective, cross-sectional descriptive study design.

Results: The months of April, August, and September 2018 portray peaks of units collected, well beyond 300 units. Type O+ was the

most frequently collected blood type. The second most collected blood group was A+, followed by B+. The least collected blood group in a tertiary hospital in Cebu, Philippines was AB+. Packed red blood cells (76.14%) were the most transfused units, followed by fresh frozen plasma (8.17%). The third most transfused products were platelets, utilizing 6.80% of units administered to patients. Similar percentages in consumption were whole blood units and cryoprecipitate, with roughly 4% each. There were 488 blood units wasted during the study period. The most common cause of wastage was due to the expiry of units, mostly platelets, followed by whole blood. Blood units reactive for transfusion transmissible infection (TTI) were the next noted cause for wastage, while damaged units were the least. For reactive units, Hepatitis B surface antigen (HBsAg) was the most common cause of wastage. Hepatitis C virus reactive units were second while Syphilis and Human Immunodeficiency Virus (HIV) reactive units followed, with around 15% and 7%, respectively.

Summary / Conclusions: The most common blood type collected in a tertiary hospital in Cebu Philippines is Type O+ (45.6%), while Type AB+ (5%) is the least collected. Packed red blood cells (76.1%) are the most transfused blood component in the institution. The most common cause of wastage is due to expired blood units (70%), most of which are platelet concentrate. The next cause for wastage is due to units reactive for transfusion transmissible infection (16%). The least cause of wastage is due to damaged blood units (14%).

P071 | Wastage of blood & blood products—time to re-think

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Background: In recent years, blood has been recognized as a therapeutic medicine of utmost importance. However, in developing nations such as Pakistan, the efficient delivery of healthcare services at reduced costs is a critical factor. Despite facing a shortage of blood donations, a significant portion of donated blood is being disposed of, leading to a widening gap between demand and supply. Blood disposal not only incurs financial losses for healthcare establishments but also poses a potential threat of disease transmission. Consequently, addressing this issue is paramount to ensure both financial stability and public health safety within the healthcare system.

Aims: This study aimed to assess the distinction between blood collection and transfusion, investigate the factors contributing to blood donation disposal, and advocate for the attention of local health regulatory authorities to ensure the safe disposal of blood and its products.

Methods: This retrospective cross-sectional study was conducted at HealthBridge Laboratory & Blood Bank from January 2021 to

December 2022. A total of 11,839 blood donors underwent thorough physical examination, following which 5 mL blood samples (2 mL EDTA and 3 mL Clotted) were collected and processed for Transfusion Transmissible Infections (Anti-HCV, HBsAg, Anti-HIV I & II, Syphilis, and Malarial parasite). Anti-HCV, HBsAg, Anti-HIV I & II, and Syphilis tests were performed using chemiluminescence immunoassay (CLIA), while the malarial parasite test was conducted through Immunochromatographic Technique (ICT). Blood was discarded according to standard operating procedures.

Results: Among the total donors, only 1.7% (207 out of 11,839) tested positive for any Transfusion Transmissible Infections (TTIs), with specific percentages as follows: 0.61% for Anti HCV, 0.50% for HBsAg, 0.008% for Anti-HIV I & II, 0.55% for Syphilis, and 0.05% for malarial parasite. These positive donors were deferred from further blood collection. Subsequently, 3.6% (420 out of 11,632) of the remaining donors provided whole blood, while the remaining 96.4% (11,212 out of 11,632) underwent blood component preparation. Storage of all blood and blood products was maintained under standard and controlled conditions. Discards included 8.57% of whole blood, primarily due to residual blood in the bag post-transfusion, 6.86% of Packed Cell Volume (PCV) due to patient vital derangements during transfusion or late return, and 0.95% of Fresh Frozen Plasma (FFP) due to red blood cell contamination. Platelets concentrate and cryoprecipitate were not discarded as they are promptly transfused after collection.

Summary / Conclusions: positivity rates, but a new challenge of blood wastage has emerged. Despite a low whole blood transfusion rate in our setting, blood and blood products are still being discarded, leading to significant financial losses for healthcare establishments and subsequently increasing healthcare service costs. Urgent attention from health regulatory authorities is warranted, as aside from financial implications, this situation may also pose risks of disease transmission within the general population. Addressing this issue requires the establishment of a centralized system for blood collection from blood banks over time, allowing for efficient utilization as needed.

P072 | Save blood, safe surgery—strategies for intraoperative transfusion safety by the power of electronic transfusion monitoring

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Background: Integration of Transfusion Safety Systems (TSS) marks a significant advancement in transfusion medicine, providing a comprehensive approach to monitoring the transfusion process using computerized systems. These systems complement clinical practices by enabling real-time tracking, documentation, and verification of essential transfusion-related data. Their integration into healthcare workflows enhances accuracy, efficiency, and transparency while mitigating risks associated with human error, thereby promoting patient safety.

P072 Table 1

OP.THEATER	CHECK-LIST	INITATE	CLOSE	% IN/CL
Traumato	755	115	79	69
General surgery	539	389	260	67
Cardiac	422	603	582	67
Urology	379	41	36	87
Urgency	311	163	129	79
Vascular	295	49	40	81
Thorax	260	49	37	75
Neuro & maxilo & plastic	363	56	33	60
Total	3.324	1.465	1.196	73%

Aims: Evaluate the implementation of an existing transfusion safety system in a high-complexity hospital: given the substantial volume and complexity of surgical procedures performed, including multi-organ transplants such as hepatic, cardiac, pulmonary, and renal transplants. To provide context, in 2023 the hospital conducted 11,378 scheduled surgical procedures (SP), 5362 urgent SP and 3801 deliveries: 21.65% caesarean. This study underscores the importance of having a robust transfusion safety system in place to manage the diverse transfusion needs arising from complex surgeries.

Methods: We utilized the transfusion safety system developed by Jub-solutions, company located in Barcelona, Spain. TSS used: HemoCod®. Transfusion activity is monitored through the unique association of each hardware device (PDA) in every operating room. Jub-solutions facilitated the exportation of these data. Transfusion in the operating room occurred in two ways: either through the HemoCod® bracelet worn by the patient or by associating the patient with the HemoCod® bracelet to a known operating room number. In the latter case, if a transfusion was needed, it was carried out using an additional label representing the bracelet number with a barcode. The analysis includes 14 operating rooms from January 2023, except for the gynaecological-obstetric and paediatric sections are currently undergoing implementation. Statistical analyses presented in terms of means and percentages.

Results: In numbers, we recorded 3457 patient-to-operating room associations (check-list): transfusion initiation began with HemoCod via bracelet in 490 cases and with an additional barcode label in 975 cases, total 1.465. The closure of these transfusions, 328 finalized with the bracelet and 876 with the additional label: total 1.196. The overall percentage of initiation/closure stands at the average 73%. Detail activity as follows:

Summary / Conclusions: An electronic TSS like HemoCod®, allows for continuous patient identification within the operating room, ensuring traceability of blood units and monitoring intraoperative activity to drive improvement actions. Furthermore, it enables analysis of transfusion frequency and the preferential use of association check list, that potentially could reduce the need for routine requests that do not culminate in transfusions. The remarkable figure of 3.324 patient-

to-operating room associations, with nearly half initiating transfusions: 1.465, underscores the potential for resource optimization. It's worth to highlight the substantial closure rate of 73% that contribute significantly to monitor transfusion follow-ups in surgery. This comprehensive approach not only enhances patient safety but also optimizes resource utilization and quality of care within the surgical setting.

P073 | Abstract withdrawn

P074 | Efficiency of red cell unit delivery by pneumatic tube system

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Background: Transportation of blood components from the point of issue to the site of transfusion needs to be safe and efficient. This study was conducted at a transfusion service (hospital blood bank) at a large tertiary care hospital. The hospital pneumatic tube system (PTS) is validated to be used for the transportation of blood components. In our setting, delivery by PTS has been shown to significantly reduce the staff resources needed for pickup and delivery of blood components. Per year, approximately 20,000 red cell (RBC) units are transfused at the hospital. Approximately one third of the RBC units are delivered by PTS. The remaining RBCs are picked up and delivered by hospital staff (HS).

Aims: The aim of this study was to evaluate the delivery time of RBC units according to whether the unit was delivered by HS or PTS.

Methods: For the calendar year 2018, data on time of RBC unit issue (TI) and time of initiation of transfusion (TT) were retrieved from the blood establishment computer system (ProSang). Delivery time (DT) was defined as TT-TI and reported in whole minutes. Data were excluded from analysis in the following situations: RBC units issued to operating rooms, ICUs or hospital wards using a remote RBC refrigerator, DT >59 min, simultaneous issue (except for the primary unit) or manual registration of TT. Data on DT across groups were compared using the Mann-Whitney U test.

Results: Delivery time data followed a log-normal distribution. During 2018, a total of 19,413 RBC units were issued. Data from 1583/19,413 (8.2%) units, 941/1583 (59%) by HS and 642 by PTS were included in the study. Mean TT was 17.7 min and 15.3 min for HS and PTS, respectively (90-percentiles: 31 and 27 min, respectively). Mean TT difference was (HS - PTS) 2 min 29 sec (14.0%; $p < 0.0001$).

Summary / Conclusions: In our setting, there was a small but highly statistically significant advantage in delivery by pneumatic tube system, this delivery mode being on average 2 min. 29 s. (14%; $p < 0001$) faster with regard to initiation of transfusion compared to pickup and delivery by hospital staff.

P075 | Availability of sufficient and quality blood services in COVID-19 pandemic during smart lockdown

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Background: Since the inception of blood transfusion services worldwide, the availability of voluntary blood donation (VBD) has been crucial for ensuring a healthy blood supply. In Pakistan, the frequency of VBD is notably low, a challenge exacerbated during the recent implementation of smart lockdown measures.

Aims: This aims of study included; Assessing the Blood Availability During Lockdown. Examining the Difficulties Encountered in Blood Donation Collection During the Implementation of Smart Lockdown in Pakistan.

Methods: The cross-sectional study took place at HealthBridge Laboratory & Blood Bank in Lahore from February 2021 to September 2021. Various factors were compared between the study duration and the period preceding the study.

Results: During the study period, a notable reduction of 9.7% in voluntary blood donation (VBD) was observed, juxtaposed with a significant increase in the demand for blood and blood components by 20%. Additionally, there was a significant decrease in the frequency of female donors. To address the heightened demand for blood, various approaches were implemented. These included obtaining 2751 blood donations through telephonic communication (18%), WhatsApp text messaging (24%), blood collection from home/office settings (19%), and blood collection from prisons (39%). These strategies were adopted to effectively manage the situation and ensure a sufficient blood supply.

Summary / Conclusions: The pandemic precipitated a severe shortage of blood, prompting the implementation of various innovative approaches to mitigate the deficit. It is imperative that these strategies be incorporated into routine practices to consistently augment the blood supply.

Management and organisation—quality management

P076 | The Lebanese Red Cross Blood Services roadmap to AABB quality certification

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Background: The Lebanese Red Cross (LRC) Blood Services a non-profit organization and member of the Red Cross and Red Crescent Movement, is mandated by the Ministry of Health in Lebanon to provide safe blood products to hospitals. Operating through 13 blood centers for the entire country, the organization faced a decline in service quality post-civil war. In response to a Ministry of Health warning in 2015, the LRC Blood Services underwent a comprehensive reorganization, establishing a quality management system, centralizing activities, and striving to meet national and international standards.

Aims: In 2018, the LRC Blood Services, committed to following AABB standards, made a concerted effort to attain accreditation from the AABB.

Methods: Despite years of dedicated work, the accreditation posed a challenge due to budget constraints preventing the implementation of Nucleic Acid Testing (NAT) for infectious diseases, a requirement for accreditation. In light of this limitation, AABB proposed an alternative solution to LRC, the AABB Quality Certificate Program. This program is a viable substitute to accreditation in our current context and achievement of the certificate is international recognition of the high standards maintained by LRC Blood Services. After procuring the “AABB Fundamental Standards for Blood Collection and Transfusion (Fundamentals)”, which are the foundation of the certificate program, the team embarked on rigorous preparations for implementing the standards and performing the self-assessment. Amidst adversities such as the COVID-19 pandemic, the riots, the huge influx of refugees, the Beirut port explosion, and Lebanon's financial crisis, the organization persisted in its efforts of centralizing testing activities, acquiring, and qualifying new equipment, training both existing and new personnel, and establishing robust internal and external quality control systems under the guidance of the “Fundamentals”.

Results: In November 2021, a week-long inspection visit was conducted and led to the issuance of the accreditation certificate in February 2022 for a period of 2 years.

Summary / Conclusions: This achievement marked the first-ever blood bank certification by an internationally recognized organization in Lebanon, fostering increased trust from partner hospitals and the community at large.

P077 | Investigating the quality of blood products from Canadian blood donors with diabetes

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Background: As of March 2021, individuals taking insulin to manage type 1 diabetes (T1D) are eligible to donate whole blood through Canadian Blood Services (CBS). This followed earlier policy changes allowing most individuals with type 2 diabetes (T2D) to donate. While recent studies support that donation is safe for individuals with diabetes, few have evaluated the quality of their donated blood components. Importantly, there is evidence that poor glycemic control can impact platelet function, red blood cell characteristics, and plasma contents—all of which may impact transfusion recipient outcomes. To ensure safe storage and utilization, and to inform other organizations about the implications of expanding donor eligibility criteria, there is a need to better understand the properties of blood products from donors with diabetes in Canada.

Aims: 1) Screen glycemic control in CBS donors with T1D and T2D, 2) Characterize blood products—red cell concentrates and single-donor buffy coat platelets—from donors with diabetes, throughout storage.

Methods: (1) CBS donors are asked whether they have diabetes at the time of donation, and this metadata was used to flag a subset of donors with T1D, T2D, or without self-reported diabetes. Following routine donations across Canada, CBS national testing centers shipped one tube of EDTA whole blood from flagged donors for clinical HbA1c testing. (2) Whole blood donations from donors with or without self-reported diabetes were separated into red cell concentrate (RCC), single-donor buffy coat platelet concentrate (PC, stored in plasma), and fresh frozen plasma. RCC was assessed bi-weekly over 42 days for storage hemolysis, oxidative hemolysis, ektacytometry (deformability and osmotic gradient), hematological indices, and blood gas indices. PC was assessed on days 1, 4, and 7 with CD62P flow cytometry (platelet activation and responsiveness to ADP), rotational thromboelastometry (intrinsic and extrinsic activation), hematological indices, and blood gas indices.

Results: (1) To date, $N = 200$ (71 T1D, 86 T2D, 43 without diabetes) specimens have been screened for HbA1c. The mean HbA1c for donors with T1D was 7.22%, T2D was 7.25%, and without diabetes was 5.50%. Of all donors with diabetes, 53% had an HbA1c over the Diabetes Canada target of 7.00%. (2) Initial data from aim 2 ($N = 43$; 2 T1D, 6 T2D, 34 without diabetes) do not suggest major clinically relevant differences in RCC or PC from donors with diabetes, though there are some notable trends. Red cell hydration (reflected by Ohyper in osmotic gradient ektacytometry) and storage hemolysis trend higher in donors with diabetes, especially donors with T2D. In PC from donors with diabetes, maximum clot firmness in extrinsically activated rotational thromboelastometry trends higher, and clot formation time trends lower in donors with T1D. A trend toward lower

storage pH was also observed in PC from donors with diabetes, though this is likely attributed to higher platelet concentrations.

Summary / Conclusions: Suboptimal glycemic control in the majority of CBS donors with diabetes could be a cause for concern for the quality of their resulting blood products. The ability to draw robust conclusions about product quality from preliminary data in aim 2 is currently limited by the current small sample size and very good average HbA1c of the donors with diabetes to date (6.26%). Data collection for both aims is ongoing, with plans for -omics analyses on banked specimens from aim 2.

P078 | Abstract withdrawn

P079 | Effectiveness of implementation of 2nd distinct Type and Screen (TS) confirmatory sample for patients with no historical record of ABO/Rh a single hospital experience

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Background: Based on data from incorrect blood components transfused and from Serious Hazards of Transfusion (SHOT) from 1996 to 2010, the most common cause of transfusion error is wrong blood in tube (WBIT) reported as near misses in 2010. WBIT can be detected based on discrepancies from historical records in the Blood Bank (BB). Transfusion of the wrong blood group or blood that has clinically significant antibodies can lead to detrimental clinical consequences. The '2nd-confirmatory Type and Screen (TS) sample method' (SCS) can potentially eliminate the risk of WBIT

Aims: We performed a retrospective analysis, looking back at 2 years of clinician adaptation and the implementation of 2nd TS sample workflow for patients with no historical result by leveraging on the hospital IT system

Methods: Upon receiving a TS from a patient, BB technologists will check the hospital Blood Bank system for any prior ABO/Rh, and antibody screen. If there was no historical result, the clinician will be contacted via Hospital Text Messaging System to send a 2nd sample for blood group confirmation. The BB technologists will add a comment to the ABO/Rh results of the first sample indicating the need for a 2nd sample. Should the clinicians request for an urgent blood transfusion without a 2nd sample, BB will issue universal 'O' cross-matched red cells. Both samples must be drawn independently. Blood samples from simultaneous sampling will be rejected

Results: SCS sent were consistent in both years. 51% of the SCS sent required blood transfusion and the remaining 49% with no SCS did not require transfusion. The compliance rate for sending SCS improved from 35.1 to 63% in patients needing transfusion from

P079 Table A: Number of 2nd confirmation sample sent against total number of Type and Screen sent that has no historical ABO/Rh

Criteria	2022*	2023	Total
Total TS sent	902	1235	2137
TS with no historical ABO/Rh (%)	280 (31.0)	365 (29.6)	645 (30.2)
2nd sample was sent (%)	111 (39.6)	146 (40.0)	257 (39.8)
Transfusion required (%)	39 (35.1)	92 (63.0)	131 (51.0)
Transfusion not required (%)	72 (64.9)	54 (37.0)	126 (49.0)
2nd sample not sent (%)	169 (60.4)	219 (60.0)	388 (60.2)
Transfusion required (%)	3 (1.8)	2 (0.9)	5 (1.3)
Transfusion not required (%)	166 (98.2)	217 (99.1)	393 (98.7)

* Data from Feb 2022 onwards (implementation of SCS).

2022 to 2023. The ratio of 2nd TS sample not sent remained the same when compared across both years. In 2022, 3 Group 'O' red cell units were issued without a 2nd sample due to urgent request for bleeding. There was a reduction in the number of blood requests without a 2nd sample from 2022 to 2023. A total of 1.3% of blood was issued without a 2nd sample over two years.

P079 Table B: Number of blood requests in Alexandra Hospital

Location of blood requests	2022	2023
General Medicine	32	61
Geriatric Medicine	1	12
Intensive Care	1	2
Orthopedics	0	2
Rehab	0	1
Surgery	0	1
Urgent Care Centre	8	15
Total	42	94
Blood requests without 2nd sample	31	22

¹ From General Medicine.

² From UCC.

From Table B, there was significant improvement over the 2 years where elective transfusion orders are only carried out after a 2nd sample. Only 5 cases (6%) of total blood requests over the 2-year period received blood units for transfusion without a 2nd sample. There was no ABO incompatible transfusion reaction reported in this period

Summary / Conclusions: With the aid of our hospital IT platform, the implementation of SCS for patients with no historical ABO/Rh has shown a positive outcome, with satisfactory clinician adherence.

P080 | The practice of jointly external quality assessment of blood stations in the Yangtze River Delta

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Background: The Quality Control Laboratory of the Blood Station is mainly responsible for quality monitoring including whole blood and component blood quality check, key materials, key equipment and environmental hygiene. At present, the External Quality Assessment(EQA)of the National Center for Clinical Laboratories does not fully include all testing items of the quality control laboratories of blood stations, and the quality control laboratories of some Blood stations do not carry out EQA. Therefore, joint multi-provincial and municipal EQA will be conducive to improving the testing level and quality control capability of QC laboratories and ensuring the quality of testing.

Aims: By organizing Blood stations in Shanghai, Jiangsu, Zhejiang and Anhui Provinces to jointly carry out EQA of relevant testing items in blood station quality control laboratories, explore the feasibility of establishing a joint EQA mechanism and test its effectiveness with a view to recommending good practices for blood stations in more parts of the country.

Methods: From 2017 to 2022, the Blood Center of Zhejiang Province, in conjunction with the blood centers of Shanghai, Jiangsu and Anhui Provinces, organized Blood stations in the province to participate in the joint EQA of blood station quality control laboratories, and carried out a regional EQA project based on the model of the "four unities". Firstly, the EQA items and sources are unified, including biochemical items (total plasma protein, methylene blue, free hemoglobin), blood cell counts (RBC, WBC, PLT, Hb, HCT), and hemagglutination (FVII: CFbg), etc., and designated commercialized reagents are used. Secondly, QCs are uniformly distributed and testing times are set at designated times. Thirdly, the uniform return format, each laboratory according to a unified template to report the results to the Zhejiang Blood Center. Finally, the analytical templates are harmonized, and all results from each laboratory are imported into the harmonized template, and mean and SD values are calculated.

Results: A total of 55 blood collection organizations in the Yangtze River Delta participated in the jointly external quality assessment of Quality Control Laboratories from 2017 to 2022. The pass rates for plasma protein testing in each laboratory were 91.11%, 92.78%, 91.67%, 95.41%, 94.29%, and 90.95%, respectively; for Factor VIII testing were 85.00%, 74.36%, 91.67%, 92.22%, 89.51%, and 68.22%, respectively; for Fibrinogen testing were 75.00%, 90.00%, 96.67%, 97.78%, 97.75%, and 97.15%.

Summary / Conclusions: Through several years of operation of the joint interlaboratory quality assessment of blood station quality control laboratories in the Yangtze River Delta, the overall mean values of the quality control laboratories are closer to the target values, there is no great deviation in the results reported by the laboratories, the

distribution of the quality control result data is more reasonable, the results of the laboratories on the duplicated specimens are more consistent, the level of testing of the quality control laboratories is generally improved, and the comparability and stability of the results are significantly improved. This project makes up for the lack of quality evaluation of QC laboratories carried out in individual provinces and cities, and can also be used as an effective supplement to the EQC of National Center for Clinical Laboratories, which has a certain value of popularization and application throughout the country.

P081 | Standardization of blood discarding rate statistics in Zhejiang Province

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Background: Blood stations collect information on blood collection, preparation, testing, storage, and distribution through computer systems. They classify, summarize, and analyze the information on unqualified and discarded blood generated at each stage. Given the preciousness of blood resources, blood stations must minimize blood waste while ensuring the safety and effectiveness of blood quality. However, due to the varying calculation rules for the discarding rate among blood stations in the province, it is difficult to conduct horizontal comparisons, and standardization of discarding rate comparisons is urgently needed.

Aims: Standardize the elements involved in calculating blood discarding rate and establish a uniform calculation rule that is applicable province-wide.

Methods: Established rules to improve the accuracy of discarded blood calculations, including specifying the scope of calculations, defining statistical criteria and time periods, determining appropriate units for measurements, and designating the department responsible for calculating dispatched blood, among other related factors.

Results: (1) The statistical method of blood discarding rate has been determined. In the basic calculation formula of discarding rate, the numerator refers to the blood that has been audited and discarded during the statistical time, while the denominator is the sum of the released qualified blood units and the discarded blood units within the statistical time. Measurement points are determined as the date of blood audit for the numerator and the date of release for qualified and discarded blood for the denominator. The time period is measured monthly or annually. (2) Formulated a standardized conversion rule for measuring blood units. The conversion from milliliters (mL) to units (U) is as follows: whole blood (U) = whole blood (mL) / 200, plasma (U) = plasma (mL) / 100, apheresis platelets (U) = actual unit number of apheresis platelets (u) / 10, pooled platelets (U) = the sum of whole blood units used to prepare platelets / 10. (3) Standardized the cryoprecipitated antihemophilic factor preparation method. Institutions using finished plasma to prepare it should adjust the denominator by adding "cryoprecipitated antihemophilic factor

preparation amount $\times 0.75$ " to compensate for the decrease in raw plasma volume. (4) Dispatched blood will be included in the calculation by the original collection department, and quality inspection wastage will be included in the statistical range of the blood discarding rate. (5) A total of 34 indicators for the discarding rate have been established, including rates for all blood types, as well as for red blood cells, plasma, and platelets. Additional indicators include rates for transfusion-transmitted infections (TTI), non-TTI results, and positive tests for ALT/HBsAg/HCV/HIV/TP/HTLV (by blood volume/sample size) and nucleic acid testing. Other indicators include lipemia, non-standard measurement, blood bag rupture/leakage, risks reported after donation, unexpected antibodies, expired units, abnormal color, hemolysis, blood clot, and other reasons.

Summary / Conclusions: The "Zhejiang Province Blood Discarding Statistics Method" was formulated to define statistical criteria and calculation formulas for standard indicators throughout the province. A comparison mechanism has been established for the province, and all blood collection and supply institutions are required to regularly collect and submit discarding rate indicators according to this unified standard.

P082 | Document management in Blood Establishments (BEs) in 2021—Polish experience

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Background: The implementation of advanced methods of collection, preparation and testing of blood has resulted in high quality and safety standards. Equally important is an effective quality assurance (QA) system which allows for the optimization and monitoring of the processes in BEs. Among others, this is supported by an efficient document management system with emphasis on standard operating procedures (SOPs). The inspection teams of the Institute of Hematology and Transfusion Medicine have observed discrepancies in the numbers of SOPs developed, implemented, coded for different areas of BE performance. The discrepancies also refer to other documents for example, Quality Manual (QM), specifications and range of responsibilities.

Aims: The study aim was to analyze document management at BEs with special emphasis on type of documents, their implementation and supervision.

Methods: Data provided by 23 BEs in response to a survey consisting of 54 questions were analyzed.

Results: The total number of SOPs ranged from < 100 to > 400; most BEs (15) had between 200 and 400. The number of SOPs for documentation ranged from 1 in 7 BEs to 30 in 1 BE. Likewise, a significant discrepancy in the number of SOPs was reported for other areas of BE performance: donor registration 1-12, medical qualification 1-15, collection of blood and blood components 1-21, preparation of blood

components 1-36, distribution 1-34, QA 23-89. In 21 BEs, the QA Department supervises SOPs; in 1 BE—heads of departments and in 1 BE—the head of the QA department and the Director's proxy for QA. 20 BEs follow a three-step procedure for SOP implementation, and 3 BEs follow a four-step procedure. In 18 BEs, lists of employees who acknowledge the SOP are an integral part of the procedure. In 12 BEs, the history of changes was included in SOP. 13 BEs use arbitrarily imposed guidelines on text style and graphics for SOPs. In 9 BEs, approval for development of a new SOP is given by the QA head, and in 1 BE—by the Quality Supervisor. In the remaining BEs, the development of a new SOP is consulted with the QA. In 2 BEs, the original SOPs are kept with the Quality Supervisor, in the remaining BEs with the QA department. In 18 BEs, the QA department is responsible for withdrawal of all out-of-dated SOPs. In 22 BEs, the specification is developed by an employee of the proper department. 22 BEs declared a single scope of duties, responsibilities and authorizations for each employee, taking into account all their functions. 17 BEs have developed organizational procedures for each department. The number of employees involved in preparing QM ranged from 1 (in 2 BEs) to 18. 22 BEs included in the QM the information on staff qualifications. 18 BEs included job descriptions. In 22 BEs, the QM presented a hierarchy of documentation with a description of the types of documents. 12 BEs declared integration of their documentation complying to the Public Blood Service Law with documentation developed under the Pharmaceutical Law. 3 BEs indicated integration with ISO, and 4 BEs declared integration with both Chief Pharmaceutical Inspectorate and ISO.

Summary / Conclusions: The total number of SOPs as well as the different numbers of SOPs in each department, indicate the need for developing an optimal number of SOPs for each BE. Different structure of SOPs and specifications, different ways of coding, writing and implementing the documents for routine use require development and implementation of a single standard procedure in all BEs.

P083 | Accreditation under ISO 15189 standards in the blood transfusion department of the General University Hospital Virgen de las Nieves improves the efficiency throughout the entire laboratory process.

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Background: Currently, the ISO 15189:2022 standard is the quality norm for clinical laboratories, which establishes the requirements to ensure the presence of a Quality Management System (QMS) and acknowledges the laboratory's technical competence. Under the ISO 15189 standard, any QMS ought to ensure that laboratory infrastructure, human resources, responsibilities and laboratory activities, specified in the applied norm, are conducted properly. The Blood Transfusion department of the General University Hospital Virgen de las Nieves adheres to the FCAT standards, which are the most effective and robust tool to guarantee blood transfusions safety and quality in Spain, to develop the

QMS that would be followed. However, to enhance efficiency of the QMS obtaining the ISO 15189:2022 accreditation was recommended.

Aims: To show the results throughout the entire laboratory process (including pre-analytical, analytical, and post-analytical phases) obtained after following the ISO 15189:2022 norm.

Methods: Descriptive research of the accreditation process that includes several phases: project planification (analysis of the baseline situation, the required resources and the responsible team), scope of the accreditation, elaboration of documents (implantation and distribution) including quality assurance processes through internal and external quality controls, training and qualification of laboratory staff, management review, monitoring of indicators and, finally, acknowledgement by the Spanish National Accreditation Body (ENAC). The app e-BDI Plus[®] was installed as an integral solution for the development and maintenance of all activities related to the quality management, and indicators for all the phases of the analytic process were established.

Results: After implementing the QMS according to ISO 15189 in our Blood Transfusion department, several improvements in laboratory performance were observed. In the pre-analytical phase, there was a progressive reduction of the incidents in both immunohematology studies unrelated to blood transfusion and blood compatibility testing. In the analytical phase, we achieved 100% accuracy in internal results and quality controls. Finally, in the post-analytical phase, the response time to request and the percentage of request forms submitted after the deadline decreased.

Summary / Conclusions: Accreditation under the ISO 15189 standard together with FCAT certification has significantly enhanced our QMS in the Blood Transfusion department, allowing us to: Recognize the technical competence of laboratory staff and equipment and guarantee the quality of materials used. Obtain clear traceability throughout the entire process, from the request form to final. Implement a systematic procedure for monitoring result validity, mitigating unwanted impacts and potential failures. Optimize the technical performance and resources management. Increase patient's safety and satisfaction.

P084 | The AABB Quality Certificate Program for blood banks and transfusion services in Low or Middle Income Countries (LMICs) as their first step on the quality journey

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Background: The AABB Fundamental Standards for Blood Collection and Transfusion (Fundamentals) are the foundation for the Quality Certificate Program. These standards were developed on good medical practice and scientific, evidenced-based data. The certificate provides international recognition for advanced laboratory safety and quality efforts. The Fundamentals consist of ten Quality System Essentials (QSEs), Organization, Resources, Equipment, Supplier and Customer, Documents/Records, Deviations/Nonconformances, Assessments (internal and external), Process Improvement, Facilities and Safety.

Aims: In 2020 during the first year of the pandemic, after 8 years of collaboration the AABB (Association for the Advancement of Blood and

Biotherapies) and AATM (Asian Association of Transfusion Medicine) developed the concept of the AABB Quality Certificate Program. The goal was a quality program based on implementation of the AABB Fundamental Standards for blood service and transfusion services in LIMCs. The virtual, successful pilot was completed in 2021 by the Rotary Bangalore TTK Blood Centre, Bangalore Medical Services Trust. A key objective of the certificate program is a pathway to full accreditation.

Methods: The program provides an opportunity for services to examine their processes/procedures against world-renowned standards to promote the highest levels of quality for donors and patients. Another goal of the program is providing an educational experience for all staff. The program consists of a self-assessment of Quality Management System (QMS) and Operations of the service utilizing the AABB self-assessment tool and the self-assessment guide. The program is initiated with a virtual kick-off meeting. Implementation of the Fundamentals is tracked through the submission of documents to the assigned AABB Quality Specialist. The specialist is dedicated to the service, reviews all documentation for accuracy and completeness, and mentors the staff throughout the program. The entire process is approximately 15 weeks, which gives flexibility to the service and allows time for everyone to participate.

Results: During the certificate program, the staff learn the Fundamentals and how each standard in the Fundamentals applies to their position in the service. The AABB QMS model describes how Operational Processes are supported by proper implementation the Quality System Essentials (QSEs) Framework. This self-assessment process creates an environment for process improvement. The awarded certificate is international recognition of the services' commitment to quality.

Summary / Conclusions: Blood and Transfusion Services that have successfully participated in the Quality Certificate Program are motivated to improve and provide quality products and services to donors and patients. Implementation of the Fundamentals reinforces a culture of quality.

P085 | Impact of standardised reporting of hospital transfusion committees on the evaluation of their performance by the Competent Authority

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Background: The role of transfusion committees (TC) is crucial for blood component therapy. Guidelines issued by numerous regulatory organizations highlight the role and proper functioning of TCs. According to Polish legislation, responsible for the supervision of TCs as well as for blood management in hospitals are blood establishments (BEs). Therefore the cooperation between TCs and BEs is of utmost importance as is forwarding correct and complete information by hospital TCs. Significant shortcomings have been determined. In this respect in 2019-2020, the Institute of Hematology and Transfusion Medicine (IHTM), as the competent authority responsible for

hemovigilance, conducted on-site and online training and prepared a publication of a manual *Standard for Activity of TCs* (Standard).

Aims: The study aim was to evaluate the impact of training and the template for TCs reports published as part of the manual on the standardization of such reports sent to BEs.

Methods: Evaluation of 321 TC reports from the period 2015-2016 (before training), 347 reports from 2019 (after some training and the first edition of the Standard) and 372 reports from 2020. According to the information provided by the BEs, 492 TCs were in operation during this period.

Results: In the period 2015-2016, no uniform reporting template for TCs was yet available and therefore a reliable comparison of TC activities was unfeasible. Moreover, not all TCs submitted reports. However, the uniform report template prepared by IHTM was made use of by only 12% of TCs in 2019, and by 48.4% in 2020. In 2019 and 2020, 64.2% and 74.2% of TCs provided information on the list of TC personnel. In 2019, 32.8% reported the number of nurses to be trained at BEs according to current regulations, and in 2020 it was 55.1%. Information on the number of RBC units ordered and transfused was reported by 40.3% and 59.4% in 2019, and 62.6% and 70.2% in 2020, respectively. The number of wasted RBC units was reported by 46.1% in 2019 and 67.7% of TCs in 2020. The number of internal audits was reported by 26.8% and 53.2%, of TCs in 2019 and 2020 respectively. The number of adverse events was reported by 39.0% and 53.2% of TCs and the number of adverse reactions 42.3% and 57.0% in 2019 and 2020 respectively. An in-depth comparison of TCs' activities based on the submitted reports will be presented on the poster.

Summary / Conclusions: 2015-2016 reports demonstrated that the lack of uniform reporting standards significantly impedes the analysis of TCs' performance. Activities such as training, implementation of standardized reporting templates in the 2019-2020 period rendered evaluation of TC performance more feasible. The number of standardized reports is steadily growing which only facilitates the cooperation with BEs in supplying hospitals with blood components. It seems however necessary to strengthen BE supervision over hospitals and to coerce reports from all TCs. The study results indicate the need for continuous training hopefully by one single unit as this would ensure standardization of the implemented changes.

P086 | Practical aspects of data collection and preparation for the development of Statistical Process Control (SPC) models and control charts in the quality control of blood components in Polish BEs

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Background: Quality control (QC) is part of the Quality Assurance System and an important tool for the daily assessment of the quality of blood components. The purpose of QC is continuous monitoring and improvement of processes directly affecting the quality and safety of blood and blood components, and thus the safety of the recipient as

well as therapy effectiveness. Correct and systematic QC provides important information on the quality of all processes performed at BEs. The use of statistical methods for monitoring the quality of blood and blood components, statistical process control (SPC included), is required by EU legislation and guidelines (EU Directive 2002/98/EC, 2004/33/EC and Guide to the preparation, use and quality assurance of blood components) and Polish guidelines (Ministry of Health Requirements for Good Practice). Data collection and analysis plays a key role in quality management, quality assurance and process improvement.

Aims: The study aim was to develop SPC models and control charts for QC of blood components for better monitoring of the QC process through i.a. determining the number of samples to be collected and factors that affect results. Control charts are used to keep track of results.

Methods: Retrospective data analysis for 2020-2021, 2022 and the 2023 (available data) was based on information collected from 5 pilot BEs. The analysis covered selected data on the preparation and QC results for five blood components most common for Polish BEs. The analysis included data on: buffy coat free RBCs in additive solution, leucocyte-reduced RBC, leukocyte-reduced pooled PC, leukocyte-reduced apheresis PC, FFP from WB and FFP from apheresis. Advanced methods of large data set analysis and statistical analysis were used, as well as selected measures of the dynamics of phenomena: time series analysis, seasonality analysis, forecasting methods. Advanced functionalities of MS Excel software (mainly Power Query) were used for data preparation and performance of multivariate analyses. Microsoft Power Business Intelligence (Power BI) software was used for data analysis and visualization.

Results: Approximately 3.7 million records in nearly 9,500 files were analyzed. They served as basis for development of statistical process control models and control charts implemented in the SPC Application—a software tool to be used by all Polish BEs.

Summary / Conclusions: Data analysis and observation of the processes in BEs led to the conclusion that effective monitoring of processes, including QC of blood components, requires mathematical and statistical models, including time series, which provide a multidimensional view of the important factors affecting the final quality of blood components.

P087 | Abstract withdrawn

P088 | Abstract withdrawn

P089 | Bacteriological control of platelet concentrate in the period from 2015 to 2023 in the Institute for Transfusion Medicine of the Republic of North Macedonia

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Background: Blood transfusion is an essential part of modern health care. A prerequisite for the efficient clinical use of blood is a well-organized transfusion service, which can provide reliable and safe

blood and blood products. Bacterial culture from platelet concentrates gives the best picture of contamination. Platelet concentrates are more susceptible to sepsis than other components, due to the fact that it should be kept at a temperature, which can have the development and reproduction of certain pathogens.

Aims: To analyze the results of bacteriological control of platelet concentrates using BacT /ALERT 3D. BacT /ALERT 3D was used for aerobic and anaerobic bacteriological cultivation since 4 November 2015.

Methods: This is a retrospective analysis of data from the register for "Microbiological analysis of sterility" in the Quality Assurance and Control Department at the Institute of Transfusion Medicine of the Republic of North Macedonia (ITM-RNM) in the period from 2015 to 2023.

Results: In the period from 2015 to 2023, a total of 8458 bacteriological analyzes were performed. In 2015-235, 2016-772, 2017-1336, 2018-866, 2019-1394, 2020-1057, 2021-1247, 2022-275 and 2023-1276 bacteriological analyzes were made. The small number of microbiological analyzes in 2015 is due to the beginning and introduction of the application of BacT / ALERT 3D SYSTEM on 04.11.2015. Then there is an increasing trend in the performed analyzes with one drop in 2018, due to the renovation of the Department/microbiology laboratory in ITM-Skopje and a decline in the trend due to a problem with the procurement of reagents in 2022. A very small number of false positive results were found. Only one true positive result (staphylococcus vitulinus) was found in platelet concentrate, in 2017. Institute of Microbiology and Parasitology at the Faculty of Medicine—Skopje confirm the result. All blood components produced were removed from use.

Summary / Conclusions: The Institute of Transfusion Medicine, as a top healthcare institution, constantly strives for the introduction of modern technologies that would lead to the improvement of the quality. Therefore, in the future, transfusion medicine will be directed towards the development of new techniques of inactivation of pathogens in blood components, as well as the search for alternative solutions for transfusion treatment.

P090 | Ensuring blood component quality with the change of collection bag—a comprehensive evaluation

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Background: The relocation of a factory producing bags for the collection and storage of whole blood (WB) presents substantial challenges in ensuring product quality and safety. It is of vital importance that the change ensures the quality and safety of bags for the collection and storage of WB, as well as the blood components (BC) stored. Additionally, this change may impact into various phases of BC management, including donation, registration, fractionation, labeling, quality controls (QC), and transfusion.

P090 – Table 1

BLOOD COMPONENT AND BAG	VOLUME (mL)	PLATELETS *(x10E6/mL) #(x10E11/U)	LEUKOCYTES (x10E6/mL)	RBC (x10E9/mL)
WB, Previous	530.7 ± 6.3	*200.7 ± 45.0	7.2 ± 4.3	4.0 ± 0.5
WB, Current	530.7 ± 7.3	*189.8 ± 48.7	6.5 ± 1.6	4.3 ± 0.7
WB, p	0.774	0.133	0.208	0.076
FP, Previous	259.7 ± 12.2	*20.8 ± 12.6	0.036 ± 0.047	0.003 ± 0.007
FP, Current	261.1 ± 14.4	*19.8 ± 9.0	0.047 ± 0.049	0.004 ± 0.011
FP, p	0.572	0.597	0.107	0.908
Platelet pools, Previous	330.4 ± 18.6	# 3.79 ± 0.34	0.05 ± 0.05	0.15 ± 0.18
Platelet pools, Current	326.2 ± 18.3	# 3.85 ± 0.56	0.21 ± 0.24	0.15 ± 0.14
Platelet pools, p	0.286	0.557	<0.001	0.548
RBC, Previous	290.1 ± 19.1	-	0.09 ± 0.08	-
RBC, Current	293.2 ± 19.8	-	0.08 ± 0.09	-
RBC, p	0.170	-	0.518	-

Aims: This study aims to assess the suitability of WB donation equipment following manufacturing changes, which may influence the QC of BCs and subsequently impact on transfusion safety.

Methods: A total of 10,584 kits for WB extraction were analyzed through visual inspection and deficiencies were recorded. For registration, fractionation and labeling, evaluation was conducted to ensure that labels remained on the products. Correct reading of codes, assembly on the automatic fractionation system, fractionation, placement of closure labels, and final labeling of BC were also evaluated. For supplies, the correct reading of components was verified. In transfusion, incidents related to ease of cutting segments, opening ports, puncturing, optimal blood flow and transfusion labeling were reported. Blood components were analyzed with special attention through QC. Whole blood units collected in the new bags (WB, $n = 266$), fresh plasma bags (FP, $n = 97$), platelet pools ($n = 104$), and leukoreduced red blood cell concentrates (RBC, $n = 298$) were examined. A Shapiro-Wilk test was conducted to check the normality of variables, and an appropriate analysis, independent samples *T*-Test or non-parametric tests (Kruskal-Wallis) for routine parameters outlined in quality guidelines were performed.

Results: Few minor defects were detected at blood collection points as well as the processing and transfusion area, and new units did not present significant incidents. Blood component specifications consistently met standards, mean and standard deviations are shown:

Summary / Conclusions: The objective was to assess the impact of changes in collecting, processing, and transfusing BC. Minor defects were observed before use in blood donation and a minimal number of units (<0.1%) had deficiencies. Few bags (<0.1%) had

manufacturing defects. No significant issues occurred during fractionation. Regarding the biochemical parameters of the BC, no significant differences were observed between the contents of the bags based on their manufacturing origin. Overall, the changes had minimal impact, with >90% compliance with quality criteria for analyzed components.

P091 | Experience of monitoring the quality control in single blood service, China

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Background: Ensuring the blood safety is of great significance for clinical transfusion. One of the key measures taken for blood safety is to carry out quality control in the blood services, including a quality management system establishment, monitoring key control points throughout the entire process, and continuously improvement strategy, so on. Although many blood services have established a quality management system, the level and methods of quality monitoring varies greatly in different blood services and regions, which will directly affects the blood safety. At present, how to optimize monitoring the quality control still needs further standardization.

Aims: Analyze the monitoring the quality control situation of our center, including quality for whole blood and its components, rate and reasons for discard blood and its components, quality for the equipment, materials and process environment, which will provide experience for the blood services in the quality control.

Methods: Quality analysis for whole blood and its components according to “the Guidelines on quality monitoring of whole blood and blood components” (WS/T550-2017) in China. If the whole blood and its components do not meet the standard in China, they will be discarded. The equipment calibration, blood bag quality, reagent quality and environmental microbial indicators will be regular monitored.

Results: Whole blood and blood components from our blood center randomly chosed for quality inspection every month, therefore 781 whole blood and blood components were analyzed throughout the year, all of them were qualified. The rate of discard blood is 2.32% in 2023, the main reasons for discard blood are lipemia and alanine aminotransferase value exceeds the upper limit. The proportion of discard blood is 0.76% due to transfusion transmitted infections. All bacterial tests were negative, including 1879 for equipments, 267 for object surfaces, 80 for staff hands, 44 for puncture sites of blood donors, 51 for blood transport vehicles. All equipment calibration is qualified during usage process. Validation tests were performed for 447 batches of blood bags, reagents, disposable consumables and disinfectants, but 3 batches of them do not meet the requirements and discard. 48 cases were reported with risks after blood donation in 2023. The overall satisfaction rate of blood donors was 98.86% in 2023.

Summary / Conclusions: A complete quality monitoring system have been established in our blood center, which can ensure the blood safety through daily monitoring. There is still need to improve the validation of reagents and decrease the risk report times from donors after donation.

P092 | Enhancing efficiency and quality in Tunisian blood banks—a 5S-Kaizen-TQM approach and its positive impact

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Background: The 5S-Kaizen-TQM methodology represents a systematic approach emphasizing incremental, cost-effective, and low-risk enhancements. Originally implemented in the Japanese industrial context, its objective extends to facilitating its effective and efficient integration into the healthcare systems of low- and middle-income countries.

Aims: This study aims to highlight the positive impact of implementing the 5S-Kaizen-TQM approach within a blood bank at a public hospital in Tunisia

Methods: The introduction of 5S-KAIZEN-TQM in the hematological department spanned a four-month period from March to June 2023. Initial efforts involved organizing seminars and training sessions on 5S and Kaizen activities for staff members. Data collection employed face-to-face interviews and brainstorming sessions. Presentation of the gathered data utilized checklists and Ishikawa

charts. Subsequently, with active participation from the staff, the initial phases of the approach were implemented and their performance evaluated.

Results: Implementation of 5S activities resulted in a reduction in item research time and increased available space. Enhancements to the working environment led to heightened staff satisfaction. Kaizen activities contributed to a reduction in process lead time, thereby increasing patient satisfaction.

Summary / Conclusions: The project's funding provides an avenue for disseminating and sustaining the 5S-KAIZEN-TQM approach throughout the healthcare sector nationwide.

P093 | Have consent practices for blood transfusion changed in the last 10 years?

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Background: Blood transfusion is not a risk-free procedure. Patients should be given adequate information in a language or format they can understand so they can provide an informed consent or refusal for blood transfusions. In 2022, Blood Matters undertook an audit of blood transfusion consent policy and practices and compared the results with a similar audit undertaken in 2012.

Aims: Identify if health service blood transfusion consent policies are available and consistent with best practice guidelines. Determine if there has been an improvement in consent policy and practice since 2012.

Methods: 140 hospitals from four Australian states and territories were invited to participate in the two-part audit. Part A – Audit of policy. Part B – Retrospective audit of blood transfusion consent documentation found in the medical record.

Results: Part A – Consent policy – 98 health services responded. In 2022, 95 (97%) health services had a policy for blood component consent. Table 1 provides a comparison of results from 2012 and 2022.

The elements that make up informed consent, as described in the Australian and New Zealand Society of Blood Transfusion (ANZSBT) guideline 2018, were more frequently reported in policies in 2022 compared to 2012. Part B – Consent practice. Eighty-seven health services submitted data for 1891 patients. Evidence of consent was documented for 1823 (96%) patients, however documentation was not consistent with all the required elements included in the ANZSBT guideline, with the largest gap in the provision of interpreters when required. Elements of consent with the lowest compliance included: Duration of consent was documented for 1493 patients (82%). Fully documented dialogue with the patient – 1130 patients (62%).

P093 Table 1:

	2022 (%)	2012 (%)
Total policy responses	98	110
Consent policy in place	95* (97)	105 (95)
Policy included elements from best practice guidelines:		
Method for documenting consent	95 (100)	95 (90)
Who can obtain transfusion consent	91 (96)	91 (87)
Period that consent is valid is stipulated	85 (89)	53 (50)
All discussion points documented	66 (69)	37 (35)
Process outlined when patient unable to give consent	89 (94)	Not asked

* Three health services reported no policy, however a statement regarding consent could be found in other sources.

69 patients reported as requiring an interpreter, with an interpreter provided for 17 patients (25%). In 2012, valid consent was self-reported by auditors at 75%, however individual consent details were not examined, making practice comparison difficult.

Summary / Conclusions: Over the last 10 years blood transfusion consent policies have improved. There is evidence that consent practice has also improved with 96% of patients having a blood transfusion consent in their medical record, although further work is needed to ensure all elements required for a valid informed consent are completed and documented, including the use of an interpreter for those patients who need it.

Management and organisation— Vein to vein registries—use of big data—epidemiology

P094 | Abstract withdrawn

Management and organisation— Social, legal, ethics blood donation and transfusion

P095 | When less is more—exploring Australian donor and non-donors' perspectives on proposed changes to donor screening questions

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Background: Several countries have replaced time-based deferrals for men who have sex with men (MSM) with gender-neutral (GN) questions, with no detected decrease in blood safety. Australian Red Cross Lifeblood (Lifeblood) will apply to our regulators to change sexual behaviour questions asked of donors to increase inclusivity whilst maintaining a negligible risk. In Australia HIV transmission continues to be primarily through MSM with heterosexual transmission commonly related to sexual contact with people from higher HIV prevalence countries. As such, while improving eligibility for people from many groups historically excluded from donating, GN approaches may also exclude some lower risk donors. Alternative non-GN approaches may prevent this, however perceptions and acceptability of these alternative approaches is unknown.

Aims: Our aim was to investigate donor and non-donor perspectives of, and preferences for, three alternative sexual behaviour screening questions.

Methods: Donors were recruited through Lifeblood, with non-donors recruited through a market-research company with sampling to maximise diversity by age, gender, location, ethnicity, and sexuality. Structured interviews were conducted online. Participants were shown the current sexual behaviour questions, followed by options for new sexual behaviour screening questions focused on behaviour in the last 3 months in a randomised order. Option 1 was GN and comprised Q1: a question for all about new and/or multiple sexual partners and Q2 (for those answering yes to Q1), a question about anal sex (Q2). Option 2 comprised Q1 and, for those answering yes, different questions for male and female donors about either male-

to-male sex (Q3) or sex with a bisexual male (Q4). Option 3 asked Q1, Q3/4, but with additional questions for those answering no to Q3/4 about sex with a person from a high-prevalence region or with a transgender person. Participants were asked their views on the questions and their most and least preferred options. Data were analysed using deductive and inductive coding. A coding framework was developed by the team after co-coding four transcripts. Themes were identified through identifying higher-level codes related to the research questions.

Results: Participants included 22 (52%) donors and 20 (48%) non-donors. They ranged in age from 18 to 68 (mean 42.5) and 69% identified as heterosexual, 17% as LGBTQ, and 14% unknown. Among all participants, knowledge of which groups are currently ineligible to donate or would become eligible with alternative approaches was poor. Most participants preferred the GN option. The main reasons related to simplicity, asking only what is necessary for safety, avoiding asking questions about a third party and avoiding categorising people based on gender, sexuality, or country of birth. Those who preferred options 2 or 3 commonly believed these approaches would be safer due to more specific questions. Conversely, asking fewer questions was believed to increase eligibility.

Summary / Conclusions: Most participants were in favour of shifting to a GN approach, with this underpinned by prioritising blood safety with asking necessary questions only. If implemented donors will need assurance that a GN approach does not compromise blood safety. Further, given poor general knowledge of who is or would be eligible to donate, clear information will need to be disseminated to donors and the public to maximise uptake among newly-eligible groups and minimise disappointment among those ineligible.

P096 | Is blood a gift or a commodity?—an update of historical theoretical principles in light of the current state-of-the-art

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Background: Blood donation (BD) and transfusion are rising inextricable questions hardly covered by classical legal and political theories that seek to protect elements of the body as a human person herself, whereas in reality, blood derivatives are sometimes placed on a commercial market

Aims: This study aims to analyse the historical theories considering blood as a classical economic good (economical theory ET¹), and those considering blood as a gift, a social bond oriented towards values and social cohesion (theory of social policy SPT²), and to determine, in light of the current state-of-the-art (evidence based and practical observations), whether their arguments remain or not only theoretical.

Methods: We have reviewed the literature to identify the key arguments of the 2 theories, to analyse them by classifying the questions addressed into 2 categories: general and social questions (A), and those related to the remuneration or not of BD (B).

Results: The results are summarized towards the most frequently mobilized thematic as assessed by reviewing the literature: A: The question of donor freedom is involving intrinsic and extrinsic motivational mechanisms, and doesn't seem to be limited to an alleged increase in individual choice through the market³. Wastage: ET considers a not-for-profit (NFP) system to encourage waste of blood products (BP), but the varied and complex national mechanisms of current healthcare price regulation call for a broader frame of analysis. The history has proven Titmuss right when he was predicting a higher rate of infectious diseases in a remunerated system⁴, but the tainted blood drama has also highlighted that unpaid BD alone is not enough to ensure transfusion safety. However, it has historically been easier for an infected patient to obtain compensations in a NFP system where the judge recognised the full liability of blood establishments (BE)⁵, whereas the US judge protected BE towards the blood shield of the *Caveat emptor*⁶. B: Blood regarded as a commodity and the payment for BD are closely linked, which has given rise to other hypotheses we evaluated in a current perspective. The motivational crowding-out induced by monetary compensation for BD has often been contested by ET, but several studies seem to demonstrate its reality⁷. The "value" of BD might be different between gift and commodity. Is BD a choice or a necessity? Is the payment given to donors a fair compensation (ET)⁸ or a risk of exploitation of the most vulnerable people (SPT)? Numerous publications underline the vulnerability-risk associated with high amounts of remuneration⁹. The remunerated models are said to be more efficient⁸ (70% of world's plasma collected), but without analysing access to care when these countries are also experiencing PDMP shortages¹⁰

Summary / Conclusions: These 2 theories have been used for over 50 years to try to determine which model would be better suited to BD and transfusion. The analysis of the literature tends to conclude that the ETs remain often theoretical and don't reflect reality, probably partly because they envisage only sequences in the transfusion chain (remuneration assessed solely in terms of volumes and not in terms of access to care or donor protection), whereas a global approach is needed to embrace the complexity of a personal action with an eminently public and social objective of making BP available, with respect for human beings

1 Arrow, 2 Titmuss, 3 Arrow, 4 Simonetti, 5 Byk, 6 Havighurst, 7 Mellström, 8 Jaworski, 9 Ochoa, 10 Strengers

P097 | Balancing patient access to plasma derived therapies with donor protection for a global health strategy

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Background: Transfusion was born out of a humanist intention to preserve human life. Historically this was associated with many risks: risks for patients linked to unknown incompatibility and infectious events, risks for donors initially associated to those of a surgical procedure, and also a societal risk with regard to an underlying risk of exploitation of human vulnerabilities.

Aims: To develop and implement an ethical global strategy for plasma collection and demand that prioritizes the safety and well-being of both patients and donors.

Methods: /

Results: Risks associated with transfusion have changed over time. Attention to donor health was primarily seen as relevant to mitigate risk for the recipient of the blood components. The effects of donation on the donor have long been seen as secondary and rarely serious. However it has changed over the past decades because it is increasingly apparent that donation can indeed have consequences for short- and long-term health of the donor. For patients, immunological and infectious transfusion risks are now better controlled, with a high level of safety resulting notably of sensitive tests for relevant pathogens, as well as optimized manufacturing processes. For plasma derived medicinal products (PDMPs) no transmission of relevant blood-borne pathogens has been documented in the last 2 decades. Nowadays, one of the main risks for patients is to not have access to treatment. Moreover, the demand is increasing yearly which is calling for a strategic independence. The supply is thus uncertain especially since entry of PDMPs onto the international market, leading to a situation where access to treatment meets call for tenders and associated risks. The debate is too often restricted to the opposition between whether or not donors should be remunerated, with an increasing pressure on donors (frequency and vol. of the donations, advertisements...), and the risk to forgo the principle of non-maleficence while obscuring the other inputs (national organisation of health care, local production or not, wastage of plasma collected and not fractionated, extraction yields, ...). We want to emphasise that while access to care is of high importance, donor protection is always a prerequisite that

ultimately ensures the resilience of patient access to donor-derived medicines. The initial humanist vocation of transfusion and the social risks forces us to go beyond a “supply and demand” approach, and think in terms of fundamental human rights. Actually, blood combines at least dignity and health protection, and the reconciliation of these two notions could make it possible to achieve an overall objective of health protection that is not considered solely from the perspective of access to care.

Summary / Conclusions: At a time when internationally efforts are made to limit trade involving the exploitation of vulnerable populations, it is appropriate to consider donation and the supply of PDMPs, within the remit of human rights, to protect health in the broadest senses. In that perspective, studies to investigate whether plasma donation leads to increased risk of health effects for the donor contribute to ensuring the integrity and ethics surrounding the process. An open and transparent communication about the results is required so that the trust of donors and public can be maintained. It is also crucial that the use of PDMPs be evidenced-based to avoid unnecessary donations, and thereby potential effects on donor health. Each donation should be fully exploited but not the donor.

P098 | Abstract withdrawn

P099 | Drepanocytosis, how changing climate and region can be a problem—a case Report

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Background: Hemoglobinopathies are the most common monogenic disorders in the world. They have variable incidences from region to region. In certain countries, they extend to most of the population, with at least one genetic abnormality affecting the structure or synthesis. Drepanocytosis is a structural hereditary hemoglobinopathy with codominant autosomal transmission, which results from an exchange of amino acids (GLU->VAL) in the β chain of hemoglobin (Hb) with an impact on its form and consequent solubility, being called HbS. It is most prevalent in tropical and subtropical Africa. The disease is characterized by hemolytic anemia, and vasculopathy with vasoocclusion phenomena that cause ischemia and chronic damage to different organs, interrupted by acute events or episodes called crises and susceptibility to infections due to functional asplenia.

Aims: JP, ♂, 21 years old, born in São Tomé and Príncipe, with drepanocytosis and α -thalassemia, residing in Viseu since 2022. To report two complicated crises since being in Portugal, requiring hospitalization, both associated with extreme temperatures. He went to the emergency room in January due to pain in his pelvis and thighs that had lasted 24 hours. On objective examination, there was only jaundice and complaints of mandibular hypoesthesia. Analytically Hb 9.1 g/dL, leukocytosis with neutrophilia, LDH 412U/L, total bilirubin 4 mg/dL, direct bilirubin 0.48 mg/dL.

Methods: He was hospitalized for analgesia and supportive treatment. On D2 initiates fever (up to 40.5°C), accompanied by massive hemolysis with a drop in Hb to 4.5 g/dL, a rise in LDH to 4300U/L, total bilirubin 7.8 mg/dL at the expense of indirect and criteria of disseminated intravascular coagulopathy (DIC). Given the profile of the patient's erythrocyte phenotype, it was necessary to request blood units from another blood bank and postpone the transfusion, as there was no compatible unit in stock. He also presented hepatomegaly with increased cytocholestatic parameters and increased inflammatory parameters. At this point only left mandibular hypoesthesia was reported. He was evaluated by maxillofacial surgery with a CT scan, which ruled out changes in the trabecular or cortical structure of the mandibular bone, ruling out areas of bone infarction, osteonecrosis, or pathological fracture, only recognizing semi-impacted wisdom teeth with pericoronitis. On D5, due to persistent fever, antibiotic therapy was escalated and scheduled for 7 days, with negative cultures and no apparent infectious focus.

Results: After sustained improvement, with good transfusion effectiveness, reduction in hemolysis parameters, and normalization of clotting times, on D14 the patient was discharged, with reevaluation in a Hematology consultation and recommendation to maintain adequate water intake and avoid crisis triggering factors, namely exposure to cold.

Summary / Conclusions: The mobilization of the world population allows rare diseases in certain environments to become increasingly frequent in clinical practice. This may also apply to the variability of erythrocyte phenotypes associated with racial determinants with important implications for transfusion safety and compatibility. The coexistence of hemoglobinopathies can worsen or improve the symptoms, depending on the combination presented. In this case, the patient had an α -thalassemia trait that leads to less hemolysis and higher Hb levels, reducing endothelial dysfunction and vasoocclusive conditions and the risk of mortality.

Blood donation—blood donor recruitment and retention

P100 | Specificity versus inclusivity and othering of ethnic minorities—exploring the impact of message type on blood donor recruitment among black communities in the UK

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Background: The under-representation of Black people in the UK blood donor panel is a crucial policy concern, as well-matched blood from Black people provides a more effective treatment of

Sickle Cell Disease (SCD) that disproportionately impacts Black communities. Addressing this shortage ensures diverse blood supplies and integrates communities into the healthcare system. Several UK-based campaigns have sought to encourage blood donation among Black people, differing in their focus on 'specificity' or 'inclusivity'. Inclusivity promotes unity and shared responsibility, while specificity emphasises health conditions within a target audience. One concern with specificity is the potential for fostering 'othering', a feeling of marginalisation and social exclusion in the target group. This is especially relevant since many campaigns target SCD in Black communities to encourage Black people to donate.

Aims: This study evaluates the relative efficacy of specific vs inclusive campaigns to encourage Black people to donate blood in the UK. An online experiment tests the effectiveness of eight different message frames varying in their degree of specificity and inclusivity.

Methods: Messages were co-designed through interviews, workshops, and discussions with stakeholders at NHSBT and members of the Black community. A $2 \times 2 \times 2$ between-subject design, varying: (1) focus (non-specific; specific) vs. (2) inclusivity (non-inclusive; inclusive) vs. (3) medium (context-free; contextualised, i.e., Instagram) was used. Participants were randomly to conditions The baseline message was a standard 'Give blood. Save lives'. The inclusive message focused on fostering diversity and representation within blood donation, the specific message on the link between SCD and the Black community, and the mixed message a combination of both. Participants were sampled via Prolific ($n = 1,342$: 656 Black people, 686 White people) to target a UK sample. The key outcome variable was 'campaign response effectiveness', a latent variable on subject's willingness to donate, encourage others to donate and not be put off donating. As a mediator, we also collected information on perceived 'othering' (i.e., marginalisation, stereotypical, overlooking diversity, etc.). Data were analysed using Stata 18, Mplus 8.7, SPSS-28, LIWC-22 & ChatGPT.

Results: The mixed campaign message generated the highest 'campaign response effectiveness' from Black people while specifically having some negative impact on White people. Moderated mediation shows that specific messages are associated with significantly higher 'othering', associated with a reduced 'campaign response effectiveness'. These effects were stronger in Black people but reduced in people for those who were aware of the need for black blood.

Summary / Conclusions: Our results highlight a unique hidden barrier of 'othering' to ethnic minority recruitment. In particular, specific campaigns targeting groups on definable characteristics may enhance 'othering'. Our results further indicate that campaigns explaining and enhancing awareness of both the importance of diversity, coupled with specific reasons why certain demographics need to donate blood, appear the most successful.

P101 | Effectiveness of a pulse oximeter device in determining hemoglobin

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Background: The determination of hemoglobin concentration is a test carried out prior to the examination of blood donation according to current legislation. The Masimo Rad-67[®] Spot-Check Pulse CO-Oximeter[®] device (Masimo[®]) uses the pulse oximetry technique, allowing continuous and non-invasive signal processing. The hemoglobin concentration can be interpreted by using the signal processor and algorithms.

Aims: Show the results obtained on the determination of hemoglobin with Masimo[®] and compare them to those obtained by the instrument that we use in routine practice (Hemocue-301) and those obtained by venipuncture with the Cell-Dyn Emerald 22 counter (gold standard) at the, Blood, Tissues and Cells Transfusion Center of Seville.

Methods: The hemoglobin of 41 altruistic donors was determined. In the medical consultation, the value calculated by Masimo[®] was recorded for each of them. The capillary blood sample was subsequently analyzed using the Hemocue-301. Venous blood samples were then taken from each donor (3 ml in an EDTA tube) for analysis using the Cell-Dyn Emerald 22 (Abbott Diagnostics), which was used as a reference counter (gold standard).

Results: We performed the following measurements: Measurement 1: Absolute value of the difference between the values of Hemocue-301 and the Cell-Dyn Emerald 22 reference. The average was 0.8 and its standard deviation 0.5. Measurement 2: Absolute value of the difference between the Masimo[®] values and the Cell-Dyn Emerald 22 reference. The average was 0.7 and its standard deviation 0.5. This means that the differences between Masimo[®] and the Cell-Dyn Emerald 22 pattern are smaller than the differences between Hemocue-301 and the same pattern. In both cases the standard deviation had the same value.

Summary / Conclusions: The sample size studied is small, however the data show that Masimo[®] is an easier instrument that allows a faster determination of hemoglobin concentration, 30 s. It is convenient for the donor to avoid a finger prick. With this instrument, the doctor can determine hemoglobin concentration during the interview, resulting in an increased and improved donor care time.

P102 | Skyrocketing new plasma donors—360° marketing strategies in action

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Background: Plasma donation is as necessary as it is unknown. Quantitative and qualitative studies on the barriers to plasma donation among blood donors demonstrate that the main obstacle is lack of awareness.

Aims: Achieve an increase from 11k donors in 2022 to 14k in 2023 incorporating 4k new donors. Transform the challenge into a national matter involving all stakeholders. Raise awareness of the impact of plasma donation on the healthcare system.

Methods: Launch a 360° marketing campaign with the following actions: Internal in-person training for all BST professionals, involving them in the challenge and educating on technical and donor care aspects. External communication campaign: press conference with media, outdoor advertising, Google Ads campaigns, social media, videos broadcasted in the subway and on YouTube. Communication to blood and plasma donors: postal mailings to retiring donors, email marketing, WhatsApp campaigns, SMS campaigns, donation point animation, newsletters, explanatory brochures, and signage. Mixed donation campaigns throughout the territory with mobile teams. Alliances with all social agents, acting as amplifiers of the campaign: public health system, hospitals, patient associations, donor associations, municipalities, professional colleges, and universities.

Results: In 2023 a record number of new plasma donors was registered, with a 52% increase representing 4106 individuals donating for the first time. The loyalty index increased to 1.91% from 1.83% the previous year, with 46% of individuals donating two or more times on average per year. Total plasma donations increased by 28% compared to 2022.

Media campaigns achieved around 100 impacts, with an economic value exceeding €250k. Online campaigns generated over 10,000 visits to the website during Plasma Donation Week. The YouTube video achieved 7.5k views on YouTube and 11.3k on Instagram. Two patient associations in our country, 13 hospitals, primary care centers of the public health system, professional colleges of medicine, pharmacy nursing and the University of Barcelona joined the challenge. 25 special campaigns were held with mobile teams during

P102 – Table 1

	2022	2023
Total Plasma Donations	23.530	30.021
Total Plasma Donors	11.826	14.542
Repeat Donors	9.118	10.436
New Donors	2.708	4.106

International Donor Day and Plasma Donor Week. 500 professionals were trained in-person in the whole year.

Summary / Conclusions: Internal and external communication is a key aspect for driving plasma donation. When we successfully convey the message to donors, they mobilize and are predisposed to engage in donation. The campaign's success cannot be attributed to isolated actions. Each action has yielded positive results, enhanced by synergies between media push, online and offline campaigns targeting current and potential donors, supported by social agents. To reach potential donors, it is essential to mobilize and activate all communication channels.

P103 | Managing Chagas indeterminate donors—changes needed for re-entry and lookback?

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Background: Due to the absence of incident infections and the low probability of acquiring Chagas disease through the bite of infected mosquitoes in the US, blood donors are tested for *Trypanosoma cruzi* antibodies only at their first presentation. Confirmation of *T. cruzi* infection in blood donors requires a reactive result on the screening assay followed by a positive result on a confirmatory assay. Under the current FDA guidance, confirmed-positive (CP) donors and donors who test indeterminate (IN) on the confirmatory assay are permanently deferred, with no pathway for reentry available. In some instances, repeat donors may be inadvertently tested, in which case, recipient notification for all transfusable products collected from the CP or IN donor is triggered. Inadvertent testing of a platelet donor with >400 prior donations who had an IN result on the confirmatory assay led to a large number of recipient notifications, consignee concerns, and the significant loss of a highly committed donor.

Aims: During the study period, the screening and confirmatory assays used for blood donation screening were formulated with the same antigens. To better serve our donors and hospital customers, we sought to investigate if an IN result was an indicator of true infection by performing supplemental testing with an orthogonal licensed screening assay.

Methods: Between Oct 2019 and Jan 2021, blood donations were screened using the Abbott PRISM Chagas assay (PRISM), with RR donations being further tested using the Abbott ESA Chagas (ESA). Residual volume from ESA-IN donations was retrieved and tested on the Ortho *T. cruzi* ELISA (Ortho).

Results: During the study period, 1326 PRISM repeat-reactive (RR) donations were identified, with 94 (7%) of these donations being ESA CP vs 211 (16%) which were ESA IN. Of the IN donations, 183 (87%) were available for additional testing. Of these, 182 (99.5%) donations were non-reactive on the Ortho; 1 donation was reactive, albeit with low S/CO values. The results of our investigation using an orthogonal assay from a different manufacturer suggest that the

majority of donors whose donations were PRISM-RR but ESA-IN, do not have confirmed infections.

Summary / Conclusions: Current FDA guidance requires permanent deferral of donors who test IN on a confirmatory assay for *T. cruzi* antibodies and does not allow donor requalification. The absence of a reentry pathway, combined with using serological assays formulated with identical recombinant antigens in blood donor screening algorithms, can result in the permanent deferral of donors without confirmed infections. In addition, inadvertent testing of repeat donors may result in the additional loss of highly devoted donors. Data obtained from our investigation show that a majority of donors who tested PRISM-RR/ESA-IN have no confirmed infections, which may enable a pathway for Chagas IN donor reentry and the revision of the requirements for recipient lookback.

P104 | Implementation of sexual risk behaviour donor screening in Canada

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Background: In 2022 Canadian Blood Services and Héma-Québec removed the three month deferral for men who have sex with men and adopted gender neutral criteria assessing sexual risk behaviors in all donors.

Aims: We assessed the impact of these changes on the safety and adequacy of the blood supply, one year post-implementation at Canadian Blood Services and 9 months post-implementation at Héma-Québec.

Methods: All allogeneic donors are asked if they have had a new partner or more than one sexual partner in the last 3 months. Donors answering yes to either question are asked if they had anal sex in the last 3 months; if yes, they are deferred for 3 months. We followed HIV rates before and after the criteria change and interviewed HIV positive donors. We assessed the number of donors answering yes to the new questions and the number deferred by age, gender, and donation status. Data on donors, donations, transmissible disease markers and deferrals were extracted from our epidemiology databases. Source plasma donors were not included. Comparisons were made using the Chi square test.

Results: There were three HIV positive donations out of 990,291 donations pre-implementation and four out of 929,384 post-implementation (0.30/100,000 vs 0.43/100,000, $p = 0.72$). All post-implementation HIV positive donors were male, Canadian Blood Services' donors in Ontario. One was non-compliant with multiple criteria. No risk factors were identified in the other three positive donors, although one had English comprehension difficulties. 2.9% of donors answered yes to a new partner and/or more than one partner; the percentage answering yes to a new partner was higher than more than one partner (2.6% vs 1.2%, $p < 0.0001$). 0.15% of donors were deferred for a new partner and/or more than one partner and anal

sex. Deferral rates were highest in first time, younger donors, with similar rates in males and females, and have decreased to 0.06% one year post-implementation.

Summary / Conclusions: Implementation of sexual risk behavior donor screening resulted in unchanged HIV rates and a manageable impact on blood availability. Gender-neutral criteria have also simplified screening for transgender donors, since donors identified as male or female are asked the same risk questions. After over 30 years, we are no longer asking donors about their sexual orientation, increasing the inclusiveness in our donor base.

P105 | Voluntary blood donation at the Transfusion Institute of Serbia in 2023—challenges of the post-pandemic period

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Background: The challenge of working after the covid pandemic represents a permanent change in lifestyle and organisation of work with the added challenge of the unfavourable demographic parameters in Serbia. In the last decade, the number of citizens has decreased by 540 000 (7.5%), the average age has increased to 43.8 years, 22% of the population is older than 65, and about 100 000 young people has left the country. In order to ensure sufficient blood reserves, changes have been made in the areas of promotion, motivation, education and organisation of voluntary blood drives.

Aims: Demonstration of promotional activities, models of education and concepts of drive organisation in 2023 which have despite the presented challenges ensured a sufficient blood supply.

Methods: Comparison with the organisation in 2019, qualitative analysis of incorporated innovations and effects of the implemented activities in practice.

Results: Traditional campaigns have been organised the Winter and Summer campaign, and “Nothing without women” campaign. Support was shown by sports associations and celebrities. A novelty in the activities was represented by the support of the Serbian Orthodox Church and the Patriarch Porfirije, as well as the patient associations Nurdor and Leuka. Corporate responsibility was encouraged even further through cooperation with public and private companies. Special attention in 2023 was given to the education of young people from the youngest age. Lectures were organized in all schools and faculties. Furthermore, special voluntary training was organised for peer education purposes. The promotion of voluntary donation, and blood drives were done as a part of different manifestations and the transfusion buses were placed in front of shopping centres. Donations were used for placement of promotional billboards and branding of public transport vehicles. At the formal gala at the Parliament of the city of Belgrade with the support of the mayor, the award “Champion of solidarity” and plaques for the most successful environments in different categories. We have intensified cooperation with the media and as a new working concept introduced promotion through digital marketing

in order for the right information to reach the wider population. The website, facebook and instagram profiles and the viber community have been updated daily. More than 2000 broadcasts, guest appearances in the media, announcements in press and on websites have been realised. In the competition Top50 sites itks.rs was announced as the winner in the category for business and social services. In 2023 75644 blood units have been collected. There were 17,568 first-time blood donors. Due to medical reasons, 12680 citizens (14.4%) have been refused. In comparison to 2019, an increase of 5404 (7.7%) in collected blood units was recorded, which represents a significant increase considering the previously mentioned changes following the pandemic, the unfavourable demographic situation and the marked dissatisfaction of citizens with the socioeconomical state in the country.

Summary / Conclusions: The realization of the mentioned activities has significantly compensated for the effect of the previously mentioned challenges. The trust of regular blood donors and citizens has contributed to high motivation for regular blood donation and a stable source of blood supply throughout the year.

P106 | Early assessment of nonanemic iron deficiency—benefiting donor health and blood supply inventory management

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Background: Iron deficiency without anemia (IDWA) is not a contraindication for blood donation, although it is very common among young women and in donors who consume low amounts of dietary iron.

Aims: The aim of this work was to select donors with hemoglobin values suitable for donation but with biochemical indices suspicious for iron deficiency in order to direct them to iron therapy or prolong the time between whole blood donations.

Methods: Since 2021, donors from various voluntary associations have been analysed, these donors were suitable for whole blood donation with hemoglobin values determined by finger puncture, but showed MCV values lower than 80 fL on the complete blood count performed at the Transfusion Service. These donors were summoned to the Transfusion Service to perform tests for the evaluation of iron reserves and, specifically, reticulocytes, reticulocyte hemoglobin content (Ret-He), serum ferritin, serum iron, transferrin saturation and sTfR concentrations.

Results: An incidence of non-anaemic iron deficiency equal to 1.9% was observed (600 donors out of 31,148 total donations). 180 (30%) are male donors with an average age of 41 and with at least three annual donations, and 420 (70%) are female donors with an average age of 25 and at least one annual donation. All donors showed Hb values ≥ 13.5 g/dL in males and ≥ 12.5 g/dL in females, but with MCV values ≤ 80 fL (mean value 70 ± 10 fL) when complete blood count was performed. In all cases, iron deficiency was confirmed by biochemical tests: Ret-He ≤ 29 pg (mean value 21 ± 7 pg), ferritin < 17

mg/dL in males (mean value 20 ± 5 mg/dL, v. r. 17-400 mg/dL) and <13 mg/dL in women (mean value 15 ± 3 mg/dL, see 13-400 mg/dL).

Summary / Conclusions: Data analysis showed that non-anaemic iron deficiency is highly prevalent in young people, women, and regular blood component donors. All donors with non-anaemic iron deficiency were treated with oral iron therapy and, 30 days after the end of the therapy, blood count and iron balance were performed again. Oral iron therapy was found to be safe and effective. We observed an average increase in Ret-He of 11pg and ferritin of 55 mg/dL, and it was not necessary to prolong the donation window in any case. The benefit of this study was both to improve the donor's quality of life through the oral administration of iron, and to reduce future donation delays, which serves to ensure a safe and continuous blood supply to patients.

P107 | Facilitating access to potential blood donor groups in southern Ghana, using blood group testing intervention to promote blood donation

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Background: The WHO recommends a blood collection index (BCI) of 10 to 20 units of whole blood per 1000 population to meet basic requirements for blood transfusion. Ghana BCI of 5.7, 5.8 and 5.8 for 2021, 2022 and 2023 respectively. These figures are well below the WHO recommendation, and are due in part to factors such as fear due to myths and misconceptions, lack of awareness and information, and "low involvement". There is therefore a huge deficit in blood supply. The need for interventions that address barriers, and promote blood donation is critical and urgent. The National Blood Service Ghana implemented a blood group testing intervention within the catchment area of the Southern Zonal Blood Centre (SZBC) to encourage blood donation by non-donors by facilitating access to, and communication with prospective blood donor groups and their members to promote blood donation and organize blood collection sessions.

Aims: This review aims to evaluate the impact of the blood group testing in combination with specially designed blood group cards and motivational messages as an intervention to gain access to and encourage non-donors to donate.

Methods: Potential blood donor groups in the catchment area of the SZBC that had not been previously approached for blood donation sessions or had not accepted to organize sessions were contacted to arrange blood group testing sessions for members. From July to November 2023, sessions were organized at corporate organizations, schools and religious organizations. Members of the groups who attended the sessions, met the age criteria for donating blood (17 to 60 years) and consented were tested for ABO RhD blood groups using the Diagast ABD PAD[®], given their test results via a special "souvenir" cards and educated on why giving blood as a voluntary

donor is important. Within four weeks after the initial visit, the groups were approached to host blood donation sessions at the same sites. During the donation sessions, participants in the ABO RhD testing sessions who were present, were interviewed to collect data on attempted, successful and unsuccessful blood donations since their blood group tests. Participants who were not present were contacted via phone call for the data.

Results: Twenty-four potential blood donation groups participated in the testing sessions. Within the period of June to November, 23 out of the 24 groups hosted blood donation sessions for their members. At the 24 sessions organized for ABO RhD group testing and blood donation education, 3816 participants consented, were registered, tested and given their ABO RhD reports and blood donation education. During follow up, 548 tested individuals responded to the follow up questions. Out of these, 350 (63.9%) reported attempting blood donation post ABO RhD testing and 198 (36.1%) reported not attempting donation. Of the number that attempted to donate, 289 (82.6%) reported successful donation while 61 (17.4%) reported that they were deferred for various reasons. Among those who donated, 278 (96.2%) reported donating as voluntary donors and 11 (3.8%) as family replacement donors. The follow up data collection is ongoing.

Summary / Conclusions: This review suggests that the ABO RhD blood group testing intervention has a potential to facilitate access to blood donor groups with regards to organizing mobile blood collection sessions for the groups and encouraging members to donate. This intervention can be adopted and used for encouraging hard to reach potential blood donor groups to donate blood.

P108 | Knowledge and self-perceptions of blood donation eligibility in Australia, and impact on blood donation

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Background: Little is known about the Australian public's understanding of the eligibility criteria for donating blood, their perceptions of their own eligibility, or how these factors influence the likelihood they will donate blood. Identifying misperceptions about common exclusion factors can inform population-level education, recruitment, and retention strategies. Enhancing knowledge of eligibility criteria may encourage more eligible individuals to donate blood, and minimise deferrals due to ineligibility. Temporary deferrals, especially those implemented at blood donation centres, negatively impact donor retention.

Aims: To assess knowledge of blood donation criteria relating to common exclusion factors, self-perceptions of blood donation eligibility, and how misperceptions may impact the likelihood of blood donation.

Methods: We conducted a cross-sectional national population survey in Australia to obtain a representative population sample. All

P108 - Table 1. Some of the most common exclusion factors and accuracy of knowledge among the Australian population

Deferral	Population Ineligible*	Correct	Incorrect	Don't know
Anemia or iron deficiency diagnosis	9.9	54.0	12.2	33.5
Weight <50kg	4.1	29.3	45.6	25.0
Age (new donor)	3.6	16.2	68.2	15.4
Ever experienced stroke	2.1	34.4	24.4	41.0
Male-to-male sex	1.7	9.9	37.9	50.7
Tattoo in unlicensed venue or overseas	1.5	10.2	56.6	33.1

* Deemed ineligible based on criteria assessed in survey, and weighted to match Australian population.

respondents were aged over 18, matching the minimum blood donation age requirement. Data were weighted to match the general population.

Results: A total of 5,178 people completed the survey, with 43% male, 57% female, 0.4% non-binary, and 0.1% who use a different term. Current donors (i.e., donated blood in past 2 years) made up 9% of the sample, 36% were lapsed donors, 54% were non-donors, and 1% did not state their donor status. Of those age-eligible to start donating (18–74-year-olds), 57.3% were eligible based on criteria assessed. Eligibility was higher among men (62.6%) than women (52.8%).

Of those eligible to donate, 26% mistakenly believed they were ineligible or did not know their status. Those eligible were more likely to be a current donor if they correctly believed they were eligible (17%), than if they incorrectly believed they were ineligible (10%) or didn't know their status (0.3%). Consistent with this, eligible respondents were less likely to be donors if they did not know that they were eligible (75%). Of the eligible non-donors, 28% were unaware that there are eligibility criteria to donate blood. Of those ineligible to donate, 56% incorrectly believed they were eligible or did not know their status.

Summary / Conclusions: Knowledge of the blood donation eligibility criteria was poor, and misperceptions of one's eligibility status were high. Misperceptions of the criteria may result in eligible individuals self-deferring, or ineligible individuals risking a deferral. Those who knew they were eligible were more likely to donate blood. Improved targeted education of the criteria may encourage those who are eligible to donate blood, and prevent those who are ineligible from attempting to donate blood until they are more likely to be eligible.

P109 | Abstract withdrawn

P110 | Sexual behaviours and blood donor selection in Europe—a changing landscape

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Background: Recent years have seen many changes to sexual risk behaviour (SRB) donor selection policies in Europe including reduced time based deferrals and the introduction of individualised risk based sexual behaviour criteria. Benchmarking of current policies and practices can support their implementation and continuous development, contributing to the overall safety and inclusivity of blood donation in Europe.

Aims: To survey SRB donor selection policies in Europe in order to identify trends in policy changes and areas where further assessment is needed.

Methods: A survey by the European Committee on Blood Transfusion (CD-P-TS) of the Council of Europe (CoE) gathered data on SRB donor selection policies from 35 of 38 representative member states (MS). This included policies applied to: men who have sex with men (MSM), use of Human Immunodeficiency Virus Pre Exposure and Post Exposure Prophylaxis (PrEP and PEP), donors with multiple or new sexual partners, sex workers and other SRB considered a risk within MS. Data collection was finalised in 2023 and verified in 2024. Replies were provided in free text form.

Results: From 2020 to 2024, 18 MS implemented changes in SRB deferral policies. Future policy changes are planned in a further 7 MS. In 20 MS, there is no exclusion or specific deferral made for MSM donors, who are selected under the same SRB criteria as all donors. 21 MS apply time based deferrals for PrEP and PEP use, ranging from 3 to 24 months. 7 MS apply permanent deferrals. Deferrals varied based on the indication of use, associated SRB and the route of administration. 7 MS had no specific PrEP and PEP policies. 31 MS apply time based deferrals to donors with multiple or new sexual partners, ranging from 3 to 12 months. This included 2 MS who apply deferrals in the case of multiple partners only, not new partners, and

3 MS only when sexual practices considered higher risk were declared in donor questionnaires. 2 MS apply permanent deferrals. There were different interpretations of "high risk" SRB and "multiple" partners. 2 MS had no specific policies in place for multiple or new sexual partners. 13 MS apply permanent deferrals for sex workers, 8 MS apply a 12 month deferral. In 14 MS, sex workers are selected under the same SRB criteria as all donors. 10 MS indicated that time based or permanent deferrals also apply to partners considered high risk, such as carriers of, or exposure to transfusion transmissible infections, injectable drug users, MSM and sex workers.

Summary / Conclusions: There has been a wave of change in Europe where related to SRB donor deferral policies. Specifically, MS have sought to change policies applied to MSM donors, moving towards individualised risk based sexual behaviour criteria for all blood donors whilst maintaining the safety of the blood supply. Further MS are considering the same as part of future planned changes. This represents a significant shift in policy. There is much variation in the application of donor selection policies and what is considered "high risk" SRB in MS. This may be representative of a diversity of blood systems, donor testing strategies, epidemiology, donor demographics, societal and public health considerations individual to each MS. This overview of current practices provides a baseline with which to review future policy changes, with areas of divergence to further address, in supporting the overall safety and inclusivity of blood donation in Europe.

P111 | Post-donation haemoglobin testing strategy in whole blood donation: A modelling study on the STRIDES trial

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Background: In blood donation in the UK, blood donors with low haemoglobin (Hb) are required to be deferred following an onsite test. Therefore, onsite testing is efficient in protecting donors from bleeding below the regulatory threshold. However, low Hb deferrals are costly, time-consuming and discouraging for donors. New strategies that determine the inter-donation recall interval based on post-donation Hb measures, that is, post-donation testing (PDT) strategies, can reduce low Hb deferrals. However, PDT strategies may lead to more below-threshold donations compared to the current strategy.

Aims: To identify a new blood donation strategy that reduces low Hb deferrals whilst maintaining a low proportion of below-threshold donations.

Methods: We proposed a new PDT strategy where the inter-donation interval is stratified based on the post-donation Hb level at the previous donation. Three further PDT strategies with limited onsite testing for donors with Hb below a certain threshold to reduce below-threshold donations were also considered by varying (i) the Hb threshold for onsite testing and (ii) the recall interval for donors with post-

donation Hb below the regulatory threshold. We evaluated these strategies using discrete event simulation (DES) modelling. The model simulated numbers and proportions of events, including donations, low Hb deferrals, and below-threshold donations for a given donor population and recall strategy. We evaluated outcomes over an 18-month period, with 500 simulations to provide uncertainty estimates. Input parameters for the DES were informed using data from a large, population-based study of blood donors in the UK (the STRIDES PDT sub-study), comprising 16,941 donors (8680 men and 8261 women) returning under the current strategy in England (onsite testing with minimum recall intervals of 12 weeks for men and 16 weeks for women).

Results: During an 18-month simulation period, the PDT-only strategy eliminated low Hb deferrals but led to substantially higher proportions of below-threshold donations for both men (6.5% vs. 2.3%) and women (11.8% vs. 4.5%). Applying limited onsite testing for donors with borderline Hb efficiently controlled the proportion of below-threshold donations whilst reducing the proportion of low Hb deferrals compared to the current strategy. Specifically, for the three PDT strategies with limited onsite testing, proportions of low Hb deferrals were lower than those of the current strategy, with results ranging from 1.0% to 3.7% vs. 5.5% in men and 2.2%-6.3% vs. 8.9% in women, depending on the Hb thresholds for onsite testing and recall intervals. Proportions of below-threshold donations were slightly higher, ranging from 3.0% to 5.1% vs. 2.3% in men and 5.4%-8.8% vs. 4.5% in women. In general, strategies that give more donors onsite testing had lower proportions of below-threshold donations, and delaying recalls for donors with Hb below the regulatory threshold had limited impacts on the proportions of below-threshold donations and low Hb deferrals.

Summary / Conclusions: The newly proposed PDT strategy with limited onsite testing has potential to reduce low Hb deferrals whilst maintaining a low proportion of below-threshold donations.

P112 | Abstract withdrawn

P113 | Abstract withdrawn

P114 | Determination of Specific Gravity (SG) of copper sulfate solution 1.066 SG as confirmation of upper limit of hemoglobin level of donor selection in Indonesia regulation

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Background: Examination hemoglobin levels at the donor selection stage using Copper Sulfate solution with a Specific Gravity of 1.053 is still one of the references by WHO because equivalent for specific gravity of Whole Blood with Hemoglobin levels of 12.5 ± 0.18 G/dL. In the Regulations of the Indonesian Ministry of Health, limit of

hemoglobin levels is 12.5 G/dL–17.0 G/dL. Considering that there is no Copper Sulfate solution reagent to confirm Hemoglobin levels of 17.0 G/dL in Indonesia, the authors tried to do an experiment to determine the Specific Gravity of Copper Sulfate solution which is equivalent to Hemoglobin levels of 17.0 G/dL.

Aims: The Aims of this experimental based research is to create a Copper Sulfate solution reagent product with Specific Gravity equivalent to a hemoglobin level of 17.0 G/dL, so it can be useful for Red Cross Blood Donor Units in various regions in Indonesia who still uses Copper Sulfate solution reagent for donor selection to confirmation haemoglobin levels are equal or more than 17.0 ± 0.18 G/dL.

Methods: The method used in this experiment is to make Copper Sulfate solutions with certain specific gravity including 1.062, 1.063, 1.064, 1.065, 1.066, and 1.067. The solution was measured using Densitometer (Mettler Toledo with tolerance ± 0.0003). We prepared wholeblood with several individuals with known hemoglobin levels and re-measured it using hematology analyzer (Sysmex XN-L series). The hemoglobin levels are 16.4 G/dL; 17.1 G/dL; 18.0 G/dL. Each Copper Sulfate solution (30 mL) is tested with each blood that has known hemoglobin levels (the distance of the blood drop to the solution is about 1 cm). Each blood was repeatedly dripped 5 times in each solution. The results of the blood droplets are observed whether they float, drift or sink in the solution.

Results: Blood test results with haemoglobin levels of 16.4 G/dL, 17.1 G/dL, 18.0 G/dL were seen to sink in copper sulfate solution with specific gravity 1.062, 1.063, 1.064, 1.065. Blood test results with haemoglobin levels of 16.4 G/dL seen floating, while haemoglobin levels of 17.1 G/dL is drifting, and 18.0 G/dL seen sinking in a solution with a specific gravity of 1.066. Blood test result with haemoglobin levels of 16.4 and 17.1 G/dL were seen floating, and 18.0 G/dL seen drifting in a solution with a specific gravity of 1.067.

Summary / Conclusions: According on the results of testing Copper Sulfate solution on blood samples with predetermined haemoglobin levels, the ideal specific gravity for haemoglobin 17.0 ± 0.18 G/dL was obtained, which is at a specific gravity of 1.066. These results explain that the blood that floats on the surface of the solution describes the value of haemoglobin less than 17.0 ± 0.18 G/dL, that is by using blood with HB levels of 16.4 G/dL. The results of blood that floats in the middle of the solution describe the haemoglobin levels equal to 17.0 ± 0.18 G/dL, that is by using blood with haemoglobin levels of 17.1 G/dL. while the results of blood that sinks describe hemoglobin values greater than 17.0 ± 0.18 G/dL, that is by using blood with haemoglobin levels of 18.0 G/dL.

P115 | Knowledge, incentives and barriers, and political ideology—blood donor behaviour in black and white people of differing nationalities (British, Nigerian) and countries of residence

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Background: Recruiting and retaining more Black donors is critical for managing health in diverse populations with diverse health needs. To better understand this, we need to examine barriers and the perceived effectiveness of incentives and knowledge donation and how incentives and barriers vary across a wide set of cultural backgrounds. Blood donation systems do not operate in political and social vacuums. As such, we need to understand how political ideologies influence willingness to donate by ethnicity. Also given health inequalities in blood between the global north and the global south, we also need to know what people from different cultural backgrounds know about different systems,

Aims: To explore how do barriers, and perceptions of incentives and knowledge of blood systems vary by cultural background and how does political ideology influence willingness to donate.

Methods: We report two studies. Study 1 assessed motivations (e.g., altruism), barriers, perceived effectiveness of incentives versus rewards, donor status, and knowledge of donation systems across four groups based on ethnicity (Black; White), nationality (British; Nigerian), and country-of-residence: (i) Black-British people ($n = 395$), and Black-Nigerian people (ii) in the UK ($n = 97$) or (iii) across the rest of the world ($n = 101$), and (v) White-British people in the UK ($n = 452$). We also sampled a Black-Nigerian Expert group ($n = 60$). Study 2 replicates this in an additional sample of Black-British ($n = 150$) and White-British people in the UK ($n = 350$) people, where we also assess political affiliation.

Results: Knowledge of donation systems in the global south was low. As a barrier, fear of poor health from donating blood, such as the inability to replace blood, as observed in Black people, White people were more concerned about fainting, and all Black groups reported a lack of trust in healthcare. For the Nigerian lay samples, rewards (e.g., food) were seen as effective, but for Black British people, financial incentives. A more left-wing political ideology was associated with a willingness to donate to Black-British non-donors.

Summary / Conclusions: Barriers and incentives vary by cultural background, indicating that a one-size-fits-all approach will be ineffective. Political ideology is important and often overlooked, indicating that political organisations could help recruitment. Awareness of health inequalities around the availability of blood needs to be highlighted in the global north.

P116 | Community-based research and engagement— understanding barriers to donation for communities of African and South Asian ancestry

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Background: Many blood operators around the world need to diversify their donor base to better meet the needs of a genetically diverse population and to maintain long-term sustainability of their donor pool. Research shows that barriers to donation for under-represented racialized communities are complex and interacting, and requires great attention to how organizational, community, and systemic barriers interact at the individual level. This presentation reports results from two qualitative case studies examining systemic barriers to donation for communities of African ancestry and South Asian ancestry. These communities have a high degree of intra- and inter-community diversity. Understanding the broader context for both communities is necessary to understand and address barriers to donation.

Aims: To understand communities' priorities and how these inform barriers to donation for communities of African and South ancestry.

Methods: We conducted two related qualitative case studies informed by community-based research methodology. Semi-structured interviews were completed with 10 key informants in each community who identified as a leader, for a total of 20. Interviews were conducted from Dec/22-Mar/23, audio-recorded with participant's consent and transcribed. Participants had the option to review their transcript for accuracy. Data were analyzed using a thematic analytic framework.

Results: Case study 1 (communities of African ancestry): Community priorities include: (1) healthcare disparities for people with sickle cell disease; (2) inequitable healthcare services; (3) gun violence; (4) Islamophobia; and (5) job security and employment equity. Barriers to donation were organized into three themes: (1) sociocultural views on blood and donation; (2) deferral criteria; and (3) mistrust and absence of blood operators. Case study 2 (communities of South Asian ancestry): Community concerns include: (1) economic concerns; (2) challenges with healthcare system; (3) inadequate housing; (4) immigration policy; (5) declining quality of education; (6) health challenges post-covid 19; and (7) Islamophobia. Barriers to donation include: (1) accessibility; (2) deferral criteria; (3) gaps in awareness and knowledge; (4) language barriers; (5) fear; (6) other commitments; and (7) lack of response from the blood operator. Key informants in both communities spoke extensively about experiences of systemic racism both at the personal and community levels in their everyday lives; however, mistrust was a barrier to donation for communities of African ancestry but not communities of South Asian ancestry. Another notable difference was the level of engagement with blood operators. For key informants of African ancestry, engagement with blood operators was minimal given their noted absence; however, key

informants of South Asian ancestry described more extensive engagement with blood operators, which they often initiated and maintained. **Summary / Conclusions:** Results suggest that barriers to donation differ for different racialized communities, and these are informed by community concerns, histories, and experiences with healthcare and state systems. Barriers to donation should be understood within the context of each community's experiences of systemic racism more broadly beyond the donation context. Blood operators should work with communities to address systemic barriers and collaborate with community leaders to understand community priorities and experiences.

P117 | Use of social media (Facebook) to mobilise voluntary donors in disasters—a case of suicide bombing in Peshawar, Pakistan

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Background: Social media has the ability to significantly alter human behaviour and has become the foundation of numerous healthcare initiatives, including blood donation drives. In January 2023, a suicidal terrorist attack occurred in a mosque in the city of Peshawar. The blast resulted in 101 fatalities and over 300 wounded individuals, resulting in a high demand for blood and blood components.

Aims: The current study assessed the efficacy of social media (Facebook) in promoting voluntary blood donations after a suicide blast.

Methods: The first goal was to assess the needs through regular communication with hospitals, and the immediate requests were satisfied with blood components from all blood groups transported to the Lady Reading Hospital (LRH), located near the blast site. Within 30 minutes of the blast, a social media post was placed on the RBC's Facebook page. The post was subsequently boosted (paid advertisement) to broaden its reach, targeting not only existing followers (56k) but a wider audience. Responses from potential blood donors (likes, comments, and messages) were closely monitored and responded to. Services were scaled up within the RBC and at the blood collection site in LRH Blood Bank (adjacent to the blast site). Blood donation camps were also set up by NGO-sector blood banks and religious institutes to accommodate the inflow of donors.

Results: During the first 24 h after the disaster, 209 voluntary donations were collected at the blood collection site, compared to an average of 62 donations in routine (3-fold increase). A total of 441 voluntary donations were collected in the first 48 h, in addition to routine blood collections. Of these 441 donors, 56.23% ($n = 248$) were 'first-time' voluntary donors, which is expected bearing in mind the circumstances and the motives. A significant number of females donated blood (27.89%; $n = 123$). The first-time donors showed higher prevalence rates for transfusion-transmitted infections compared to regular donors (4.61% vs 4.23%). Of the 441 donors, 309 (70.06%) were informed via RBC's Facebook page, 89 (20.18%)

by TV, and the rest by announcement in mosques (6.57%; $n = 29$) and through friends or family (3.17%; $n = 29$). The number of donations in January 2023 was 3.6% higher than the number of donations in January 2022 ($p < 0.0001$). Similarly, data analysis showed that in January 2023, the number of female donors ($p = 0.0018$) and first-time donations ($p < 0.0001$) considerably increased compared to January 2022. The number of deferred donors ($p = 0.035$) also increased (from 0.33% to 1.13%), which is a consequence of the large number of first-time donors and female donors. In 2023, out of 62,185 blood donations collected, 16.01% were from voluntary donors ($p < 0.0001$), compared to 12.54% of voluntary donors in 2022 and 10.32% in 2021.

Summary / Conclusions: There was a substantial increase in blood donations post-disaster, particularly among 'first-time' voluntary blood donors. This altruistic behaviour needs to be capitalised by converting these first-time donors into regular voluntary donors through sensitization. The increase in the number of female donations is encouraging, as only 2.04% of females donate blood annually. The effective use of social media confirms that it is a promising approach to eliciting a prompt community response.

P118 | Do donors talk to others about blood donation, and if not, why not? A survey study

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Background: Donor-recruit-donor interventions are considered one of the most cost-effective recruitment strategies. Positive accounts of blood donation (i.e., word of mouth [WoM]), by people whose opinion we value and trust can increase awareness, trust, commitment and positive feelings toward blood donation. While donors indicate willingness to engage in positive WoM, comparatively few donors appear to engage in it. However, little is known about what motivates donors to engage in WoM, the barriers they perceive or their actual practice of attempting WoM about blood donation.

Aims: The aim of this research was to explore WoM in blood donors, with a particular focus on the triggers for, audience and valence of their WoM, along with their motivations, perceived barriers and ability to engage in blood donation related WoM.

Methods: An online survey was distributed to 11,551 active blood donors, with 1599 responding. Of the 1227 providing useable data, 53.1% were male, aged 18-75 years (Mean = 45 years), with 55% born in Australia. Most (81%) were experienced donors (13% novice, 6% new) with a mean donation history of 33 donations, over 10 years (range 0-52).

Results: Talking about blood donation either in person or online was not something respondents commonly did (>64% indicated they talked in person less than monthly, and >84% of social media users posted online about donation less than monthly). Interactions

were most often with family and friends, and least likely at work or in the broader community. However, talking about blood donation in person and online occurred significantly more often ($p < 0.05$) than talking about donating money, goods, or time. Engaging in WoM was most often triggered by personal experiences (donated blood [93%], experiences in centre [77%], questions about personal schedule [74%]) or highlighting of need in media (63%), with motivation to engage in WoM stemming from wanting to ensure supply (those who need blood will receive, 89%), viewing WoM as another way to give (to the community, 90%) or as part of their role as a donor (61%) and wanting to help others make the 'right' decision (to donate, 82%). Some respondents (<24%) saw WoM as having positive reputational benefits for themselves. While most respondents (97%) reported saying at least mostly positive things about donating blood, 56% indicated receiving negative WoM from others about blood donation. Consistent with this, concern about the reactions of others to WoM was identified as a barrier both in terms of risk to the self (e.g., concern with being seen as bragging; 33%), their relationship with their conversation partner (e.g., learning something about the other they didn't want to know, 11%) and general disapproval (of recommending blood donation, 6%). However, the biggest barriers to WoM were belief that donating is a personal choice (70%) and that their conversation partner wouldn't be able to donate (e.g. insufficient time, 18%). Despite this, most donors felt confident to talk about blood donation (78%).

Summary / Conclusions: Blood donation is not often talked about and triggers most often used to start conversations occur infrequently (e.g., donating, media call outs). Further, while donors feel able to engage in WoM, they see interpersonal risks in doing so. Given the documented benefits of WoM, blood collection agencies should work with donors to help them effectively navigate the risks they perceive in engaging in WoM about blood donation.

P119 | Negative impact of questions on travel and sexual history on ethnic minority donor recruitment

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Background: People from ethnic minorities are less likely to donate blood; thus, encouraging greater diversity of donors is psychologically and clinically beneficial (e.g., treatment of sickle cell disease: SCD). Deferral criteria that differentially affect people from minority communities may reduce the effect of any successful recruitment campaigns. We explore the effect of two commonly used deferral questions that are more likely to impact people from ethnic minorities (1) having had sex with anyone who may ever have had sex in parts of the world where AIDS/HIV is very common, (High-Risk-Population or

HRP question), and (2) travelled and returned from a tropical area. We explore if these put potential donors and result in negative feedback to communities to not donate blood.

Aims: Does exposure to HRP and travel questions negatively impact potential donors and the wider community, creating a barrier to recruiting ethnic minority donors?

Methods: 891 people (Asian = 182, Black = 141, Mixed = 182, White = 456), of whom 761 were current donors, were sampled through NHSBT and community samples. They completed an anonymous online survey between June and August 2019. They read the two questions on travel and HRP-SSA and indicated if this would put them off donating and if they would tell people in the community not to donate. We also assess perceived racial discrimination in the NHS and social isolation

Results: The result showed that 34% of black non-donors would be out off after reading the HRP-SSA question, and 17% of Black donors tell others not to donate. After reading the travel questionnaire, 17% of black non-donors would not donate, and 11% stated they would tell others not to. The association between ethnicity and being put off donating and telling others not to donate was mediated by increased perceptions of racial discrimination in the NHS.

Summary / Conclusions: Deferral questions that differentially affect people from ethnic minorities result in both putting of donors and leading to negative feedback not to donate. Such negative feedback is likely to spread and is extremely hard to counter. Blood services need to justify to the donor why these questions are asked and to remove them where possible. Indeed the HRP question was removed as part of the FAIR project work on individualised approach to donor selection in the UK?

P120 | Practice of reservation management for plateletpheresis donors in Hangzhou of China

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Background: The long plateletpheresis collection time (including uncertain waiting time) is one of the main reasons that negatively affects the recruitment and retention of plateletpheresis donors. Also for this

reason, plateletpheresis donors tend to choose holidays rather than working days to donate and the concentration of holiday collection challenges both the service ability and clinical supply. Based on this background, our center started to carry out multi-dimensional reservation management of plateletpheresis donors, mainly including: 1. With the help of information technology, establish plateletpheresis reservation management platform. Donors could make reservation 7 days in advance through the mobile Alipay app, and the on-site call system is ranked according to the principle of priority and reservation almost no need to wait. 2. Improve the collection service capacity of working days, thus attracting blood donors to transfer to working days. 3. Strengthen the communication with those who do not follow the reservation or make reservation and actively guide them to change their behavior.

Aims: To summarize the practical experience of reservation management of plateletpheresis donors and evaluate its effect.

Methods: The proportion of following the reservation or not (reserved and registered or unregistered), and the proportion of making reservation or not among donors who were registered (registered and reserved or unreserved) and their respective proportion of qualified donors (qualified and reserved or unreserved) were analyzed, as well as the proportion of plateletpheresis donation during holidays and working days was analyzed from 2021 to 2023.

Results: From 2021 to 2023, the proportion of plateletpheresis donors who followed the reservation (reserved and registered) and the proportion of reservation donors (registered and reserved) among the registered donors increased year by year, with 81.82%, 87.00%, 91.12% and 71.76%, 81.60% and 85.11%, respectively ($p < 0.001$); During the three years, the qualified rate of the reservation donors were about 85%, significantly higher than that of the non-reservation donors whose average rate were 69.57% ($p < 0.001$); the average number of plateletpheresis donors of holidays and working days were 118.6, 115.6, 114.2 and 80.4, 90.9, 99.7 respectively from 2021 to 2023. Their gap was gradually decreased and their ratio was 1.47, 1.27 and 1.15 respectively.

Summary / Conclusions: The reservation management promotes the sense of both making and following reservation for the plateletpheresis donors and forms benign mutual promotion with the good experience of plateletpheresis donors, which is conducive to the recruitment and retention of blood donors. It also effectively promotes the daily balance of plateletpheresis collection so as to better guarantee the clinical platelet supply.

P120 - Table 1

	Reserved+Registered	Reserved+Unregistered	Registered +Reserved	Registered +Unreserved
2021	29470 (81.82%)	6547 (18.18%)	29470 (71.76%)	11596 (28.24%)
2022	36089 (87.00%)	5393 (13.00%)	36089 (81.6%)	8137 (18.4%)
2023	38776 (91.12%)	3777 (8.88%)	38776 (85.11%)	6785 (14.89%)
χ^2	1483.8		2524.6	
p	<0.001		<0.001	

P121 | Abstract withdrawn

P122 | #Blood donation - how European blood banks use social media to reach donors and non-donors

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Background: One of the main barriers to donating blood (products) is lack of awareness and knowledge about the need for more donors and the donation process. Traditionally, socialization through family, schools, and friends has played a crucial role in shaping behaviours and identities however, in recent years, social media has emerged as an equally influential element in this process. While previous studies explored health communication and blood donation-related communication via social media, these efforts have been sporadic and lack a comprehensive perspective. Hence the academic literature lacks a theoretical understanding of current practices of online communication in the field of blood collection. This study aims to bridge this gap by exploring the current practices of blood collection agencies, focusing on their social media strategies and communication tactics. By understanding these practices, we can facilitate the establishment of a direct link between academic research and the practical needs of the field. This alignment could advance the impact of academic research and enable organizations to develop more efficient and effective communication strategies and blood donation campaigns, ultimately benefiting both donors and patients.

Aims: The primary objective of this study is to investigate how blood product collection institutions across Europe utilize social media to communicate with donors and non-donors.

Methods: During May and June 2023, we conducted 22 semi-structured interviews with communication and marketing specialists from 13 different European countries. We employed thematic coding to identify key themes within the data.

Results: Social media is an important component of the communication strategies employed by blood banks in Europe, used not only for engaging with current and potential donors but also for societal positioning and establishing collaborations with external entities, such as public institutions and private companies. Key platforms used by blood banks include traditional ones such as Facebook and LinkedIn, alongside newer ones like TikTok and Spotify. The content shared spans on a broad spectrum, from educational materials and memes to stories of donors and patients. For creating this content, blood banks rely on in-house production, collaborations with creative agencies, and user-generated content. The strategies employed in distributing the information are equally diverse, including collaborations with influencers, paid online advertising, audience targeting, and the use of micro-content. Finally, we observed new trends in social media usage, including data-driven communication decisions, the adoption of new platforms (e.g., Snapchat), and formats (e.g., podcasts), along with emerging challenges like misinformation and privacy concerns.

Summary / Conclusions: While the analysis of the interview content is still in progress, the preliminary results offer already valuable

insights. For researchers, he findings will support them in developing targeted interventions for donor recruitment, assessing the effectiveness of social media campaigns conducted by blood banks, or understanding the impact of digital communication on donors behavior. Equally, these findings offer practitioners a comprehensive and current overview of the field, shedding light on recent online strategies and practices for engaging with donors and non-donors, alongside identifying emerging challenges and trends that are shaping the future of donor communication.

P123 | Abstract withdrawn

P124 | Abstract withdrawn

P125 | Age-specific blood donation turnout among first-time blood donors during the major COVID-19 outbreak in Taiwan

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Background: During the COVID-19 pandemic, the Central Epidemic Command Center implemented various epidemic alert measures to prevent sustained community transmission. As a result of the pandemic and these control measures, there has been a significant reduction in blood donation, which has disrupted blood supply and transfusion services. The major COVID-19 outbreak was characterized by restrictions on indoor and outdoor gatherings, closure of public venues and the implementation of social distancing measures in Taiwan.

Aims: We aim to examine the shifts in age distribution among individuals donating blood for the first time during the peak of the outbreak at four blood donation centers.

Methods: We retrieved data on first-time blood donors from the Taiwan Blood Services Foundation blood donor database at blood centers in Taipei, Hsinchu, Taichung, and Kaohsiung for the duration of the study. The study period was divided into four time frames: (1) the pre-COVID-19 pandemic era (November 1, 2019–January 20, 2020), (2) COVID-19 alert levels 1 and 2 (January 21, 2020–May 18, 2021), (3) the major COVID-19 outbreak period with alert level 3 restrictions (May 19, 2021–July 6, 2021), and (4) the period of implementation of an effective epidemic control and management policy coexisting with the virus (April 1, 2022–July 31, 2022). The age groups analyzed were: (1) 17-24 years (youngest cohort), (2) 25-40 years (adult group), and (3) 41-65 years (middle age group). We used the Chi-square test to assess any differences in age-specific turnout across the study periods at the four blood centers. All statistical analyses were performed using the statistical analysis system (SAS) software, version 9.4. Statistical significance was specified at $p < 0.05$.

Results: A total of 230,334 individuals participated in blood donation across all centers during the study periods. Taipei center had the highest number of donors (75,489, 32.8%), followed by Kaohsiung

(61,684, 26.8%), Taichung (54,088, 23.5%), and Hsinchu (39,073, 17.0%). The distributions of donors varied significantly throughout the study periods, both overall and across different areas (all $p < 0.001$). Period 3 had the lowest turnout at all centers (3.96% in total, 3.80% in Taipei, 4.32% in Kaohsiung, 3.92% in Taichung, and 3.76% in Hsinchu) compared to the other periods (in total, period 1: 15.12%, period 2: 67.45%, and period 4: 13.47%). When examining age groups, the percentage of first-time donors varied significantly across all centers throughout the four periods ($p < 0.001$). It is noteworthy that the youngest donors exhibited the highest percentage in Hsinchu, Taichung, and Kaohsiung during all four periods. However, in Taipei, they only had the highest percentage during periods 1, 2, and 4. During the major outbreak era, the percentage among the youngest donors was 33.31, while that for adults was 42.79.

Summary / Conclusions: This study highlights a substantial decline in the proportion of young first-time donors at the Taipei blood donation center during the major COVID-19 outbreak. Consequently, it emphasizes blood centers need to modify and enhance their outreach and retention programs, particularly in situations as the COVID-19 pandemic. Furthermore, this study serves as a valuable reference for assessing potential shifts in blood donors' behaviors during future public health emergencies.

P126 | Influence of blood donation motivational domains and fear of COVID-19 on perception towards blood donation, among young blood donors in the post-pandemic era

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Background: Encouraging young people to donate and to continue to retain them as regular blood donors is one of the cornerstones within the national blood programme. An important consequence of the COVID-19 pandemic was a donor shortage, substantially contributed by a decline in young donors who could not donate due to restrictions in operations of colleges and schools. The disruption of regular blood collection activities during the pandemic have had an impact on promotional activities with consequences on donor motivation to donate, especially among young individuals. The additional fear of being infected by COVID-19 may affect the motivational factors and influence their perception of donation.

Aims: We aimed to assess perception and motivational factors that drive young people to donate blood and identify if the COVID pandemic had influenced their donation behaviour.

Methods: Blood donors between the age of 18 to 29 years were interviewed through an online questionnaire during donation sessions. The Blood Donors Function Inventory (BDFI) was used to assess donor motivation within the 5 domains of values, understanding,

social, protective, and enhancement while an in-house questionnaire was used to assess perception towards blood donation. Assessment of fear toward COVID-19 was made through the Fear of COVID-19 Scale (FC-19S). All responses were scored on a 5-point Likert scale.

Results: A total of 215 respondents were recruited. 189 (88%) of respondents were between the age of 17-24 years, with the remaining between 25-29 years. First time donors accounted for 111 (51.6%) of the respondents. The highest ranked motivational domain was 'values' (median score: 4.5; CI: 3.5, 5.0) followed by 'enhancement' (4.2; CI: 2.6, 5.0). The lowest ranked domain was 'protective' (3.7; CI: 1.9, 5.0). Median perception score was 4.0 (CI: 4.0, 5.0) and showed no significant difference between genders. FC-19S scores were significantly higher among females (3.0; CI: 1.2, 4.1) as compared to males (2.4; CI: 1.0, 4.3; $p = 0.002$, Mann-Whitney U-test). Weak to moderate correlations between fear of COVID-19 and the 5 motivation domains were observed, with 'values' showing the weakest correlation ($r = 0.154$, $p = 0.024$) and enhancement showing the strongest ($r = 0.394$, $p < 0.001$). No significant correlation was observed between the perception scores and fear of COVID-19. Ordinal Logistic Regression indicated that understanding, social, and protective motivation were positively associated with higher levels of perception toward blood donation. Enhancement motivation was negatively associated with perception level (OR = 0.38; CI: 0.18, 0.82; $p = 0.014$).

Summary / Conclusions: Young donors continue to show altruistic 'values' as the most common motivator for donating blood even post-pandemic. 'Fear of COVID-19' has shown no impact on donor perception towards blood donation. On the contrary, higher 'fear' appears to correlate with increased motivation, especially among those who consider 'enhancement' as a motivator. We speculate that individuals with higher levels of fear towards COVID-19 may be more motivated by enhancement because they see donating blood to achieve personal satisfaction as a coping mechanism to confront their fear. Individuals who seek 'enhancement' through blood donation however demonstrate lower perception of the blood services. Identifying how these motivational domains influence perception will allow the donor services to target these areas to avoid poor donor perception in future.

P127 | ISBT World blood donor day social media campaign

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Background: In an era dominated by digital connectivity, social media platforms have emerged as powerful tools for disseminating information and mobilizing communities, including. Within the realm of healthcare, particularly blood donation initiatives, social media offers unprecedented opportunities to raise awareness and engage donors. In 2022, the Clinical Transfusion working party initiated annual social

P127 – Table 1

	2022	2023
Volume of mention	94	209
Social media reach	312 K	1.6 M

media campaign for World Blood Donor Day (WBDD), aiming to celebrate the occasion and emphasize the importance of blood donation in supporting clinical transfusion. These campaigns were named after the World Health Organization slogans for 2022 and 2023.

Aims: The aim of this abstract is to outline the activities conducted during these campaigns and assess the level of engagement from the online community. Through this analysis, we seek to provide insights into the effectiveness and impact of these initiative in promoting blood donation awareness and mobilizing public participation globally. Primary engagement metrics include the utilization and traction of the official campaign hashtags, #ISBTWBDD2022 and #ISBTWBDD2023, tracked using BrandMentions, Hootsuite and SocialInsider for the respective years.

Methods: Working party and ISBT members were invited to post over two weeks on blood donation (June 1st-14th) with invitations sent via email through ISBT networks and social media accounts. Participants were asked to post on various platforms (X, LinkedIn, Instagram, Facebook) and were encouraged to use official hashtags and tag ISBT social media accounts and engagement with other posts. In 2023 campaign, a toolkit of posts was also designed by the office and shared with the ISBT members for use. A dedicated webpage was established on the ISBT website for this campaign, providing comprehensive information and social media assets for users to share across their platforms. Additionally, the 2023 campaign featured an opening and closing videos by the ISBT Regional Director of Africa Region and the Working Party Chair, which were posted on the social media accounts of ISBT.

Results: The use of hashtags in both campaigns, increased reach and engagement, with significant participation from ISBT members, the public, and different institutions. Posts included personal experiences with blood donation, blood donation drives, thank-you messages to donors, educational content on clinical indications for blood transfusion, and recruitment calls for blood donation. The 2023 campaign had participation from 22 countries, compared to 16 in 2022, with higher mentions and reach (Table 1). The 2023 email campaign reached 11,124 recipients, with a 65% open rate. Activity peaked on WBDD (June 14th) and included posts in multiple languages, such as English, Spanish, French and Portuguese. The winners of the 2022 and 2023 campaigns were Dr. Kamini Khillan (a working party member) and the National Blood Service of Ghana.

Summary / Conclusions: This WBDD social media campaign highlights the power of digital platforms in raising awareness and promoting blood donation. It also highlights the tremendous efforts undertaken by the public and blood centers. Continuing this annual initiative holds promise for further mobilizing communities and fostering solidarity for saving lives through blood donation.

P128 | Abstract withdrawn

P129 | Evaluation of deferral patterns and correlation between donor return among temporary deferred and accepted whole blood donors at National Blood Centre, Sri Lanka from May 2020 to April 2022

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Background: National Blood Transfusion Service (NBTS) in Sri Lanka is a centrally coordinated system which relies on 100% voluntary non-remunerated donors. However, donor deferral remains a challenge, affecting donor return and retention rates. Investigating the impact of deferrals is crucial and large scale studies are scarce in Sri Lanka.

Aims: This study aims to evaluate the donor deferral patterns and their impact on donor retention in Sri Lanka.

Methods: This retrospective descriptive cross-sectional study was conducted in National Blood Centre, from May 2020 to April 2022 using data from donor declaration forms and blood bank management system. 1605 donors were sampled out of 59,821 and details were recorded and analyzed. Donor demographic data and deferral characteristics were evaluated and return events of donors were calculated. Factors affecting first return after a donation (period of first return-PFR), yearly donation and return rates (YDR,YRR) were evaluated. Effect of deferral at the index donation (first donation during study period) on donor return behavior was evaluated by populating 2 groups from the donor sample (Group 1: Donors who were not deferred during entire study period, Group2: Donors who were temporary deferred during index donation, but not deferred thereafter during study period).

Results: 1417 (88.3%) allogenic donors were eligible and 188 (11.7%) were deferred from donating at the index donation. Majority of the donors were first time donors (77.1%) and males (71.8%). 11.7% of donors were deferred of which 9.7% were temporary deferrals. Commonest causes for temporary deferrals were low hemoglobin (27.7%), low blood pressure (13.8%) and high blood pressure (12.2%). Mean YDR is 2.87 (6.2SD), Mean YRR is 3.26 (6.3SD). Mean YDR and Mean YRR were both relatively higher in males, random donors (RDs), inhouse donors and donors who were accepted at index donation. Group 1 donors had higher mean YRR and YDR than Group 2 donors. YDR and YRR from highest to lowest was as follows: RDs who were never deferred during the study period > RDs who were deferred only during index donation > FTD who were never deferred during the study period > FTDs who were deferred during the index donation. Group 1 had a mean PFR of 185.77 days while Group 2 had a mean PFR of 141.80 days. Kaplan Mayer curves revealed RDs had a statistically significant earlier mean first return (139.6 days) than FTDs (202.9 days) ($p = <0.001$). Donors who had <1month deferral

had higher mean YDR(FTD-1.2706/RD-3.2958), YRR(FTD-1.9250/RD-3.4299) than donors >1month deferral.

Summary / Conclusions: Donor return to donation program was significantly affected by gender, acceptance or deferral at index donation, RD or FTD status at index donation and mode of donation. FTDs displayed significantly different return behavior when subjected to temporary deferrals compared to RDs. FTDs experienced a negative impact on their return rate due to both temporary deferrals and particularly, deferral periods exceeding one month. These findings highlight the importance of implementing strategies to minimize temporary deferrals, especially among FTDs. Immediate recruitment efforts following deferral periods and potential reductions in mandatory deferral lengths could demonstrably improve donor retention and overall program productivity.

P130 | Blood donor attendance - can a phone call make a difference?

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Background: Apheresis is a procedure in which whole blood is collected, separated in different components, one of these components is retained and the remaining are returned to the donor. An apheresis equipment that uses centrifugation is used to perform this type of collection. The most common use of this technology is collection of platelets unit. Platelets collected through this technology have some advantages over random donor platelets (RDP) because one apheresis platelet unit is usually enough for one recipient. This decreases the risk of transfusion-transmitted disease, alloimmunization rates and allergic transfusion reactions. In 2020, as COVID pandemic emerged in Portugal, blood banks had to implement strategies to maintain blood donor attendance despite the “stay at home” messages. In the São João Blood Bank in Oporto, donation of single-donor apheresis unit of platelets (SDP-apheresis) is previously scheduled by the nursing staff, mostly with more than one month in advance. During the pandemic, it was decided to reinforce this procedure, implementing an additional measure: a clinician should contact each donor of SDP-apheresis by phone, in the week before donation scheduling. The main objectives of this contact were to reassure donors regarding their fear of infection during blood donation (essentially COVID related), to remind the donor of his appointment and to question if there was any new situation that would prevent donation. Although the pandemic period passed, the procedure was maintained.

Aims: Evaluation of the impact of an additional measure that consists of making a phone call to the platelet donor one week before donation schedule.

Methods: We compare apheresis-platelet donation numbers in the three years before and after this measure implementation.

Results: From 2017 to 2019, the average donation of apheresis-platelet was 1036 donations/year. In the next three years (2020-2022), apheresis-platelet donation average was 1492 donations/year. These numbers correspond to an increase of 44.5% in SDP-apheresis donation.

Summary / Conclusions: This communication strategy reinforces the need for donor donation, increasing donor sense of commitment. On the other hand, knowing in advance the lack of availability of a donor, allows a better management of platelet collections. Simple strategies can optimize donor attendance and lighten the burden of those who need to manage blood components stock.

P131 | Knowledge and attitude of blood donation in young blood donors in lower middle income countries—first of its kind in country setup

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Background: Voluntary and replacement blood donation are explored in this study, with a focus on the safety and altruistic motivation associated with voluntary donors. The abstract addresses the challenges, such as misconceptions and negative attitudes, hindering voluntary blood donation. The importance of community-based interventions to improve knowledge and attitudes toward blood donation is emphasized.

Aims: The primary aim of the study is to assess the knowledge and attitudes of young blood donors, specifically targeting 12th-grade students in Bhopal, Madhya Pradesh. The study seeks to evaluate the impact of motivational lectures, delivered both online and offline, on the awareness and perception of blood donation among students. Additionally, the abstract highlights the need to analyze feedback and recommendations received from the participants.

Methods: The study involves community-based interventions, primarily motivational lectures conducted for 12th-grade students. Due to the COVID-19 pandemic, a combination of online and offline modes is employed. Consent is obtained from both students and their parents or guardians, and data is collected and securely stored in password-protected files. The abstract outlines the duration of the study (15–18 months) and the preference for government schools as the target audience.

Results: The results section presents key findings from the study. The survey sheet was taken just after delivering the motivational lectures. The feedback and response of participants to awareness towards blood donation was assessed. It reports a high level of awareness among students regarding blood groups and indicates a positive response to motivational lectures, with 95% of students finding them very useful. Specific feedback from participants is outlined, including

suggestions for incorporating more visual elements and personal stories related to blood donation. The section also provides statistics on students' personal connections to blood transfusions and donations within their families.

Summary / Conclusions: In conclusion, the abstract scores the efficacy of motivational lectures in creating awareness and shaping positive attitudes toward blood donation among young donors. It recommends further assessment of such programs in the first year of institutes, coinciding with the legal age for blood donation in India. The abstract also proposes educational visits to blood banks as part of school activities. Overall, it emphasizes the significance of community-based initiatives in fostering a culture of voluntary blood donation and ensuring a safe and healthy blood supply.

P132 | Major reasons of temporary deferrals of donors aged 18-23 in years 2019-2023

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Background: Blood service in Poland is based on voluntary and non-remunerated donations. Regional Blood Donor Centre in Poznań as well as other regional centres (total of 23) are the only entities authorized to collect, process, store and distribute blood and its components to hospitals in the region of their activity but they are also responsible to provide sufficient amounts of blood and its components. Regional Blood Donor Centre in Poznań is one of the largest blood centers in Poland with the total number of donations exceeding 100,000 per year. Regional Blood Center in Poznań collects blood in its main location, 13 satellite branches and also organises mobile unit collections, many of which take place in the donors' workplaces but also universities and schools. Taking into consideration the aspect of ensuring sufficiency in blood and its components and also from the marketing point of view it is essential to recruit young donors but also know the reasons why they are deferred from donating blood and its components.

Aims: The aim was to identify and analyse main reasons of deferrals of young donors from giving blood and subsequently modify the communication and marketing strategy targeted at them.

Methods: The analysis was made using the data obtained from the computer system 'Blood Bank' which is in operation in Regional Blood Centre in Poznań, Poland. We have analysed the total number of deferrals of donors aged 18-23 in the years 2019-2023.

Results: The total number of temporary deferrals in the age group 18-23 (both sexes) totalled 14,466 in years 2019-2023 which accounted for 23 % of all temporary deferrals. The major reasons in age group 18-23 were: low haemoglobin level (26.21%), no access to veins (7.11%), low blood pressure (6.25%), taking medications (4.64%), improper body weight (4%) and surgical procedures (3.13%). In the group of young men the results were: low haemoglobin level (14.59%), medications (6.15%), low blood pressure (5.78%), surgical

procedures (4.75%), skin diseases (4.62%) and improper virus screening test results (4.49%). In the group of young women the results were: low haemoglobin level (34.49%), no access to the veins (9.37%), low blood pressure (6.58%), improper body weight (4.60%), medications (3.56%) and tattoos (3.42%). Three major reasons for deferral were the same for both groups although they ranked in a different way: low haemoglobin level, medications and low blood pressure (rank 1, 2, 3 for men; rank 1, 3, 6 for women).

Summary / Conclusions: As it results from the analysis the effective communication (educational materials, content on the web site and in social media, topics discussed during educational meetings) with the young donors should be mostly targeted on aspects such as keeping good healthy diet, proper preparation for the donation and using medications. This communication should be differentiated for both sex groups. Bringing awareness to activities/situations with potential risk of virus infection is essential in both groups but probably with a bit more concern in the group of men (deferrals: surgeries—rank 4, positive virus test results—rank 6), whereas as far as the women are concerned more attention should be brought to ways of proper preparation for the donation but also on tattoos as a potential risk of virus infection (deferrals: rank 6 in women, not included in the top reasons in group of men).

P133 | Analysis of donors' deferrals due to positive results of virus tests in Regional Blood Donor Center in Poznań, Poland

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Background: Every donation is tested for the presence of antigen Hbs, antibodies anti-HIV1/2 and anti-HCV, presence of RNA HCV, DNA HBV, RNA HIV and the markers of Treponema pallidum infection. In case of positive virus test results the donors is deferred permanently from donating blood and its components. In such case donors collect virus test results in person and are referred to specialists for further treatment.

Aims: The aim of the thesis is to analyze the deferrals of donors of whole blood and its components that received positive virus test results.

Methods: The comparative analysis of donors with permanent deferrals in years 2022 and 2023 was carried out using data obtained from computer system Bank Krwi (system in operation in Regional Blood Center in Poznań, Poland), virus screening test reports regarding blood samples collected at the time of donation for every donor and reports confirming donors collecting their virus test results. The obtained data was divided into following categories: (1). positive result of the INNO-LIA test for the presence of the anti-HCV, no RNA HCV was detected. (2). RNA HCV was detected; screening test for the presence of anti-HCV was reactive. (3). HbsAg confirmatory test was positive. (4). DNA HBV was detected, anti-HBc test was positive. (5). RNA of the HIV was detected, the results of the anti-HIV 1/2 test were positive. The data was analyzed according to the kind of virus resulting in infection, donors' sex and their frequency of giving blood.

The source of infection was also identified using questionnaires completed by the physician when informing the donors about their positive test results. Data from 2023 was contrasted with the data from 2022.

Results: The number of deferred donors due to positive test results fell by 65%: from 26 in 2022 to only 9 in 2023. The analysis of the laboratory virus test results has shown: HCV infection was confirmed in 11 donors in 2022 and in 3 donors in 2023 (4 vs. 2 donors without the active replication of the virus). HBV infection was confirmed in 11 donors in 2022 (occult virus infection in 2 donors) and in 2 donors in 2023. There were 4 HIV infections in 2022 and 4 in 2023, which accounts for 15.4% of donors' deferrals due to positive virus tests results in 2022 and 44.5% in 2023. The number of deferred donors due to positive test results totaled 88% of deferrals due to all causes in 2022 and 67% in 2023 which is prevailing in the first-time donors. The number of men deferred due to positive test results totaled 65% in 2022 and 78% in 2023. 60% of the deferred donors due to positive test results claim they do not know the potential source of the infection, 25% indicate surgical, dental as well as cosmetic procedures and tattoos as the source of virus infection; 2% identify blood transfusions as the source. Remaining donors admit to having sexual contacts with multiple partners (which refers in 50% of the cases to the group of donors infected with HBC). 80% of donors with positive HBsAg claim they have not been vaccinated against HBV.

Summary / Conclusions: Such high number of virus infections in 2022 may be linked to the outbreak of the war in Ukraine. Because of that we have observed increased number of first-time as well as recurring donors (after a very long time) that made spontaneous decision to donate blood.

P134 | Abstract withdrawn

P135 | Understanding emotional drivers of blood donors—implications for donor retention and future donation intentions

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Background: Despite availability of generalized idea that higher level of fear in blood donors are associated with less donor retention, we do not know how different spectrum of emotions are related to willingness of blood donors to return back again.

Aims: This study recognizes intensity of 10 positive and 10 negative emotions live during donation and provides relationship between change in emotions and their association with willingness to return. Using the Positive and Negative Affect Schedule (PANAS scale), emotions were assessed at different stages of the donation process.

Methods: This observational time series study was conducted on first time whole blood donors over the period of 1 year in the blood centre of All India Institute of Medical Science. After donor registration and

informed consent at reception area, donor screening and examination was performed as per the national guidelines, and fit donors were asked to mark PANAS scale in the waiting area. Repeat PANAS scale was applied during phlebotomy and third time in the refreshment area. Before leaving the blood centre, all donors were asked question about their 'willingness' for return in the feedback form. The data was analysed using SPSS and R software and nonparametric tests were used to compare emotions of two groups of donors (with willing and not willing to donate) at different time points.

Results: Out of 360 donors, who successfully completed feedbackform, 83.3% showed willingness to return ($n = 300$, $CI = 79.0\%-87.0\%$). Friedman tests were performed to explore the statistical significance (at 95% confidence interval) for the changes in emotions from the waiting area to the various follow-up timepoints. Wilcoxon-Mann-Whitney Test was used to compare the two groups in terms of emotions at each of the timepoints. Donors who were willing to return back for donation, felt significantly *higher guilt* and *higher shame* in waiting area ($p = 0.046$, 0.040 respectively) and felt *higher nervousness* during donation in comparison to donors who are not willing to return back ($p = 0.026$). While emotions such as distress, excitement, upset, strength, fear, hostility and pride exhibited notable changes from before to after donations, yet these changes did not differ significantly between the groups willing and not willing to return for future donations ($p = 0.543$, 0.526 , 0.119 , 0.350 , 0.933 , 0.863 , 0.527 respectively).

Summary / Conclusions: Overall Positive emotions stay prominent throughout donation process in both the groups. Contrary to existing evidence, the experience of fear during donation does not seem to influence the intention to donate again. Rather than centering on categorizations such as 'negative' and 'positive' emotions, implementing strategies tailored to address sensations of guilt, shame, and nervousness in designated areas of the blood center can foster a more favorable inclination towards repeat donation.

P136 | Abstract withdrawn

P137 | Optimizing donor experience; strategies based on indicators

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Background: Society is in constant change. Understanding the factors that affect it at any given time is key to understanding citizen behavior and being able to influence it. That is why the Blood and Tissue Bank has always seen it as essential to inquire about donors' satisfaction and their expectations regarding donation.

Aims: Establish specific indicators of donor satisfaction and their expectations to develop continuous improvement actions that foster their loyalty and ensure repeat donations.

Methods: After each donation, and 48 h later, a survey is sent to the donor via email to evaluate the service provided. We focus on three

P137 – Table 1

	2021	2022	2023
Adverse reaction	1704	1683	1518
Complaints for adverse reaction	0.62	0.50	0.36
Total complaints	4.63	4.19	3.68
Complaints about staff treatment	1.16	1.12	0.94
Complaints about extraction	0.94	0.93	0.72

P137 Table 2

	2021	2022	2023
Surveys sent	283.951	294.436	319.044
Surveys answered	62.684	57.408	50.968
Regular donor NPS	86.25	86.25	86.72
Plasma NPS	84.19	85.16	84.58

aspects: Detecting if there has been an adverse reaction post-donation, allowing for a much broader hemovigilance control by providing personalized medical follow-up to all donors who experience any symptoms. Assessing service satisfaction, obtaining an NPS indicator. Donors rate their donation experience from 0 to 10 and indicate if they would donate again. Identifying specific donor complaints, which are addressed personally along with the responsible personnel at the donation center or mobile donation campaigns where they were attended.

Results: In 2023, the number of adverse reactions recorded post-donation has decreased. This is especially attributed to two improvement actions: (a) Upon identifying the most common reactions, specific informational material for donors was created, and staff was trained to reinforce this post-donation message. (b) Each recorded adverse reaction receives personalized follow-up from a BST doctor. As a result of the improvement actions, the rate of complaints for this reason has also decreased.

The complaint index has been decreasing over these three years. Improvement actions have been focused on the most critical areas according to donors; (a) Complaints related to the treatment of blood bank personnel have decreased following specific training adapted to donation service; (b) Each complaint is also handled personally between the donor and the service that attended to them.

Despite the decrease in survey responses, the NPS indicator has remained stable for regular donors. In the case of plasma NPS, there has been an increase in satisfaction, especially noticeable when broken down by centers, due to the training actions activated for 2023 in the marketing plan to attract new donors.

Summary / Conclusions: Maintaining continuous communication with the donor is essential to improve their satisfaction and ensure repeat donations. Thanks to the information collected through surveys sent to donors, it has been possible to monitor the mentioned indicators and apply improvement actions in areas of interest to donors. A reduction in the complaint index and improvement or maintenance of the NPS has been demonstrated.

P138 | Demographic change of blood donors in Montenegro

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Background: About 20 000 units of blood are collected annually in Montenegro. Nearly 60% of that blood comes from family/replacement donor who gives blood when it is required by a member of his/her family and 40 % from voluntary non-remunerated blood donors. As the whole society continues ageing, the demographic trend is getting older which could be great impact on blood donations and blood supply services

Aims: The aim of this study is to investigate and analyze the age distribution and demographic data of blood donors in the past seven years in Montenegro

Methods: The statistical data on blood donors and blood donations were compiled from the annual report of Montenegro. Blood Services from 2017 to 2024. The donor rate was calculated by the number of donors in per 1000 population. Donor rates were calculated and separately by sex and age

Results: The share of blood donors in the total number of donations for a certain year, classified by age groups 18, 19-23, 24-30, 31-40, 41-50, 50-65, was compared for a period of seven years from 2017 to 2024, and this was done in two categories by gender, specifically for women and specifically for men. The goal was to monitor blood donors by gender and age in order to gain insight into the structure of donors in Montenegro and to know in which direction we should go with further education and promotion of blood donation and to which age structure should be paid special attention. **Women:** 18 years from 0.83% in 2017 to 0.59% in 2023; 19-23 years from 1.17% in 2017 to 1.38% in 2023; 24-30 years from 1.30% in 2017 to 1.63% in 2023; 31-40 years from 1.80% in 2017 to 2.64% in 2023; 41-50 years from 1.87% in 2017 to 2.46% in 2023; 50-65 years old from 1.32% in 2017 to 1.82% in 2023. **Men:** 18 years from 1.40% in 2017 to 1.13% in 2023; 19-23 years from 4.76% in 2017 to 4.61% in 2023; 24-30 years from 13.45% in 2017 to 10.76% in 2023; 31-40 years from 28.45% in 2017 to 27.60% in 2023; 41-50 years from 24.11% in 2017 to 26.61% in 2023; 50-65 years old from 19.52% in 2017 to 18.75% in 2023

Summary / Conclusions: The blood donors in Montenegro are mainly in the age of 31-40 and 41-50 and when we look at the trend in the last 7 years we have a decline in the group of men aged 18, 19-23, 24-30, 31-40 and the growth in the age group of 41-50 which indicates the necessary work with donors especially in the younger categories. Share of donors older than 30 years is 83.5% and younger under 30 only 16.5% for 2023 and it is in decline as compared to 2017 where the ratio was 77% versus 23%. As for the female population when we look at the trend in the last 7 years we have a decline in the 18 year old group, a slight increase in the 19-23 and 24-30 group, more significant increase in the 31-40 group and the age group of 41-50 as well as

in the group of 50-65, which indicates the necessary work with donors, especially younger categories, but otherwise intensive work with the female population which barely exceeds 10% of the total blood units taken, which is a slight increase in relation to the period of 7 years ago when the share was above 8% but we should also take into account the fact that the share of women over 30 years old is 66% in 2023 and is increasing compared to 2017 where this ratio was 60%

P139 | Research on the analysis of key factors affecting blood donation based on interpretable deep learning methods

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Background: With the continuous and rapid growth of clinical blood demand in China, the contradiction between blood supply and demand is increasing, and the lack of blood source and poor public motivation to donate blood are still the main problems currently faced.

Aims: This study aims to explore the key factors affecting the amount of unpaid blood donation and their intrinsic relationship, and we combined the collection of big data, deep learning techniques, and their interpretability to conduct a comprehensive and in-depth analysis of the relevant variables. We successfully collected a total of 21,131 blood donation data related to the above variables for in-depth analysis. These data were categorized into training and test sets of 80% and 20% respectively. In this study, we have taken "Total Blood Donation" as the main object of study and used other factors (excluding Total Blood Donation and Number of Whole Blood Donations) as inputs for the unsupervised study.

Methods: We adopt a novel interpretable deep learning method, EVNet, which incorporates advanced dimensionality reduction techniques to effectively capture the intrinsic structural features of high-dimensional data, and retains the important attributes of the data with the help of the lasso loss function through a gate layer, thus realizing in-depth parsing and understanding of the original data. and understanding of the original data.

Results: By analyzing the importance of these features, we found that: honorary certification of the donor, frequency of blood donation, single blood donation and specifics of the most recent blood donation are the key factors influencing the amount of blood donated. In addition, socio-economic status, occupation and blood group also influence blood donation to some extent. (1). Availability of honorary card (1.0): This is the most important characteristic, implying that donors with honorary cards are likely to contribute the most to the volume of blood donated. This may be due to the fact that donors who have received an honorary card are more likely to be long-term and repeat donors. (2). Highest single donation of component blood (0.7897554): This indicates that the higher the single donation, the greater the contribution to the total blood donation. (3). Frequency of Blood Donation—Total (0.68018156): The frequency of blood donation is also a key factor. Frequent blood

donors contribute more to the total number of blood donations. (4). type of last blood donation (0.6700109) and volume of last blood donation (0.6524984): these characteristics indicate that the type and volume of last blood donation also play an important role in predicting future blood donation behavior. (5). Occupation (0.5488994): these socio-economic factors may influence an individual's willingness and ability to donate blood. (6). gender (0.25562042) and age (0.2851166): these basic demographic characteristics have some effect on blood donation but not as significant as other characteristics.

Summary / Conclusions: Entering the information age, the technology of collecting and analyzing big data provides unprecedented opportunities. This information can help blood donation centers and related organizations develop more effective strategies to encourage blood donation and optimize resource allocation. With the help of big data, this study collects a large amount of information about blood donors, their behavioral patterns, and other related data, thus providing rich materials for in-depth analysis.

P140 | Navigating donor deferral—pre and post-donation considerations in blood donation

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Background: In Pakistan, only 0.05% of people donate blood voluntarily. They need more motivation as they are screened well. Blood donation and transfusion saves lives in health emergencies. Good blood transfusion service is essential for health care. Developing countries give 60% of the 118.2 million blood donations in the world, but the rates vary. Safe blood donors and testing make blood transfusion safe. WHO says blood transfusion should use blood that matches and is free of HIV, HCV, HBV, syphilis, and malaria.

Aims: This study aims to find out the rate of deferral before, during, and after donation among healthy blood donors. This study also measured how common hepatitis B and C were in local blood donors and proposed how to make blood transfusion safer.

Methods: A total of 221 donors were meticulously selected based on the questionnaire criteria established by the College of American Pathologists (CAP) and American Association of Blood Banks (AABB) standards. During the blood collection process, all donors underwent screening for Hepatitis B, C, Immunodeficiency virus (HIV), Syphilis, and malaria in accordance with the guidelines issued by the World Health Organization (WHO). The screening was done using the Chemiluminescence immune assay (CLIA) method, which is known for its high sensitivity and specificity. Additionally, the malaria test was conducted using the Immunochromatographic Test (ICT) method, which is a rapid diagnostic test that has been shown to have high diagnostic accuracy. The rigorous screening process employed in this study ensured that only safe and healthy blood units were collected, which is of paramount importance in ensuring the safety of blood transfusions.

Results: The Study included 221 potential donors over the period of 1 year. 67.8% of donors were eligible for donation. 32.1% were

deferred for various reasons. The most frequent reason for temporary deferral was low hemoglobin (45%), followed by underweight, flu/allergy, high blood pressure (7%), and Low vein strength 12%. The most frequent reason for permanent deferral was hepatitis C, followed by hepatitis B and syphilis (5.3%).

Summary / Conclusions: The study findings reveal that a staggering 32.1% of all blood donations were deferred due to low hemoglobin levels and infections with hepatitis B virus (HBV) and hepatitis C virus (HCV). These alarming statistics necessitate the implementation of assertive measures to address the underlying causes of deferrals.

P141 | Abstract withdrawn

P142 | Revolutionizing blood donor screening—my GPTs AI-driven approach aligned with Thai Red Cross standards

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Background: The efficient and accurate screening of blood donors is crucial in transfusion medicine. Traditional methods, while effective, can benefit from the integration of artificial intelligence (AI) to enhance decision-making processes.

Aims: This study aimed to evaluate MyGPT, an AI model, for its accuracy and applicability in determining blood donor eligibility in line with the Thai Red Cross guidelines.

Methods: My GPTs was trained using a comprehensive dataset from the Thai Red Cross's blood donation manual. The model underwent testing with 20 diverse queries, encompassing various eligibility criteria, medical histories, and current health guidelines.

Results: The AI model achieved a 100% accuracy rate in these tests, correctly interpreting and applying the Thai Red Cross guidelines across all queries. This performance demonstrates MyGPT's robust understanding of the criteria and its potential as a reliable tool in the screening process.

Summary / Conclusions: MyGPT's exceptional performance in this study suggests significant potential for AI applications in transfusion medicine, particularly in enhancing the efficiency and accuracy of blood donor screenings. This technology promises to support healthcare professionals in making informed decisions, paving the way for more advanced, AI-assisted medical processes in the future.

P143 | The end of user fees to access healthcare in Portugal and the impact on blood donations - a single center experience

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Background: Before 2022, in Portugal, the only incentive offered to blood donors was an exemption from paying fees to access the National Health Service (SNS). After the 1st of June 2022, the

collection of user fees ended in almost all services of the SNS, which left blood donors without any monetary or non-monetary incentive.

Aims: We aimed to evaluate the effect of this measure on the number of blood donations in a Portuguese blood bank.

Methods: In this retrospective study, data from all volunteer blood donations performed in the Tondela-Viseu Hospital Center blood bank from January to December 2023 was analyzed and compared with the same period in 2019 (i.e. pre-pandemic). This data included the number of blood collections, total number of donors, first-time donors, and donor gender information.

Results: In 2019, a total of 2380 blood donors (59% male, 41% female) performed 3825 total blood donations in the blood bank from Tondela-Viseu Hospital Center. Of these, 401 were first-time donors (16.8% of the blood donors). In the same health unit, in 2023, the total blood collections were 3714, which represents a decrease of 2.9%. However, we recorded a 1.7% increase in the number of donors (2420 donors, with similar sex distribution to 2019). Likewise, the number of new donors also rose to 500 (20.7% of the blood donors).

Summary / Conclusions: We observed a reduction in the number of blood units collected in our center, but a growth number of blood donors, essentially due to first-time donors. This means that the decrease in blood availability was caused by a higher percentage of suspended and/or eliminated donors. The lack of prerequisite knowledge that is the main cause of these events can be linked to the increased number of first-time donors. In contrast to what was previously hypothesized, we cannot conclude that the end of incentives to access healthcare in Portugal had a negative impact on the blood supply. Donor recruitment measures appear to have been effective. It is essential to continue investing in training and disseminating information to these donor populations to avoid donations decreasing. Further studies must be performed to evaluate the national impact of this political measure.

P144 | Abstract withdrawn

P145 | Hospital-Based blood donation center's response to national blood shortage around COVID-19 pandemic

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Background: Blood shortage preparedness is essential for blood banks, but the COVID-19 pandemic challenged traditional supply chains. This study examined how a hospital-based blood donation center responded to the national blood shortage from 2020 to 2023.

Aims: To compare national red blood cell (RBC) supply trends with donations at a local hospital-based center from December 2019 to April 2023, focusing on periods of national shortage.

Methods: Blood donation data was analyzed for three periods: pre-COVID (December 2019-February 2020), initial COVID (March-June 2020), and post-initial surge (July-September 2020). Data from October 2020 to April 2023 was also reviewed, encompassing Delta and Omicron variant surges.

Results: The local center trends mirrored National RBC supply trends at the local center, particularly the March/April 2020 peak following the US Surgeon General's blood donation appeal. During the initial COVID surge, a major blood supplier announced a severe shortage, cancelling over 400 blood drives. Local donations significantly increased, with 136 and 189 units collected in March and April 2020 respectively, compared to 37 and 35 in January and February.

Summary / Conclusions: Hospital-based blood donation centers, though representing only 6% of US blood collection organizations, proved crucial in supplementing national blood supply during COVID-19 shortages. They helped mitigate the impact of reduced fulfillment from major suppliers and national supply chain disruptions. The findings highlight the valuable role of local centers in ensuring blood security during crises.

P146 | Blood donor characteristics and return behavior in Abu Dhabi population

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Background: Blood donors in United Arab Emirates are volunteer non- remunerated donors. Efforts have been done to recruit and retain young blood donors. This study evaluates the donor characteristics, and how these characteristics changed over time.

Aims: To describe characteristics of blood donors at Abu Dhabi blood bank over the course of 10years and evaluate the results of the recruitment efforts of young donors and retaining them.

Methods: Abu Dhabi blood bank is the main supplier of blood and blood components to all hospitals in the emirate of Abu Dhabi. Electronic Records from 2013-2023 were extracted from the blood bank information system and evaluated and compared using statistical analysis.

Results: 460491 blood donors presented to donate throughout the study period (2013-2023). 88.3% were male when only 11.7% were female. 72.3% of the donors fell in the age category of 26-45 years, 12.5% between 17 and 25 years. The number of repeat donors is increasing over the years to reach 50% of the total donors. It seems young donors are becoming repeat donors which goes in parallel with the increase of donors category aged 26-45 years old from 35.5% to 72%.

Summary / Conclusions: There is a change in donor demographics over time in our center. The young donors are retained as the number of donors of the upper age category is increasing, this is a proof that the young donors continue to donate. The main reasons for this retention being an increased awareness about importance of blood donation and having a pleasant and satisfactory first-time experience in donating blood. It also includes the efforts put in by the staff in calling and reminding donors to come for blood donation. Further studies are required to identify potential causes of low participation of female and new young donors.

P147 | Abstract withdrawn

P148 | Evaluation of the Military Blood Transfusion Centre's blood collection activities from 2018 to 2023

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¹collecte, ²distribution, ³laboratoire, centre militaire de transfusion sanguine, ⁴hopital militaire de tunis, hopital militaire de tunis, tunis, Tunisia

Background: The need for labile blood products is constantly increasing as a result of advances in medicine and longer life expectancy.

Aims: The aim of this study was to analyse collection activities and to study performance indicators and their evolutionary trends in order to optimise the contribution of the Military Blood Transfusion Centre (CMTS) to achieving national self-sufficiency in blood products.

Methods: This is a retrospective descriptive study conducted on the CMTS over a period of 6 years based on an analysis of annual activity reports from 2018 to 2023 involving total missions, blood donations in fixed and mobile sites.

Results: During this period, the blood collection service provided 79037 blood donations. An annual increase in blood donations was observed describing an ascending curve. CMTS's national contribution also passed from 9% in 2018 to 13% in 2023. Donations at mobile sites reached 65.8% comparative to 34.2% at fixed sites. The average number of outings was 143 missions/year. Over the last 6 years, collection yields have gradually increased, from 55 donations/outing in 2018 to 70 donations/outing in 2023. The number of blood bags collected at fixed sites increased from one year to the next, with an average of 4503 donations per year. The highest growth rate at fixed sites was observed in 2021 (+1.99%).

Summary / Conclusions: The efforts made by the CMTS to increase the donation rate are confronted to constraints linked mainly to the specific nature of the volunteers targeted. An action plan has been made since the end of 2018 to improve the care of military donors during mobile blood drives and to promote fixed-site donation with the aim of optimising the recruitment and fidelization of blood donors.

P149 | Analysis of the efficacy of blood donor mobilization via WhatsApp

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Background: The primary objective of the Blood Bank of La Rioja is to ensure an adequate number of blood donations to meet the transfusion needs of the Autonomous Community of La Rioja. In this context, it is essential to consistently promote blood donation to maintain a stable flow throughout the year, given the expiration of blood

P149 – Table 1

Municipality	2020	2021	2022	2023
A	5	10	2	8
B	3	5	2	9
C	6	3	5	7
D	4	10	8	3
E	7	0	6	8

components. However, there are occasions when reserves of certain blood groups decrease, necessitating actions to remind the population of the importance of blood donation during periods of reduced donor activity and when transfusion needs in our hospitals are sustained or increased.

Aims: In this study, we analyse the efficacy of a call to action conducted via WhatsApp to donors of the O- blood group who had donated blood at least once in the previous two years, carried out during the months of October and November of the year 2023.

Methods: To conduct this analysis, we compared the number of donors from this blood group who made blood donations in the municipalities where donors were notified via WhatsApp in the same months of the years 2020, 2021, 2022, and 2023.

Results: As can be observed in the following table, a significant increase in the number of donors is observed in all municipalities compared to the previous visit of the blood bank:

In most municipalities, there is an increase compared to previous years, except for Municipality D: Municipality A: Second-best record in four years. Municipality B: Best record in four years. Municipality C: Best record in four years. Municipality E: Best record in four years.

Summary / Conclusions: The utilization of instant messaging tools such as WhatsApp can be effective in reminding donors of the significance of their contribution, particularly during critical periods where donation activity decreases and transfusion needs increase. Increase in Participation: The results demonstrate a rise in the number of donors across the majority of analysed municipalities compared to previous years and the immediately preceding campaign. While there was a widespread increase in the number of donors, some variability is observed among municipalities. Certain municipalities exhibit significant increases compared to previous years, while others show more moderate growth.

P150 | Profile of blood donors in a Tunisian blood bank

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Background: Studying donor demographics and understanding the reasons for donor unsuitability can help develop intervention strategies to increase donation rates and address rejection issues.

Aims: The aim of our work was to describe the various profiles of donors, their motivation and the immediate complications associated with donation.

Methods: The study involved 1084 Tunisian blood donors in a hospital blood bank between June 2023 and November 2023. Questioning of blood donors (DDS) was conducted using a standardized questionnaire completed during the medical interview.

Results: The donors were mainly male (71.2%) and had a mean age of 37.9 years with extremes [18 years, 65 years]. Among the blood donors, 480 (44.3%) were first-time donors, while 329 (30.4%) had donated once, 112 (10.3%) twice, 64 (5.9%) thrice, and only 97 (8.9%) had donated four or more times. In 99.8% of cases, the donation is motivated by the fact that family members have needed a blood transfusion. During the study period, 14.9% (161/1084) were considered ineligible to donate. One hundred and forty-six of these donors were eliminated during the interview. The most common reasons for pre-donation exemption were chronic or acute pathologies in 38.4% of cases, and alcohol consumption within 24 h in 4.8% of cases.

Summary / Conclusions: Encouraging voluntary blood donation is essential, as is improving the targeting of interview questions in order to identify donors at risk of transmissible diseases.

P151 | A novel predictive algorithm for post-donation platelet count—a model software used at the point of care

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Background: According to the FDA, automated cell separators must ensure that donors reach a minimum post-donation platelet count (PLT post) of at least 100×10^3 platelets/uL. To this end, these machines possess proprietary internal algorithms that can only be utilized when the equipment is not in use; otherwise, they would not be accessible during the donor selection process, who await the final decision of whether they will be eligible for platelet (PLT) donation by apheresis, especially those donors who have lower platelet counts, between 150 and 200×10^3 platelets/uL.

Aims: To evaluate the performance of the novel algorithm to predict post-donation platelet count in a population of donors with platelet counts less than 200×10^3 platelets/uL.

Methods: A total of 207 predictive PLT_{post} counts of plateletpheresis obtained by the Trima Accel V.7 cell separator algorithm (Terumo BCT, USA) (Actual algorithm) and the novel algorithm were evaluated. All donors had pre-donation platelet counts between 150 and 200×10^3 platelets/uL. The target yield was 3×10^{11} in all cases. The setting of the Trima was minimal PLT_{post} count 100×10^3 platelets/uL. Formula of the novel algorithm PLT post: % Return of total PLTs ($\times 10^{11}$) = % Total donor platelets returned ($\times 10^{11}$) $\times 100$ / Total donor platelets ($\times 10^{11}$). PLT post **PRE NOVEL** = PLT pre ($\times 10^3$ platelets/uL) % Return of total PLTs ($\times 10^{11}$) / 100. PLT post **NOVEL** = PLT post **PRE NOVEL** $\times 15$ / 100 + PLT post **PRE NOVEL**.

P151 Table 1: Results of analytical performance evaluation of Novel Predictive Algorithm PLTpost

	Current Algorithm PLTpost	Novel Algorithm PLTpost	Bias % (CI 95%)	Desirable bias (%)*
Predictive PLTpost ($\times 103$ platelets/ uL)	139.59 \pm 15.61	142.37 \pm 10.85	-2.2 (2.77 to 1.67)	5.9

* Obtained from Desirable Biological Variation Database specifications - Westgard.

Results: 207 predictive PLTpost results were evaluated, of which 96.12% (198) were men. The average weight and height were 74.65 kg \pm 10.1 and 1.68 m \pm 0.07, respectively. The average PLTpost in the current algorithm was 142.37 $\times 103$ platelets/uL \pm 10.85 and in the novel algorithm was 139.59 $\times 103$ platelets/uL \pm 15.61, which was significant ($p < 0.05$). A very strong positive correlation was observed between the novel and the current PLTpost algorithm ($R^2 = 0.99$, $p < 0.05$). A linear regression analysis was performed obtaining the following equation: (Y) = 46.480 + 0.687 (X), where: (Y) is the current algorithm and (X) is the novel algorithm. Using a Bland Altman test found the mean bias of -2.77 $\times 103$ platelets/uL (CI 95% of bias: -3.48 to -2.06 $\times 103$ platelets/uL, $p < 0.0001$), while the percentage bias was -2.22% (CI 95% of bias: -2.77 to -1.67%, $p < 0.0001$). Regarding the evaluation of clinical relevance, the percentage bias was compared to desirable bias (-2.22% < 5.9%, respectively). Therefore, the bias found had no clinical relevance (Table 1). The novel algorithm had a sensitivity and specificity of 99.03% and 100%, respectively.

Summary / Conclusions: The novel algorithm was a statistically significant difference without clinical relevance; therefore, it can be used in donor care as external software, saving time in the donor selection process when cell separators are in use or are located in places far from the clinical interview site.

Blood donation—blood donor health

P152 | Seasonal and daily variation in haemoglobin in the Dutch blood donor population

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Background: Haemoglobin (Hb) is used as an important donor deferral criterium worldwide. Hb levels, however, do not only vary between but also within individuals, for example depending on time of year or day as observed by us and others. This variation may be of relevance particularly for those individuals whose Hb levels are around the deferral cut-off. Daily and seasonal fluctuation may be due to environmental factors (e.g. temperature), behaviour and homeostatic processes, but also related to observations that many processes in the body show intrinsic circadian rhythms with a period of approximately (circa) one day (diem).

Aims: We aim to characterise these fluctuations in Hb levels further by adding modelling and chronobiology expertise (the science of periodic phenomena).

Methods: In order to investigate the nature, size and significance of the Hb variation, nearly 7.5 million capillary Hb measurements of new, whole blood and plasma donors collected between 8 AM and 8 PM at the Dutch blood bank over the past ten years were included.

Results: Preliminary analyses show that part of the variation in capillary Hb levels can be explained by a non-linear association between Hb levels and time of day or year. In accordance with other studies, we observed higher Hb levels in the morning and in winter than in the late afternoon and in summer. We will further explore this variation, considering the impact of group differences as well as environmental and other factors that may play a role in the Hb variation. In addition, we will assess daytime and seasonal variation in other blood (count) parameters.

Summary / Conclusions: In the future, this knowledge could contribute to data-driven donor assessment at blood banks and the interpretation of clinical data.

P153 | Abstract withdrawn**P154 | Should we need to consider insulin resistance in candidates for apheresis procedures? An observational and prospective study.**

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Background: According to Mexican regulations, it is mandatory to provide post-donation meal; however, the guidelines are limited to volume and not nutritional specifications. Candidates living with diabetes are eligible to donate; but, 61% of the population is unaware of their condition. The infusion of citrate-dextrose solution in plateletpheresis and the lack of regulation in the nutritional quality of post-donation meal are factors that could potentially increase the risk of hyperglycemia in donors with insulin resistance (IR).

Aims: To assess the association of the HOMA index with post-donation and post-meal glycemic status in plateletpheresis donors.

Methods: A prospective cohort was designed with plateletpheresis donors randomly recruited at the Instituto Nacional de Cancerología-México, approved by IRB. Peripheral blood samples were collected at three time points: (A) pre-donation, (B) post-donation, and (C) 90 min post-meal for biochemical and hormonal analysis. Pre-donation anthropometric assessment and clinical history were obtained during interview. The post-donation meal (661 Kcal) was the Institute's standard, total intake was supervised within 30 min. IR was evaluated using the Homeostasis Model Assessment (HOMA). The lower tertile was defined as no insulin resistance group (NIR), while the middle and higher tertile were considered as exposure group, defined as the IR group. Multiple linear and logistic regression models were adjusted using acyclic diagrams.

Results: We recruited 105 plateletpheresis donors (70% male), with a median age of 34 years and a 12-h fasting period. Regarding HOMA index, NIR group showed values of 1.15 (0.96-1.33) and IR group showed 2.61 (2.1-3.8). Additionally, no differences we observed comparing NIR with IR groups in age, procedure duration, infused citrate, and time elapsed between meal and analysis at time C. The IR group had higher BMI, HbA1c, and triglyceride levels. In both groups, glucose levels increased at time B compared to A, and levels at B showed no changes compared to C. However, only in NIR group glucose returned to baseline levels at the end of the study. At time C, IR showed a maximum glucose concentration of 500 mg/dL, which was higher than in NIR with a maximum of 176 mg/dL. Insulin and C-peptide levels were consistently lower in NIR throughout all

evaluations. The logistic regression models showed that IR group had increased probability of having glucose levels >101 mg/dL at post-donation and post meal, compared to NIR group (OR: 4.70, 95% CI: 1.34-16.46, $p = 0.015$, and OR: 2.74, 95% CI: 0.97-7.77, $p = 0.057$, respectively). Linear regression models showed (β coefficient) an increase of 13.40 mg/dL in glucose levels per unit of HOMA (95% CI 8.08-18.71, $p < 0.01$) for the IR group at post-donation, and at post-meal, there was an increase of 15.77 mg/dL (95% CI 8.8-22.70, $p < 0.01$). Both models were adjusted for age, BMI, fasting time, infused citrate, and procedure time.

Summary / Conclusions: Insulin-resistant candidates (HOMA index >2.11) are 4.7 times more likely to present metabolic alterations in glucose levels after plateletpheresis compared to the NIR group. This effect was consistent with plateletpheresis and a 611 Kcal meal, yet only the NIR returned to baseline glucose levels. Considering prevalent risk factors in the Mexican population, candidates' metabolic unawareness, and post-donation recovery time, developing meals emphasizing caloric replenishment and a low glycemic index could be a viable strategy.

P155 | Returning genetic information to blood donors—hemochromatosis

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Background: The Blood Service Biobank is specialized in transfusion medicine related questions, such as blood donor health. Almost all (99.5%) of the biobank donors in Finnish Red Cross Blood Service Biobank have given their consent to receive health related findings. Post donation iron supplementation is routinely offered to all frequent donors and to women under 51 years, except donors with clinical hemochromatosis. HFE C282Y allele in homozygote form is the most common cause for hereditary hemochromatosis in Caucasian population and is recommended to return by American College of Medical Genomics.

Aims: The aim of the study was to return genetic risk information to blood donors and to prevent the provision of post donation iron supplementation in future blood donations.

Methods: 82 HFE C282Y homozygotes were identified in blood donor population, $N = 43,688$ based, on genotyping array results. Identified blood donors were informed of their genetic risk for hemochromatosis by letter and guided on further consultation and testing to health care. In addition, they were asked to return a form to Blood Service where they were asked if they had been aware of their genetic risk towards hemochromatosis, can the HFE C282Y status be stored in blood donor registry and if they wished to participate on further survey regarding receiving genetic information from biobank.

Results: 100% ($N = 82$) of the primarily identified HFE C282Y homozygotes were verified in independent laboratory with clinical grade method. 75.6% ($N = 62$) of the blood donors returned the form. Only 9.7% ($N = 6$) were aware of their genetic risk for hemochromatosis

prior the study. 100% ($N = 62$) gave their permission to record the HFE C282Y status to blood donor registry. 98.4% ($N = 61$) expressed their willingness to participate in a survey.

Summary / Conclusions: We demonstrate high occurrence of blood donors not being aware of their genetic risk for hemochromatosis. By avoiding excess iron supplementation in future blood donations, returning the genetic data may have an impact on donor health in the long run. In addition, we show that biobank material can be utilized for personalized blood donation policy. Moreover, the results highlight a high overcome in response rate and willingness to participate in further survey in the matter. We are currently conducting a survey for the blood donors regarding their experiences receiving the genetic risk information and whether they had expected to gain such an information. Furthermore, the aim of the survey study is to understand whether the donors had gained any short-term impact on prevention of iron accumulation.

P156 | PFAS levels over time in plasmapheresis and whole blood donors

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Background: Perfluoroalkyl substances (PFASs) were introduced to the environment in the fifties because of its desirable properties in non-stick or stain—and water resistant products and more. They accumulate in the environment and in the human body (half-life > 44 days–4years). PFASs with high certainty cause suppressed immune response, low birth weight and delayed mammary gland development, increased cholesterol levels, liver damage and kidney and testicular cancer.

Aims: To measure the level and possible development of the concentrations of several PFASs in blood and plasma donors over 4 month.

P156 – Table 1

Baseline (mean values)	HFPD, $n = 15$	RFPD, $n = 15$	Whole blood, $n = 14$
Age (years)	44.3 (9.8)	48.7 (9.8)	48.9 (12.4)
EBV (mL)	5940.0 (563.9)	5745.3 (483.2)	5758.8 (334.9)
PFOA, ng/mL	0.52 (0.17)	0.57 (0.20)	0.52 (0.21)
PFNA, ng/mL	0.23 (0.09)	0.34 (0.13)	0.29 (0.09)
PFDA, ng/mL	0.10 (0.05)	0.16 (0.07)	0.13 (0.05)
PFUDA, ng/mL	0.08 (0.06)	0.16 (0.10)	0.13 (0.07)
Sum PFHxS, ng/mL	0.39 (0.18)	0.51 (0.23)	0.53 (0.32)
Sum PFHpS, ng/mL	0.05 (0.02)	0.06 (0.04)	0.06 (0.03)
Sum PFOS, ng/mL	1.94 (0.93)	3.30 (1.98)	2.98 (1.87)

P156 – Table 2

PFAS diff pre-to post donation	HFPD, $n = 15$	RFPD, $n = 15$	Whole blood, $n = 14$
PFOA	−0.42*	−0.24*	−0.05*
PFNA	−0.18*	−0.11*	−0.01
PFDA	−0.07*	−0.05*	−0.01
PFUDA	−0.05*	−0.03*	−0.01
Sum PFHxS	−0.26*	−0.15*	−0.03
Sum PFHpS	−0.04*	−0.02*	−0.01*
Sum PFOS	−1.60*	−1.09*	−0.12

Note: * marks significant difference $p < 0.05$.

Methods: Pre and post donation blood samples stored from an ongoing RCT plasmapheresis project was sent to Dept Laboratory Medicine in Tromsø, Norway where PFASs was measured by ultrahigh pressure liquid chromatography tandem mass-spectrometry (UHPLC-MS/MS, Waters, Milford, MA, USA). Samples are secured from the individual donors before first project donation and post donations after 4 months. Furthermore, we have three RCT groups. HFPD donated plasma 14–24 times, RFPD 3–8 times and control donated WB 1 – 2 times. Only PFASs with measurable amounts were included in this study.

Results:

Summary / Conclusions: We have found a reduction in PFAS concentration that correlates with number of plasmapheresis donations. Also with whole blood donation the concentrations were decreased for PFOA and PFHpS. Regular plasma donors, donating within sustainable donation frequencies and donation volumes will have lower levels of PFASs. Moreover, for patients with problematically high PFAS levels, plasmapheresis is an effective treatment. Plasma from such patients should be discarded and not used for PDMPs.

P157 | The efficacy and effectiveness of eating and drinking interventions to reduce vasovagal reactions in blood donors - a systematic review of controlled experimental studies

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Background: Blood establishments guarantee the safety of blood donors by making maximal daily efforts to provide a comfortable donation process and minimize the risk of adverse events. Eating and drinking interventions are donor protection practices that are commonly used to prevent or reduce vasovagal reactions (VVRs).

Aims: To conduct a systematic review to identify, analyze, synthesize, and critically appraise the best available scientific evidence that investigates the efficacy and effectiveness of eating and/or drinking interventions before, during and/or after a whole blood donation on VVRs.

Methods: Six databases (The Cochrane Library, MEDLINE, Embase, CINAHL and Web of Science) and a trial registry (ClinicalTrials.gov) were searched until 11 May 2023. The inclusion criteria were: Population: whole blood donors. Intervention: consumption of food and/or beverages before, during and/or after a blood donation. Comparator: no intervention, placebo, or any other intervention. Outcomes: pre-syncope or syncope VVRs and related signs/symptoms. Design: (non-)randomized controlled trials (RCTs). Meta-analyses were executed to pool the effect measures across studies (where possible). The Grading of Recommendations, Assessment, Development and Evaluation (GRADE) approach was used to assess the risk of bias and overall certainty of the evidence.

Results: Six individual RCTs, 2 cluster RCTs, and 5 non-RCTs were included: Pre-donation water ingestion (250–500 mL) likely results in a large reduction in on-site VVRs, compared to no water (2 fewer per 100 donors, 7 studies, moderate-certainty evidence). This reduction was present in different subgroups (males vs females, first-time vs repeated donors) (low-certainty evidence). Pre-donation isotonic drink or water (500 mL) and a snack likely results in a large reduction in on-site and off-site vasovagal reactions, compared to regular practice (= advice to drink pre-donation and to eat a snack after the donation) (2 fewer per 100 donors, 1 study, moderate-certainty evidence). Pre-donation water (280 mL) and a gel cap containing sucrose with 250 mg caffeine may result in a moderate reduction in blood donor reaction ratings, compared to pre-donation water (280 mL) only (1 study, low-certainty evidence). Pre-donation sweetened lemon water (300 mL) with additional salt (2.5 g) may result in a large reduction in off-site vasovagal reactions, compared to sweetened lemon water (300 mL) only (1 fewer per 100 donors, 1 study, low-certainty evidence).

Summary / Conclusions: Pre-donation water ingestion or isotonic drink probably results in a large reduction in on-site and off-site VVRs. Pre-donation water ingestion with caffeine consumption or salt supplementation may result in a VVR reduction, compared to water ingestion only. Future large trials are required to increase the certainty of the effect of these and other interventions in the prevention of VVRs.

P158 | JAK2-V617F mutation among blood donors and donors with incidentally found polycythemia—a systematic review and meta-analysis

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Background: Although blood donors are generally healthy individuals, it is essential to acknowledge the potential existence of an underlying preclinical stage of a myeloproliferative neoplasm, particularly in blood donors with polycythemia. Identification of blood donors with polycythemia vera is valuable because the safety of blood donations from patients with myeloproliferative neoplasms is not established, and it provides an opportunity to advise these donors to seek medical attention for further evaluation and provision of appropriate management. There is currently very limited available evidence regarding the prevalence of polycythemia vera (PV) and Janus kinase 2 V617F (JAK2) mutation among blood donors, the appropriate upper cutoff for hemoglobin levels, or a consensus on the approach to such individuals.

Aims: This study aims to systematically review the available evidence on the prevalence of Jak2 mutation and PV among all blood donors and blood donors with polycythemia.

Methods: We conducted a literature search using two search engines, EMBASE and MEDLINE, from inception to August 14th, 2023. The studies that investigated the prevalence of JAK2 V617F mutation and/or polycythemia prevalence among blood donors with normal hematocrit and high hematocrit levels were included with no language restrictions. Risk of bias was assessed, and data were extracted. A random effects model meta-analysis was utilized to estimate the pooled prevalence and 95% confidence interval. Subgroup analysis was done to differentiate donors with normal hemoglobin vs those with polycythemia. Heterogeneity was evaluated by I² statistics. The study is registered with PROSPERO, number CRD42023456878.

Results: A total of 14 studies on 57,005 blood donors reported the prevalence of JAK2 mutation among blood donors with normal and high hematocrit (HCT). The overall proportion of JAK2 mutation among blood donors from all studies was estimated at 0.019 (95% CI 0.008 - 0.034). Subgroup analysis shows a prevalence of 0.027 (95% CI 0.002–0.073) among studies including donors with polycythemia only vs 0.012 (95% CI 0.003–0.026) among studies that included all blood donors. Only five studies reported the prevalence of PV among blood donors. The overall proportion of PV among donors was 0 (95% CI 0.00–0.00). However, the subgroup analysis indicated a proportion of PV diagnosis of 0.02 (95% CI 0.00–0.07) among blood donors with polycythemia.

Summary / Conclusions: This study reveals a very low prevalence of JAK2 mutations and PV among healthy blood donors; the slightly higher prevalence of these conditions among individuals with

polycythemia is not surprising. This finding may be somewhat reassuring from a blood bank perspective. However, further efforts are needed to establish the upper limit values of hemoglobin for donation deferral and to investigate pathways for those donors.

P159 | Results of an alternative donor deferral strategy that accounts for measurement variability

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Background: On-site deferral for low hemoglobin (Hb) levels poses a significant challenge for blood establishments worldwide, as they cause material wastage, and consume valuable staff and donor time. At most blood establishments, donors are deferred based on the result of a single pre- or post-donation test, not taking into account the individual's history or measurement variability. An alternative approach which uses the donor's average Hb value from multiple measurements can provide a more appropriate reference for assessing donor eligibility. In this method the historical mean Hb value is used to interpret individual measurements, thereby improving the ability to identify irregular outcomes (Janssen, Transfusion, 2022).

Aims: This study aims to quantify the benefits of an alternative deferral algorithm based on a donor's average Hb compared to currently applied strategies that use single pre- or post-donation Hb measurements only.

Methods: Using the algorithm proposed by Janssen (Transfusion, 2022), in which measurement variability is used to assess the deviation of individual Hb measurements from average Hb levels over a donor's career, we reassessed low-Hb donor deferrals in The Netherlands, Finland, Belgium, Denmark and South Africa. We quantify the effects of the algorithm by calculating how many on-site deferrals might have been avoided (where donors are deferred whilst their mean Hb is not below the deferral threshold) and how many donations would not have been allowed (where donors would have been deferred because their current Hb is above the threshold, but their mean Hb level is below the threshold).

Results: The preliminary findings presented in Table 1 demonstrate that implementation of the proposed algorithm would lead to a 66-98% reduction in deferrals across participating blood banks. The advantages of this revised policy are particularly pronounced for blood establishments utilizing pre-donation Hb measurements with high measurement variability. Conversely, our analysis reveals that, although rare, certain donors are genuinely ineligible for donation (demonstrating consistently low mean Hb levels), with occasional measurements above the eligibility threshold. Under the current donor deferral policy, these individuals are permitted to donate.

Summary / Conclusions: While the reduction in low-Hb deferrals with this algorithm vary per country, we show that a substantial reduction in donor deferral rates can be achieved in every setting. Moreover, this strategy offers the potential to identify donors who may be ineligible but are presently overlooked. There are also limitations to the proposed algorithm, in that there is an increased risk of un-noted inadvertent drops in Hb in individual (low Hb) donors. Further studies and development of a more advanced donor deferral algorithm is needed that allows finding the right balance between donor yield, donor health and donor risk.

P159 Table 1. Results of alternative donor deferral strategy compared to current strategy, confidence level for deferral 99%

Country	Measurement	Deferral rate (%)	Proportion unnecessary deferrals (%)	Proportion should not have donated (%)
The Netherlands (n = 1,163,750)	Pre-donation	5.3	76	0.0004
Finland (n = 832,845)	Pre-donation	2.8	66	0.0002
Belgium (n = 1,243,737)	Post-donation	5	68	0.0024
Denmark (n = 262,036)	Post-donation	3.5	68	0.0001
South Africa (n = 3,538,786)	Pre-donation	19	98	0.0049

P160 | A study of iron status of premenopausal first-time female donor associated with subsequent hemoglobin deferral

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Background: The report of the National Nutrition and Health Survey in Taiwan (2012-2015) revealed that there were nearly 1/4 premenopausal females with anemia or iron deficiency (ID), for whom pregnancy and menstruation were the dominant causes. Blood donation in this population could increase the risk of ID and iron deficiency anemia (IDA). According to the annual report of the Taiwan Blood Services Foundation (TBSF) from 2021 to 2022, low hemoglobin (Hb) deferral accounted for about 49.2% of all reasons of deferral before blood donation and approximately 85% of them were female. It is known that iron deficiency is the main cause of low Hb deferral, therefore, understanding the iron status among first-time premenopausal female blood donors facilitate the development of strategies mitigating low Hb deferral caused by blood donation.

Aims: Our primary objective was to assess whether different iron status of first-time premenopausal female blood donors was associated with low hemoglobin deferral on return donations.

Methods: Copper sulfate screening passed first-time premenopausal female blood donors aged 20-50 who were eligible for and underwent whole blood donation were recruited. Their blood samples were collected at the commencement of phlebotomy. Serum samples were analyzed for serum ferritin (SF) by Beckman Coulter AU5800 and whole blood samples were analyzed for complete blood count (CBC) including Hb by Sysmex-XN 1000. The routine method for Hb screening before blood donation was copper sulfate method and the records of low Hb deferral were followed. Nonanemic group defined as Hb ≥ 12 g/dL and divided into three sub-groups: Normal group (Nor) defined as SF ≥ 26 ng/mL; Low ferritin group (LF) defined as SF between 12 and 26 ng/mL; Absent iron stores group (AIS) defined as SF < 12 ng/mL. Anemia group defined as Hb < 12 g/dL and divided into two sub-groups: iron deficient anemia (IDA) defined as SF < 12 ng/mL; non iron-deficient anemia (Non-ID anemia) defined as SF ≥ 12 ng/mL. Statistical analysis was performed using IBM SPSS Statistics version 24.

Results: From March 2018 to January 2021, a total of 2696 first-time female blood donors were recruited. Iron status in Nor ($n = 1699$), LF ($n = 595$), AIS ($n = 262$) groups accounted for the proportion 63.0%, 22.1%, and 9.7% respectively. Although all of participants passed the copper sulfate method, there were still 140 participants (5.2%) with Hb < 12 g/dL by CBC method, of these 79(2.9%) were IDA and 61(2.3%) were Non-ID anemia. As of June 30, 2022, there were 5093 returned donations and 631 low hemoglobin deferral (copper sulfate method) records had been followed. The rates of low Hb deferral were 7.9% (239/3014) in Nor group, 13.4% (173/1287) in LF group, 24.0% (137/570) in AIS group, 43.7% (52/119) in IDA group, and

29.1% (30/103) in Non-ID anemia group. In nonanemic groups, participants with AIS had a higher risk of low hemoglobin deferral (OR = 3.39, 95% CI: 2.49-4.61) than those with Nor.

Summary / Conclusions: Low Hb deferral is associated with iron status among first-time premenopausal female blood donors. According to iron status, we can develop different strategies to reduce the incidence of low Hb deferral and retain blood donors, such as iron supplements or extending the minimum interdonation interval.

P161 | Evaluation of the impact of a donor management system on donor deferrals for low haemoglobin at the Southern Zonal Blood Centre, Ghana

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Background: A major barrier to blood transfusion services in sub-Saharan Africa is the chronic non-availability of adequate blood supply. Blood donor deferral costs both donors and the blood service valuable time and resources, in addition to losses to blood supply. In Ghana, the estimated prevalence of anaemia is 42.4% in women of reproductive age and 18.8% among peri-urban men, higher than the estimated global prevalence of 30.2% and 13.7% respectively. Also, 17.2% of all prospective donors in southern Ghana are deferred, and of these 35.3% are due to low haemoglobin (Hb). Key to reducing the negative impact of donor deferral due to low Hb is an effective donor management system. At the Southern Zonal Blood Centre (SZBC) of the National Blood Service Ghana, individuals deferred due to low Hb are referred to the Clinical Services unit for counselling and management. The Hemocue method is then used to confirm exclusion (Hb < 12 g/dL for females and < 13 g/dL for males) from donation or otherwise. Those with confirmed low Hb are further evaluated clinically to establish the cause of anaemia. Individuals found to be iron deficient are treated at the unit, and those with other causes of anaemia are referred to a specialist.

Aims: This study assessed the impact of the donor management system on the management and return donation attempts among donors who were deferred for low Hb.

Methods: We conducted a retrospective analysis of data on all potential blood donors. Who presented at the static donor clinic of the SZBC from January to December 2023, who were found to have low Hb using Copper sulphate method, and were referred to the Clinical Services unit for counselling and management. A follow-up review of the blood donor database was also made to determine the proportion of post-treatment blood donation attempts among those diagnosed and treated for iron deficiency anaemia.

Results: Among 2572 persons who attempted to donate blood at static clinic located at the SZBC, 329 (14.7%) were deferred for various reasons. Of these 60 (18.2%) were deferred for low Hb and referred for counselling and management. Thirty-seven (61.6%) of the referred individuals reported to the clinical services unit. Among those reporting to the unit (aged 17 and 56), 48.6% were females, 51.4% were males;

32.4% and 67.6% were first-time and repeat donors, and 10.8% and 89.2% were voluntary and family replacement donors respectively. After counselling and further clinical assessment, 20 (54.1%) of those reporting were found to have iron deficiency anaemia and managed with iron supplementation and nutritional counselling. A follow-up review of the blood donor database showed that 7 (35%) of the 20 donors managed for iron deficiency, all repeat blood donors, subsequently attempted to donate. Of those who tried to donate again, 5 (71.4%) were successful and 2 were deferred on account of low Hb.

Summary / Conclusions: The study has shown a potential of the existing donor management system to reduce losses to the blood supply by increasing the proportion of deferred donors who return to donate blood after correction of iron deficiency anaemia. Strengthening and rolling out of the existing system for effective donor notification, referral, counselling and management across the catchment area of the SZBC is recommended. Further research into factors that affect donor follow-up on referral is key to improving the proportion of referred donors that report for counselling and management.

P162 | Familial pseudohyperkalemia among Danish blood donors—a large-scale study of prevalence and impact of donation on donor health

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Background: Familial pseudohyperkalemia (FPHK) is a variant of the genetic disorder hereditary stomatocytosis. FPHK can be associated

with macrocytic anemia but is often asymptomatic—the only observable feature may be leakage of K⁺ from red blood cells ex-vivo at low-temperature storage. In transfusion medicine, this is problematic due to the risk of transfusion-induced hyperkalemia and the potential risk of donation-induced anemia.

Aims: We aimed to determine the prevalence of FPHK in Danish blood donors and to investigate differences in hematological parameters among carriers and non-carriers including the effect of donation on red blood cell count (RBC) and mean corpuscular volume (MCV).

Methods: The study is based on the Danish Blood Donor Study. Upon inclusion, participants are chip genotyped (Infinium GSA, Illumina). Imputation quality was assessed by INFO score and compared to danMAC5—a cohort of 8671 whole genome sequenced Danes. Four of six published FPHK variants were available. Hematological measurements (Sysmex XE-2100D, Sysmex Corp., Kobe, Japan) were available from two Danish regions. Data on donation history were available from time of inclusion. Sex and age were collected from public registries. Carriers were combined into a case-group and compared to non-carriers using descriptive statistics. We used regression models to assess the effect of carrier status on RBC and MCV at inclusion adjusted for sex and age. Similarly, we assessed the effect of donation on RBC and MCV adjusted for sex, age at inclusion, number of donations and baseline RBC/MCV values.

Results: Prevalences of the FPHK variants are presented in Table 1.

No differences in demographics, number of donations after inclusion, RBC or MCV at baseline or at latest donation were found in carriers vs. non-carriers (Table 2). Carrier-status had no significant effect on the changes in RBC and MCV over time ($p = 0.75$ and $p = 0.84$, respectively).

Summary / Conclusions: We found an overall prevalence of FPHK-carrier status in Danish blood donors of 2.90%. The prevalence for rs57467915 is 10-fold higher than previously reported (Andolfo, *Hematologica*, 2016), but is in accordance with the danmac5 cohort. We did not find an effect of carrier status on hematological parameters at inclusion or any adverse effect of donation on RBC and MCV. These preliminary data suggest that FPHK is a common phenotype in Danish blood donors and that healthy carriers can safely donate. We will proceed to investigate the implications for blood product quality and for recipients.

P162 Table 1

Prevalence of variants in DBDS, N = 100,180. ABCB6 SNPs, ascension #	INFO score	Carriers, no. (%)	Minor allele freq. (%)	Minor allele freq. in danmac5 (%)
rs148211042, R723Q, "Cardiff"	0.86	236 (0.24)	0.12	0.16
rs754667801, R375Q, "Lille/Flemish"	-	0 (0.0)	0.00	-
rs57467915, R276W, "Irish"	0.90	2668 (2.7)	1.34	1.4
rs61733629, V454A, "Bolivian"	-	0 (0.0)	0.00	<0.03

P162 Table 2

Characteristics of donors with complete sysmex data, N = 12,031	Non-carriers, N = 11,678	Carriers, N = 353	p
Female sex, no. (%)	6582 (56)	187 (53)	0.23
Age at inclusion, years, mean (SD)	30.8 (11.8)	30.8 (11.5)	0.92
No. of donations since inclusion, mean (SD)	14.0 (10.9)	14.3 (12.1)	0.67
Hematological values, mean (SD)			
RBC count at inclusion, $\times 10^{12}$ /L	4.79 (0.38)	4.81 (0.37)	0.32
RBC count at latest donation, $\times 10^{12}$ /L	4.80 (0.39)	4.82 (0.39)	0.31
MCV at inclusion, fL	89.5 (3.77)	89.4 (3.45)	0.87
MVC at latest donation, fL	90.4 (3.94)	90.4 (3.51)	0.72

P163 | Significant lower Reticulocyte Haemoglobin Equivalent (Ret-He) was found in blood donors with iron deficiency

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Background: Blood donation may cause iron deficiency especially in regular blood donors. In Indonesia, laboratory donor selection for eligibility relies solely on acceptable haemoglobin level 12.5 g/dL, but this may not exclude latent iron deficiency. Some tests can be used to diagnose iron deficiency, such as ferritin level or iron status which may not be suitable as routine examinations in the blood donation setting. Serum ferritin can reflect body iron stores, but it is time-consuming and expensive. Ret-He can depict actual iron availability faster and more affordably, making it a potential option for detecting iron deficiency in donors.

Aims: To analyze the relationships between Ret-He and iron deficiency in blood donors

Methods: The subjects were whole blood donors at the Dr. Sardjito Hospital Blood Establishment, Yogyakarta, Indonesia. Laboratory tests were performed for complete blood count and Ret-He using Sysmex hematology analyzer, serum ferritin, serum iron, transferrin saturation, unsaturated iron-binding capacity (UIBC), and total iron-binding capacity (TIBC). CRP level ≥ 5 mg/L was used to exclude donors experiencing inflammation.

Results: After excluding 13 subjects caused by CRP level, a total of 137 blood donors consisted of 109 (79.56%) males, and 28 (20.44%) females were recruited in the study. 55 (40.15%) subjects have been donated blood for ≥ 10 times. The mean age of subjects was 39 ± 11 years, with the predominant age group of 36-45 years (32.8%). Based on serum ferritin level ≤ 30 μ g/L, 13.9% of donors were in an iron-deficient state. The median of Ret-He value in the iron-deficient group (29.9 [23.90–31.30]pg) was significantly lower than in the non-iron-deficient group (30.70 [20.40–33.10]pg) ($p = 0.007$). Using a transferrin saturation cut-off of $< 20\%$, 24.1% donors were iron deficiency, and there was a significant lower Ret-He compared to the non-iron deficiency donors (29.9 [23.10–32.7] pg vs. 30.8 [20.4–33.10] pg)

($p < 0.008$). The female donor group showed a higher proportion of iron deficiency compared to males (29.2% vs. 20.2%). Ret-He had a significant positive correlation with serum ferritin ($r = 0.302$; $p < 0.001$) and transferrin saturation ($r = 0.218$; $p = 0.010$).

Summary / Conclusions: Ret-He was significantly lower in donors with iron deficiency, had a significant positive correlation with serum ferritin and transferrin saturation. As Ret-He examination can be easily and efficiently performed, it might be considered for use as an additional examination in the selection of blood donors or as an early screening for iron deficiency dedicated for regular blood donors. Further multicenter studies with more heterogeneous subjects and stricter exclusions to avoid other factors that may affect Ret-He is recommended.

P164 | Donor awareness on taking iron supplementation after blood donation in Singapore

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Background: Low haemoglobin levels pre-donation is the leading cause of blood donor rejection in Singapore. Despite efforts to provide post-donation iron supplements to replace donation-induced iron loss, this persisting issue emphasizes the need to assess the perception of blood donors regarding iron supplementation after donation.

Aims: Our survey aimed to evaluate the blood donors' knowledge and awareness of iron supplementation after donation, with the objective being to optimize the blood donation experience and improve overall well-being among donors.

Methods: The survey was conducted from March to December 2023. Posters with a QR code to access the online survey were placed in the post-donation refreshment area. The survey has a total of 15 questions on donor demographics, interaction with donor centre staff, compliance with iron supplementation and sources of information on iron supplementation.

Results: A total of 1865 donors participated in the survey, 58% were male and 7.1% were first time donors. 34.7% were aged between 31 and 40 years, 26.6% between 21 and 30 years, 24.6% between 41 and 50 years, 10.9% were >50 years and 3.2% between 16 and 20 years of age. 661 (35.4%) respondents reported a history of low haemoglobin; 71.7% ($n = 474$) were female and 96.4% ($n = 637$) were repeat donors. Among the different age groups, donors >50 years old have the highest low haemoglobin rate (42%). 392 (21%) respondents discussed low haemoglobin with donor centre staff, and 355 (90.6%) found the importance of iron supplements well-explained. Among those who spoke to donor centre staff, 82% ($n = 322$) had a history of low haemoglobin, and 13% ($n = 51$) were advised to consult a primary care physician. Only 910 (48.8%) respondents took the iron supplements provided by the donor centre (10 tablets), 61% took it post donation, 20% before donation, and 19% sporadically. Of these, 46.8% ($n = 426$) had a history of low haemoglobin. 256 respondents with history of low haemoglobin who took iron supplements finished the iron supplements and 132 requested additional iron or purchased their own. Most respondents took iron supplements 2-3 times per week (42%) which is recommended by blood centre, daily (28%), and weekly (12.8%). 17.1% only took one dose. 35.6% ($n = 235$) of respondents with a history of low haemoglobin did not take iron supplements. The commonest reason for not taking iron supplements is the perceived lack of need for supplemental iron followed by preference for dietary sources, forgetfulness, and concerns about side effects. Sources of information on the importance of taking iron supplements were donor centre staff, educational leaflet, and the internet.

Summary / Conclusions: The survey results highlight the need for additional measures to emphasize the importance of iron supplementation in blood donors. Measures include targeted educational and awareness campaigns leveraging social media platforms to inform blood donors about the importance of maintaining adequate iron levels (even in donors with normal haemoglobin levels) and the impact of iron deficiency on overall health and its potential consequences. Personalized counselling sessions can be offered to donors identified as at-risk for iron deficiency. Integrating follow up communication into the donor mobile application to remind donors of the importance of maintaining iron levels. By implementing these strategies collectively, we aim to improve awareness and enhance compliance with iron supplementation among blood donors.

P165 | Management of anaemia in donors of whole blood at a tertiary level hospital in the region of Madrid

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Background: More than 7500 people visit every year the Donor Unit of Alcorcon Foundation University Hospital to donate whole blood. Around 900 donors are rejected and 12% of those are due to low capillary haemoglobin level (cHb). Currently these rejected donors have no follow-up. This translates into worried and unsettled donors, increases in the cost and morbidity due to the delays in diagnosis of potential pathologies such as colon cancer, and the loss of potential future donations with the impact that this has on the number of donors, already stretched due to the aging population.

Aims: Introduce a new healthcare circuit for an early detection, prevention and treatment of anaemia in whole blood donors that will reduce the number of rejections or their transformation into plasma donors. Improve the quality of the service given to donors and the management of medical resources. Support the European "SUPPLY" program focusing on improving self-efficiency of derivative plasma products. Support one of the world nutritional aims of the World Health Organization for 2025, to reduce anaemia in the population and is aligned with one of the objectives of the agenda 2030 for the Sustainable Development of the United Nations Organization focused on managing anaemia in woman between the ages of 15-49.

Methods: Description of a new healthcare circuit implemented in November 2023 in a third level hospital of the Community of Madrid.

Results: The circuit has the following steps: Determine cHb: After an interview and the initial recognition of the donor, a point of care analyser is used to determine cHb. If cHb is <12.5 g/dL in women and < 13.5 g/dL in men, two more measurements are taken and a mean is calculated. If the donor has anaemia (determined in women with <12 g/dL and in men <13 g/dL), the nurse analyses previous haemoglobin results. Further enquiries are made to determine the possible cause and the haematologist is notified. Medical evaluation: The haematologist reviews the medical history and interview the donor looking for risk factors (poor diet/veganism, malabsorption, blood losses...), and finally do an individual approach: o Increase the time between donations for frequent donors. o Personalize diet recommendations with iron-rich food. o Promote plasma donation - in women with cHB >12 and men >13 g/dL. o In women with cHB<11 and men <12.5g/dL further tests are carried out to determine hemogram, creatinine, vitamin B12, folic acid, ferrokinetics test. Furthermore the donor is referred to

a general practitioner (GP) to invest etiology and treat. From the implementation on November 1 2023 to February 8 2024, the number of donors who visit the donation unit was 1759, but 310 were rejected for different reasons, including 19 people who had low levels of cHB. Of the latter, 100% received dietetic recommendations. Likewise, 6 of them were taken blood tests and were referred to a GP, of which 100% received oral ferrotherapy and a colonoscopy was requested due to rectal bleeding in one patient.

Summary / Conclusions: Anaemia in whole blood donors is a concern for both the donor as it could underly health issues and to the blood donation services as it reduces an already limited number of donations. Through early intervention, both could be addressed. A new protocol was introduced in November 2023 that offers health professional the ability to early detect and treat anaemia. Although the full effects of this new protocol are yet to be realized, the early signs are encouraging.

P166 | Characteristics and contributions of high-frequency plasma donors in The Netherlands

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Background: The Netherlands, as most European countries, does not collect enough plasma to be strategically independent, but aims to increase collections. Both male and female plasma donors may donate every two weeks up to 26 donations per (calendar) year. High-frequency plasma donation is associated with risk of declines in immunoglobulins and other plasma proteins, with unknown health effects.

Aims: The aim of this study was to identify and characterize high-frequency donors, and to evaluate their contribution to plasma collections.

Methods: First, we selected all plasma donors from 2023 that donated only plasma since 1-1-2022 and made four groups based on the average annual number of donations in 2022 and 2023: Low Frequency (LF) 1-6, Medium Frequency (MF) 7-12, High Frequency (HF) 13-18 and Very High Frequency (VHF) >18. Secondly, we selected all donors who made more than 100 donations in the last 20 years.

Results: In 2023, 73,101 donors (52% female) made 363,504 (successful) plasma donations (56% by males), on average 4.9 times. Only 4.1% of plasma donors were HF or VHF. Almost 1.6% of the male donors were VHF, and only 0.3% of the female donors. LF donors were generally younger (44 ± 15 for females and 47 ± 15 for males) than VHF donors (54 ± 15 for females and 58 ± 13 for males). Both the drawn volume (682 ± 140 vs 735 ± 118) and the estimated blood volume (4.9 ± 0.8 and 5.4 ± 0.8) were lower in LF donors compared to VHF donors. The average Hb level and deferral rate for low Hb seemed to not differ between the groups. Only 20% of VHF donors came to the donation center on the same weekday for all their donations and 28% on the same part of the day (morning, afternoon, evening), whereas almost 49% of the LF donors came on the same weekday and 66% on the same part of the day. Although the

percentage of VHF donors is low (0.9%) their contribution to the total amount of plasma collected from donors is 4.0% of the number of donations and 4.2% of the collected volume in 2023. In the past 20 years a total of 13,569 donors (74% male donors) made more than 100 plasma donations, 67% of these donors was still active. Their average age was 64 ± 10 years. The average Hb at these donors' first donation (9.4 ± 1.0 for males and 8.5 ± 1.0 for females) did not differ from their last donation (9.4 ± 0.8 for males and 8.6 ± 0.7 for females). Only 5999 of these donors donated only plasma in 2023 and 80% were in the LF or MF group in 2023.

Summary / Conclusions: In the Netherlands the number of VHF plasma donors is low, especially for female donors. Their contribution to the total number of collected plasma donations is equally low, but their donated volume is 4.2% of the total. This corresponds with the higher likelihood for VHF donors to be male, older, have higher estimated blood volumes, and thus donate larger volumes of plasma per donation. This preliminary study shows that only few donors donate at high frequencies, but with relatively high contributions to plasma collections. More equally distributed donation frequencies may be warranted to better protect donor health while maintaining supply.

P167 | Abstract withdrawn

P168 | Association of blood donation with the risk of cardiovascular disease in whole blood donors

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Background: The reduction in blood viscosity and iron store has been proposed to be associated with the reduction in the risk of cardiovascular disease (CVD). However, the results of epidemiological studies regarding to whole blood donations and the risk of CVD remain inconsistent.

Aims: To investigate whether regular whole blood donation decreases cardiovascular risk.

Methods: We conducted a retrospective follow-up study. We identified whole blood donors with a donation duration of more than 10 years from Taiwan Blood Services Foundation (TBSF) blood management system (BMS) database. A "10-year qualification period" was used, during which the number of whole blood donations was counted in this qualification period. To minimize health donor effect (HDE) bias, we included only those donors who remained active in donating during the 10th year. The health status of these donors was examined through linkage with the Taiwan National Health Insurance (NHI) databases. The NHI program's coverage rate for Taiwan's 23 million citizens reached 99.0% after 2005. The NHI database includes complete outpatients visits, emergency visits, and hospital admissions. The primary outcome was hospitalization for

cardiovascular, cerebrovascular, and peripheral vascular diseases. Donors were followed from the end of the qualification period until the occurrence of the events of interest, death, or the end of follow-up (2021/12/31), whichever came first. We performed Cox's proportional hazards model to estimate the hazard ratio (HR) and confidence interval (CI), assessing the association between high donation frequency and the risk of CVD hospitalization.

Results: A total of 371,791 male and 270,830 female donors were included in the analyses. The median years of follow-up was 11.38 years (Q1-Q3, 7.16-15.28). The rate of CVD hospitalization was 3.31 and 1.90 per 1000 person-years for males and females, respectively. After adjusting for first donation age, first donation year, body mass index at the end of qualification period, geographic regions, and a disease history of hypertension and diabetes, the risk of CVD hospitalization in male donors with equal or more than 10 donations in the qualification period, compared to those with fewer than 10 donations, was 0.94 (95%CI: 0.91-0.98, $p = 0.0009$). For female donors, the risk was 1.00 (0.96-1.06, $p = 0.7634$), indicating no significant difference. Sensitivity analyses using propensity score matching revealed a consistent result over the specified time frame.

Summary / Conclusions: The study results provide evidence supporting that frequent whole blood donation is associated with a diminished risk of CVD hospitalization in male donors

P169 | Are Canadian blood donors more likely to have COVID-19 vaccination than the general population?

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Background: Blood donors world-wide were indispensable for monitoring anti-SARS-CoV-2 antibodies generated by infection and vaccination during the pandemic. Vaccination against SARS-CoV-2 and other seasonal infections such as influenza reduces both the incidence and severity of infections. Because winter respiratory infections can impact on donor availability, it is important to understand the level of protection within the donor base. However, donor vaccination behaviours are under-studied.

Aims: We aimed to compare the percentage of Canadian blood donors with SARS-CoV-2 vaccination antibodies with the percentage of the general population who received at least one dose of vaccine each month during initial vaccine deployment. We also report donor attitudes towards SARS-CoV-2 vaccination.

Methods: Canadian blood donors were randomly selected for SARS-CoV-2 antibody testing over 2021 ($N = 165,240$). The percentage of donor samples with vaccination antibodies were compared with the percentage of general population who received at least one dose of vaccine in each month of 2021 except February. A random sample of

Canadian blood donors were surveyed about vaccination intent and attitudes ($N = 4558$ participated, 30.4% response rate).

Results: The percentages of the general population vaccinated and donors with vaccination antibodies increased from 1% to over 90%. General population vaccination was greater early in vaccine deployment than donors ($p < 0.05$), greater in donors than the general population by mid-2021 ($p < 0.05$) but they were similar by the end of 2021. While 52.6% of surveyed donors had received vaccine in May 2021, a further 41.1% intended to when eligible. Most donors thought COVID-19 infection could be serious (83.5%) and that it was important to be vaccinated even if previously infected (77.8%).

Summary / Conclusions: Early pandemic vaccine prioritization to at-risk individuals and healthcare workers gave rise to higher general population vaccination percentages, while donors had higher vaccine antibody percentages as vaccine was deployed to progressively younger age groups. This is consistent with observations of seasonal influenza vaccination. Since blood donors may be more willing to receive vaccination, they may be at lower risk of infection, and if they are infected may have shorter duration of illness. However, as less than a quarter of the general population had the SARS-CoV-2 booster shot in the fall of 2023, further studies of blood donor vaccination behaviour are warranted to understand to what extent donors may have reduced vaccination rates.

P170 | Abstract withdrawn

P171 | Whole-blood and plasma donor beliefs about the health impacts of donation and impact on donation frequency—a survey study

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Background: As blood collection agencies (BCA) rely on established, repeat donors to ensure a stable supply of blood the health and well-being of donors is of critical concern. While health is often raised as a reason for lapsing or reducing donation frequency, we do not know the specific health beliefs donors hold, and whether these affect donation-related decision-making.

Aims: To investigate the donation-related health beliefs of donors and how these relate to subsequent donation behaviour.

Methods: Novice (1-2 donations in past 2 years) and experienced (3+ donations in the last 2 years) whole-blood (WB) and plasma donors were asked in an online survey if they had considered the health impacts of donating (yes/no), and to select the health impacts they believed resulted from donating (1 = Not an impact, 5 = Definitely an impact). Negative binomial regression models determined the impact of health beliefs on donation frequency within 6 months (plasma) and 12 months (WB) by study group, controlling for age and gender.

Results: Of the 1127 (RR 11%) respondents, 54% were WB donors (55% female; mean age 51.6 years [± 13.7], mean of 8.8 prior donations [± 17.7]). Plasma donors were mostly female (62%; mean age of 47.4 years [± 14.5], mean of 4.4 prior donations [± 5.3]). Around half of respondents (53%) had thought about physical health impacts of donation while around one-third (33%) had thought about mental health impacts. For WB donors, there was generally low agreement (below scale midpoint) with physical health impacts of donating. The most strongly endorsed negative physical health beliefs were lowering of iron levels (1.99 ± 1.29), reduced physical activity (2.03 ± 1.20) and feeling tired, weak, low energy (1.97 ± 1.17). For plasma donors reduced physical activity (1.83 ± 1.08), bruising (1.83 ± 1.00) and feeling tired, weak, low energy (1.80 ± 1.04) were most strongly endorsed. Positive mental health impacts were more strongly endorsed than physical health impacts. For WB and plasma donors the most strongly endorsed mental health impact was 'feeling good from knowledge that you've helped others' (4.74 ± 0.72) followed by 'feeling like you're giving back to community' (4.59 ± 0.83). 'Feeling like donating is good for your mental health' was more strongly endorsed by plasma than WB donors (4.04 ± 1.29 vs 3.84 ± 1.42 , $p = 0.0098$). Between 82% (novice plasma) and 96% (established plasma) attempted to donate again at 6 or 12 months. Donors who had lower endorsement of 'light-headedness, dizziness or nausea' (novice WB: IRR = 0.890, $p = 0.030$; novice plasma: IRR = 0.868, $p = 0.007$), higher endorsement of donating being good for mental health (established WB: IRR = 1.093, $p = 0.003$), and lower endorsement of donation lowering iron levels (established WB: IRR = 0.924, $p = 0.040$) donated more frequently in the follow-up period. No significant predictors were found for established plasma donors.

Summary / Conclusions: The physical impacts of donating are salient considerations for WB and plasma donors. While positive impacts of donating were strongly endorsed, most were not associated with donation frequency. In contrast, beliefs about negative physical impacts, such as feeling dizzy and iron loss, were less strongly endorsed but negatively affected donation frequency. Findings indicate opportunities to educate donors about the physical effects of donation, and how to manage them, to encourage donor return.

P172 | Abstract withdrawn

P173 | Abstract withdrawn

P174 | A survey of association between rs11549465, rs855791 and rs651007 and biochemical indicators of iron status in regular blood donors of Tehran Transfusion Center

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Background: Iron deficiency anemia is a common problem for frequent blood donors, and it can harm their health and quality of life. Genetic variations, particularly genes related to iron absorption and transportation in the blood, may play a role in contributing to iron deficiency in these donors.

Aims: We evaluated the association of polymorphisms rs11549465, rs855791, and rs651007 with iron levels in male regular blood donors.

Methods: A total of 130 male regular donors participated in this study. After obtaining demographic information from the donors, serum iron, ferritin, and TIBC tests were performed on their blood samples. The genotypes of the targeted polymorphisms were determined using High-Resolution Melt Curve Analysis and Allele-Specific PCR methods. All collected data were subjected to statistical analysis using the R software.

Results: Among 130 regular blood donors, 13 (10%) had low ferritin levels, 9 (6.9%) had low iron levels, and 16 (12.3%) had higher than normal iron levels. There was a significant association between the rs11549465 polymorphism and ferritin levels of donors ($p = 0.02$). However, no significant association was observed between the rs855791 and rs651007 polymorphisms and iron assessment indices.

Summary / Conclusions: The results obtained from the measurement of serum iron, ferritin, TIBC, and TSAT were 6.9%, 10%, 12.3%, and 13.8%, respectively, indicating that some of the blood donors suffered from iron deficiency without anemia and iron-deficiency anemia. This study also investigated the impact of genetic polymorphisms on iron status, revealing that some of these polymorphisms affect iron levels and related parameters. Given the importance of blood donation in transfusions and patient care, it is essential to identify individuals at risk of low iron levels. This identification can help blood donation centers accurately identify at-risk groups and take appropriate measures to protect these individuals after donation.

P175 | Analysis of erythrocyte parameters and iron nutrition levels of plateletpheresis donors in Hangzhou of China

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Background: With the development of society, the improvement of medical technology level and the increasing health needs of the people, the number of various organ transplantation operations has increased year by year, and the clinical demand for platelets has also increased year by year. The number of blood donors and the frequency of blood donation have increased significantly. Whether the increasing frequency of platelet donation will affect the health of platelet donors has become the focus of blood collection and supply organizations at home and abroad.

Aims: The purpose of this study is to find out the population who need to pay attention to iron nutrition level in blood donation service in this area, so as to effectively guide the practical work and improve the iron nutrition level and overall health literacy of platelet collection donors.

Methods: A total of 993 plateletpheresis donors were included in this study, including 798 males and 195 females. The results of red blood cell parameters and iron nutrition parameters of blood donors were

compared and analyzed in different groups according to the gender, age and number of blood donations.

Results: Ten point eight percent of male donors and twenty-seven point seven percent of female donors had low serum ferritin (SF) levels in this study. There were differences in blood donation frequency parameters, red blood cell metabolism, iron metabolism parameters and their changing trends between male and female donors. The mean levels of serum iron (SI), SF, total iron binding capacity (TIBC), hemoglobin (Hb), hematocrit (HCT) and other parameters of male blood donors in different age groups showed a decreasing trend with the increase of age groups, but there was no significant statistical difference between the results of female blood donors in different age groups. The level of SI, SF, transferrin (TRF), Hb, HCT, mean corpuscular volume (MCV) of male donors tended to decrease with the increase of blood donation times in the past year, while TRF and TIBC tended to increase. Different from male blood donors, with the increase of the number of blood donations in the past year, the TRF of female blood donors had an increasing trend, and the TIBC and TRF had a decreasing trend. However, Hb, HCT, MCV, MCH and SF levels of female donors showed no significant downward trend as the number of donations in the past year increased.

Summary / Conclusions: Blood collection institutions need to focus on iron nutrition levels in older and frequent male donors, and young fertile female donors. Regular monitoring of iron nutrition levels and health education such as eating habits are recommended for these populations.

P176 | Effect of Covid-19 pandemic on anticipated young blood donors—a study from lower middle income countries

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Background: Voluntary blood donation in young blood donors who will be turning 18 years of age which is legal age for blood donation in India are explored in this study, with a focus on the safety and altruistic motivation associated with voluntary donors. The abstract addresses the severe challenges faced during the covid 19 pandemic creating a hinderance in voluntary blood donation. The importance of community-based interventions to improve knowledge and attitudes toward blood donation in young blood donors is emphasized.

Aims: The primary aim of the study was to assess the knowledge and attitudes of young blood donors, specifically targeting 12th-grade students in Bhopal, Madhya Pradesh. The study seeks to evaluate the impact of motivational lectures, delivered both online and offline, on the awareness and perception of blood donation among students. After the motivation lectures it is anticipated to have young blood donors to increase the voluntary donor pool.

Methods: The study involves community-based interventions, primarily motivational lectures conducted for 12th-grade students. Due to the COVID-19 pandemic, a combination of online and offline modes is

employed. Consent is obtained from both students and their parents or guardians. The abstract outlines the duration of the study (15–18 months).

Results: Motivational lectures thus help to bring a positive attitude in the student towards blood donation. However, only giving the didactic motivational lecture doesn't have a significant impact on actual blood donation. So, parents influence is equally needed to add more impact on such intervention programmes. Hence, Parents counselling should be done simultaneously along with such implemented programmes. The work of motivation has been done by us to the students but the lack of motivation to parents for sending their ward wasn't done which lead to unpromisable results. Covid Pandemic also became a major hinderance as there was fear amongst the parents that the children can also get susceptible to Covid 19.

Summary / Conclusions: (1) Covid 19 pandemic was one of major limitation which hampered the sample population and also affecting the post motivational donation due to fear of acquiring the covid infection. (2) Limited study period of 15 months was insufficient to access the effect of intervention of motivational lectures on blood donation. (3) Lack of interaction with the parents of these students during the study period was another limiting factor for blood donation which if done would have added more impact on our motivational lectures.

P177 | Vel negative individuals, homozygous for the SMIM1 loss-of-function variant, have reduced energy expenditure and weight gain

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Background: Function and associations to disease have been extensively studied for ABO and RhD blood groups because of routine testing and their importance in transfusion medicine. Variants in the ABO blood group have previously been linked to increased risks of

bleeding disorders (blood group O) and thrombotic events (blood groups A, B, and AB). However, knowledge of the function and disease associations for most other blood groups is limited. In this study, we present that Vel negative individuals homozygous for the 17-bp nonsense-deletion in the Small Integral Membrane Protein 1 (*SMIM1*) present in 1 in 5000 and 1 in 2000 of the UK Biobank (UKB) and Danish Blood Donor Study participants, respectively, associate with dyslipidemia and lower thyroid hormone levels. These alterations were accompanied by reduced resting energy expenditure and a gain in weight.

Aims: To characterize the metabolic and cardiovascular consequences of lacking *SMIM1*, i.e. absence of the Vel blood group protein.

Methods: We performed linear and logistic regressions on the UKB (90 *SMIM1*^{-/-}, 11,849 *SMIM1*^{+/-}, and 396,559 *SMIM1*^{+/+}) to investigate genotype-phenotype associations. Findings were replicated in English blood donors (25 *SMIM1*^{-/-} and 180 controls), Danish blood donors (43 *SMIM1*^{-/-} and 645 controls), and Danish patients (30 *SMIM1*^{-/-} and 450 controls). Single-cell RNA-Sequencing (scRNA-seq) was used to investigate possible mechanisms. Results are presented as regression coefficients (β) and odds ratios (OR) with Benjamini-Hochberg method for false discovery rates correction of p-values (q-values), and significance level at 0.05.

Results: In *SMIM1*^{-/-} UKB participants, we found an association with increased weight ($\beta = 0.22$, $q = 4.61 \times 10^{-2}$), more prominent in women than men, which equates to 4.6 and 2.4 kg on average, respectively. This observation was replicated in the blood donor cohorts. Analysis of UKB-plasma biochemistry showed increased triglycerides, urate and liver-enzymes levels. A clinical metabolic assessment showed reduced energy expenditure. Association analysis for cardiovascular events in UKB showed an increased risk for cerebral bleeds (OR = 5.53, $q = 6.88 \times 10^{-4}$) and thrombotic stroke (OR = 3.46, $q = 2.32 \times 10^{-2}$). scRNA-seq showed expression of *SMIM1*-transcripts in the pituitary glands, suggesting a central hypothyroidism as a possible cause of the traits observed.

Summary / Conclusions: Vel-negative individuals, homozygous for loss-of-function variants at the *SMIM1* locus, exhibit altered lipid metabolism, due, at least in part, to reduced energy expenditure, resulting in a gain in weight. The present study also suggests that Vel-negative individuals suffer from mild hypothyroidism. Further studies are required to understand the role of this small transmembrane-protein in the pituitary-thyroid axis and whether thyroid hormone supplementation can reverse the weight gain.

Blood donation—blood collection including apheresis

P178 | Impact of donor stress level on the coagulation factor VIII of apheresis machine donor

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Background: Response to stress consists of three parts: psychological, hormonal, dan physiologic. Psychological response covers an increase level of anxiety, fear and tension. Hormonal response covers an increase excretion of adrenaline and cortisol. Physiological response covers an increase of heart rate, blood pressure and changes in heart rate variability. Not only that, blood hemostasis may change and induce the increase of coagulation.

Aims: To determine the effect of the donor's stress response on blood clotting factor (Factor VIII) in the Fresh Frozen Plasma collected by apheresis.

Methods: Twenty-seven random apheresis donors were tested by Dass Test. The plasma was collected with Amicore apheresis, and was processed into Fresh Frozen Plasma. Then, all the bags were tested for Factor VIII. The FVIII is still considered good if it is ore that 0.545. The result of the Dass Test and Factor VIII were analyzed using the linear regression analysis method and statistically assessed using ANOVA and t- test.

Results: According to the Dass test, 45.8% of the donors are under stress, while the other 43.2% are not. The result of the FVIII was still high in 62.1% of the donor, and not good in 37.9%. The Dass test result have an impact in the FVIII level. ANOVA test gave a significant result ($p < 0.05$) and t-test also gives a significant result ($p < 0.05$).

Summary / Conclusions: Stress response of the donors that was checked using Dass Test before donating the plasma with Amicore apheresis machine have an impact in the result of Factor VIII level in Fresh Frozen Plasma. There is a need of more samples before Dass Test could be implemented for all the apheresis donor.

P179 | Factors associated with systematic differences in capillary and venous hemoglobin measurement in a large diverse blood donor cohort

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Background: Requiring a minimum level of hemoglobin (Hb) in presenting blood donors helps ensure the potency of products and guard against drawing blood from an anemic donor. Hb measurements at blood drives using portable devices with a capillary (C) sample are known to be less accurate than those measured on hematology analyzers using a venous (V) sample. At least two reports (Cable, Transfusion, 2012; Tong, Vox Sang, 2010) suggest that CHb is not an

unbiased estimator of VHB, with systematic bias occurring on the basis of donor sex and other factors. The prevalence, magnitude, and nature of any such bias have potential implications for donor health, transfusion outcomes, and blood center operations.

Aims: To estimate and characterize the difference between VHB and CHb in a large cohort of blood donors.

Methods: This study represents a re-analysis of the enrollment visit for the REDS-III RBC Omics donor population, a large US cohort enriched for racial/ethnic minority donors and high-frequency donors. Successful enrollment included a productive whole blood and venous sample collection. CHb and demographics were retrieved from operational data, VHB was performed locally on hematology analyzers, and plasma ferritin was batch tested at a central laboratory. The mean difference between VHB and CHb measurements was calculated, overall and by race, sex, age, iron status, blood center, and CHb level grouped into 0.5 g/dL bins. Linear regression was conducted to assess the association between each covariate and VHB-CHb controlling for other covariates.

Results: Of 12,871 RBC Omics subjects with VHB, CHb, and ferritin values (96% of total enrolled), the average VHB-CHb difference was -0.38, indicating overall CHb was on average nearly 0.4 g/dL higher than VHB. Both unadjusted and modeled differences of VHB-CHb varied by sex, race, iron status, blood center, and CHb bins of 0.5 g/dL increments, but not by age. In linear regression, female sex, iron status and CHb level were associated with greater divergence of VHB and CHb values. Controlling for other covariates, predicted values for VHB-CHb in females were -0.52 g/dL (95% CI -0.56, -0.50) compared to males. In subjects with ferritin <12ng/mL predicted VHB-CHb was -0.44 g/dL (95% CI -0.47, -0.41) compared to those with ferritin ≥26ng/mL. Across the range of CHb used to establish donor eligibility, a monotonic trend was observed where higher CHb levels increasingly over-estimated VHB values (i.e., the difference became increasingly negative). For subjects with CHb in the range 12.5-12.9 g/dL, VHB was predicted to be 0.44 g/dL (95% CI 0.37, 0.51) g/dL higher than CHb. For subjects with CHb 13.0-13.4 g/dL, VHB was predicted to be lower than CHb by -0.17 g/dL (95% CI -0.22, -0.13), with that difference increasing to -1.31 g/dL (95% CI -1.36, -1.26) for those with CHb >16.0 g/dL.

Summary / Conclusions: Systematic differences were observed between VHB and the CHb values used to qualify donors for blood donation, with modeled differences varying by demographic, biological, and operational factors. This analysis confirms earlier reports that at a given CHb used to qualify a blood donor, female donors have substantially lower VHB than male donors. Female donors who are iron-deficient may be particularly susceptible to mis-estimation of true Hb levels, potentially increasing the risk of both drawing a unit from anemic donors and inducing attendant symptoms or adverse outcomes.

P180 | Impact of bidirectional data interface on operational errors in platelet apheresis collections - a comparative analysis before and after implementation

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Background: Through the bidirectional data interface, real-time information exchange between the apheresis equipment and Blood Bank Information Systems (BBIS) allows a significant reduction in human errors and improves the quality of the platelet collection process and products. By providing a seamless flow of information, the bidirectional data interface ensures that all necessary information is communicated accurately, contributing to eliminating manual data entry errors.

Aims: This study aims to describe the frequency of operational errors before and after the implementation of bidirectional data interfacing. Additionally, the study intends to evaluate the impact of bidirectional data interface on these errors in platelet apheresis collections, including, but not limited to donors.

Methods: We conducted a retrospective comparative analysis of operational errors in the collection of platelet apheresis on the parameters: gender, weight, height, hemoglobin, platelet count and blood type. We compared the frequency of errors recorded in 6643 collections before and 5714 collections after the implementation of the bidirectional data interface in our service.

Results: Before the implementation of the bidirectional interface, 55% of procedures had at least one discrepancy in the donor information entered into the apheresis device. The parameters that most frequently showed differences were Hemoglobin/Hematocrit (45% of procedures and $p < 0.05$) and Height (17% of procedures and $p = 0.07$). After implementation, the frequency of errors reduced significantly to just 2% ($p < 0.05$), reporting discrepancies in donor information and the parameters that showed the most discrepancies were Height (8% of procedures and $p < 0.05$) and Weight (0.5% of procedures and $p = 0.06$). The study also observed a decrease in interactions between staff and the apheresis equipment; i.e. before implementation, it required an average of 62 touches on the equipment screen to complete a procedure, while after implementation, only 46 were needed (a decrease of 25.8%). As a result of the reduction in errors, we also observed, a reduction in products flagged as possibly contaminated with leukocytes. Before the implementation of bidirectional interfacing, 5% of platelet products were flagged as possibly contaminated with leukocytes and after implementation, this number decreased to 3% ($p = 0.05$).

Summary / Conclusions: Operational errors can significantly affect the safety and efficacy of platelet apheresis procedures. As apheresis devices rely on the information entered to calculate the parameters that are essential to ensuring the safety and efficacy of the procedure, errors during this phase can lead to serious consequences. The cases in which there was a discrepancy between the donor data entered into the

apheresis equipment versus the BBIS were caused by technical problems that resulted in the equipment being disconnected and consequently making it impossible to transmit the data or were related to procedures in which the user manually altered the data due to some reason. This is the first study that has been able to demonstrate the impact of using data management such as bi-directional interfacing in pro-apheresis platelet procedures and how this reduces operational errors.

P181 | Increased plasma volume collected on a plateletpheresis separator

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Background: Blood Service (SFS) of the Belgian Red Cross like other European blood establishments must increase plasma collection notably to meet growing demand on immunoglobulins. One source of plasma is the secondarily collection of plasma in a specific bag during a platelet donation using apheresis separator like Trima Accel[®] (Terumo BCT). Recently volume of plasma collection bag on Trima set has been increased by 17%. This change represents an opportunity to increase plasma collected during platelets collection without exceeding legal requirements: 650 ml plasma per collection, anticoagulant (AC) excluded.

Aims: The aim of the study was to evaluate plasma volume gain and possible donor impact with adapted Trima Accel settings used in two collecting platelets centres of the SFS.

Methods: Trima set (ref: 82410) is used for concomitant collection of platelets (simple or double doses - suspended in 38% of plasma / 62% of PAS) and plasma in an additional bag. We decided to modify the settings in order to increase the volume of total plasma collected. In the Trima of the SFS, we have 3 parameters scheduled to protect the donor. When one of these limits is reached, the quantity of plasma collected stops. This is to protect lightweight donors or with a small total blood volume (TBV). 2 of these parameters were modified. The volume of plasma collected cannot exceed: A maximum of 15 % of TBV. A volume limit by donor according to his weight: 10 mL/kg became 10.5 mL/kg. A maximum volume collected by donor according to his weight: ≤60kg: maximum 500 mL became 600 mL. >60 kg: maximum 600 mL of plasma became 640 mL. We tested the new settings in 2 collecting centres between 06/11/2023 and 07/12/2023. This was tested on a panel of 196 donations (23% of double doses). The tolerance of the donors was followed up during this period analysing incident / complication sheets. The only recommendation made to them was to hydrate a little more than usual during donation. We compared these results to a reference period for these 2 sites, June 2023 (N = 249, 24% of double doses).

Results: Plasma volume collected including plasma fraction on platelet bag was compliant with legal requirements for all donations (maximum at 648 ml without AC). Average plasma volume collected including AC was 457 ± 62 ml during reference period and 544 ± 56 ml during test period (with AC). Resulting in an average increase of 87 ml per collection procedure. Duration per procedure was similar between the

2 periods. No more incidents or complications were identified in donors during this test period on the basis of incident / complication sheets.

Summary / Conclusions: The SFS of the Belgian Red Cross now collects more plasma in the plasma bag during plateletpheresis on Trima Accel[®] with the same level of donor safety and without impact on adverse events.

P182 | Increasing collection capacity by adopting a new plateletpheresis device

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Background: One of the efforts to ensure transfusion self-sufficiency is to obtain high-quality blood components while ensuring maximum donor safety and comfort what may be one of the conditions for their regular donation behavior. Blood Establishment (BE) in Rzeszów constantly improves the quality of blood components and donor safety by equipping the center with modern medical devices. In our BE in year 2023 we performed 1986 platelet apheresis by use of: AmiCORE and Amicus Apheresis Systemas from Fresenius Kabi—two distinct separation devices. We have been working with the Amicus for many years, and the AmiCORE system was implemented in year 2022. Both devices include process leukoreduction and saline infusion for donor comfort. The AmiCORE system is already used in many centers in Europe, and offers simplified operation of platelet collection, as well as collection with platelet additive solution. When evaluating new apheresis technologies for mean platelet count and residual leukocytes, it is also important to assess donor comfort to prevent donor discouragement.

Aims: To assess donor safety, comfort and performance of both AmiCORE and Amicus separators.

Methods: All staff collecting blood components were trained on the AmiCORE System by Fresenius Kabi instructors. The device settings were adjusted to obtain a platelet level of at least 4.0×10^{11} /unit. We assessed 156 collections performed on AmiCORE and 120 performed on Amicus for in vitro platelet quality at first day. After 5 days of storage the pH was determined for 61 donations collected on AmiCORE and 38 collected on Amicus. Results are given as mean values. Platelet donations were tested on a new generation analyzer adapted to quality control tests in blood components—Sysmex XN-1000. pH assessment

P182 Table 1: Amicus Separator and AmiCORE Apheresis System Results

Device (No. of samples tested)	Volume [mL]	PLT $\times 10^{11}$ / unit	WBC $\times 10^6$ / unit	pH (samples tested)
Amicus (120)	296	4.2	0.03	7.29 (38)
median	296	4		7
AmiCORE (156)	303	4.1	0.03	7.29 (61)
median	304	4		7

was performed by use of pH-METER CP-511. For data collection from all AmiCORE devices we used Data Collection System DXT 3.7 and for Amicus devices—iTrace system.

Results: Mean platelet yield was 4.1×10^{11} for AmiCORE vs. 4.2×10^{11} for Amicus donations. Average procedure time was 49.8 min for AmiCORE and 50.3 min for Amicus. All platelet units met the EU standard of $< 1.0 \times 10^6$ leukocytes and pH level after 5 days storage. Swirling was observed in all platelet units throughout the storage period. No aggregates were observed in the final products. No adverse reactions were reported by donors, even not citrate triggered symptoms while using both devices.

Summary / Conclusions: AmiCORE in comparison with Amicus met all criteria for platelets, leukocytes, pH and unit volume per national requirements. AmiCORE is an easy-to-use apheresis platform to collect platelets in plasma and PAS with minimal variation from pre-defined device setup. The efficiency of both devices is similar, as indicated by the comparable procedure time, volume and number of platelets in the products. The DXT system used in AmiCORE enables the collection of data from all new generation Fresenius Kabi devices which facilitates work. While using both the AmiCORE and Amicus Systems no adverse reactions occurred in donors, which is most likely related to the features such as: the use of saline for kit priming, infusion and reinfusion, numerical flow rate control, and the use of an active cuff system that optimizes flow during collection.

P183 | How to operate a sustainable plateletpheresis programme in LMIC—experience from Namibia

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Background: The Blood Transfusion Service of Namibia (NAMBTS) has a sole mandate to ensure availability of blood components, including adequate platelets for transfusion in Namibia. The blood service launched the plateletpheresis programme in 2007 using one device and a second device was purchased two years later. However, due to a 9% average year on year increase in platelet demand and increasing cost of plateletpheresis, NAMBTS needed to explore ways to meet demand and optimize plateletpheresis collection to achieve high output at the lowest cost possible. To achieve this objective, NAMBTS implemented two strategies

Aims: **Strategy 1:** Identify a platelet collection device that can collect high yield outputs at the current or lower cost. **Strategy 2:** Increase the productivity of Trima Acel by increasing split rate of high yield platelet donations. The minimum yield per product is set at $2.4 \times 10^{11}/L$ for an adult dose and $6.0 \times 10^{10}/L$ for a paediatric dose.

Methods: **Strategy 1:** Trima Acel device was selected and evaluated against the existing device (device A). Existing plateletpheresis donors with an Hb ≥ 12.5 g/dL and pre-donation platelet count $\geq 230 \times 10^3/\mu L$ and who met all other platelet donation criteria were recruited for the evaluation. A total of 47 donors participated in the evaluation, 20 donors

donated on brand A and 27 donated on the Trima Accel brand. The average pre-donation platelet count for all donors was $317 \times 10^3/\mu L$.

Strategy 2: Donors were carefully selected and priority was given to donors with a history of producing higher yield donations and a pre-donation platelet count of $230 \times 10^3/\mu L$ was preferred. Donations with yields above $6.0 \times 10^{11}/L$ are split into two adult dose products while those above $4.0 \times 10^{11}/L$ but below $6.0 \times 10^{11}/L$ are split into one adult dose and paediatric dose products.

Results: **Strategy 1:** The average pre-donation platelet count for all donors was $317 \times 10^3/\mu L$. Donors who donated on brand A device had a higher average pre-donation platelet count ($353 \times 10^3/\mu L$) than the average pre-donation count ($306 \times 10^3/\mu L$) of those who donated on the Trima Acel device (p -value = 0.0011). However, the average platelet output yields from the Trima Acel was significantly higher than those from brand A (p -value = 0.0040). The average donation time for donors who donated on the Trima Accel was shorter than that of brand A (p -value = 0.0084).

Strategy 2: Donations with yields above $6.0 \times 10^{11}/L$ are split into two adult dose products while those above $4.0 \times 10^{11}/L$ but below $6.0 \times 10^{11}/L$ are split into one adult dose and paediatric dose products. The split rate of apheresis platelets between from the previous three financial years have shown a continuous growth of 36% for 2021, 50% for 2022 and 93% for 2023 respectively.

Summary / Conclusions: We concluded that the Trima device achieved higher productivity than brand A. The Trima Accel became our dedicated brand for platelet collection while the other brand was diverted to erythrocytapheresis. Both strategies were implemented successfully, resulting in the tremendous reduction of the collection cost of apheresis platelets by saving US \$205, 000.00 (during year 2023) and improved availability of platelets for transfusion.

P184 | Evaluation of a new apheresis device for single and double dose platelet collection and comparison with existing device—study at a major stand-alone blood center

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Background: Jankalyan Blood Centre, Pune is a major stand-alone blood centre in West India catering to a 150 kilometres radius and at times also to any place across the nation. The center supports patients across 100+ hospitals and has been performing plateletpheresis for last 11 years with over 1200 procedures per year. Amicus (Fresenius Kabi) is the already existing cell separator used for collecting platelets in Platelet Additive Solution (PAS) and concurrent plasma. To cope up with increasing workload, the center decided to validate AmiCORE, the new apheresis system/cell separator by Fresenius Kabi, Germany. AmiCORE facilitates a very user-friendly set installation, machine handling, trouble shooting, and retrieval of procedure data. It also ensures leucodepletion and saline infusion. Due to intermittent flow control, it

P184 – Table 1

	SD	DD
N	82	40
Donor Pre-count	$261 \times 10^3/\mu\text{L}$	$332 \times 10^3/\mu\text{L}$
WB processed	2322 mL	3505 mL
ACD used	267 mL	361 mL
Procedure Time	53.2 min	74.6 min
Donor post count	$208 \times 10^3/\mu\text{L}$	$207 \times 10^3/\mu\text{L}$
Target Yield	3.5×10^{11}	6.0×10^{11}
Product volume	280 mL	560 mL
Actual Yield	3.43×10^{11}	6.04×10^{11}
A/T ratio	0.98	1.007
Collection efficiency	70.02%	73.10%

P184 – Table 2

	AmiCORE	Amicus
No of procedures	45	45
Donor Pre-count	$258 \times 10^3/\mu\text{L}$	$272 \times 10^3/\mu\text{L}$
WB processed	2340 mL	2319 mL
ACD used	258 mL	223 mL
Procedure Time	52 min	50.9 min
Donor post count	$212 \times 10^3/\mu\text{L}$	$229 \times 10^3/\mu\text{L}$
Target Yield	3.5×10^{11}	3.5×10^{11}
Product volume	280 mL	280 mL
Actual yield	3.42×10^{11}	3.58×10^{11}
A/T ratio	0.98	1.02
Collection efficiency	70.00%	71.97%

gives tremendous donor comfort and minimises adverse donor reactions.

Aims: To evaluate the procedure performance and platelet quality of the AmiCORE system for single and double dose platelet collections in PAS. Additionally, the device performance was compared to Amicus.

Methods: 122 platelet procedures were performed on the AmiCORE system between January to December 2023. Of these, 82 were single dose (SD) and 40 were double dose (DD) collections. Healthy volunteer donors were recruited as per the national apheresis donor eligibility criteria. All platelet products were stored in PAS – InterSol (Fresenius Kabi) in the PAS:Plasma ratio of 70:30. The target platelet yield was 3.5×10^{11} for SD and 6.0×10^{11} for DD. The Citrate Infusion Rate was set at 1.25 mg/kg/min with a fixed WB:ACD ratio of 10:1. Maximum cycle volume was 250 mL. Concurrent Plasma of 200 mL was collected in all SD procedures. Donor pre- and post-donation platelet count and procedural characteristics were recorded. Platelet product quality parameters were measured. Donors were observed for any adverse reactions. 45 SD AmiCORE procedures were compared to 45 SD Amicus procedures done during the same

time period of October – December 2023. The donor and procedural characteristics were similar.

Results:

The average residual WBC count was $<0.22 \times 10^6$. Mean pH was 6.8 with high quality swirling and no aggregates. Bacterial testing showed no growth for both SD and DD procedures. All the procedures were well-tolerated by the donors and no adverse reactions were observed during and after any of the procedures.

Summary / Conclusions: Platelets prepared on AmiCORE met our internal quality standards, The device performance was satisfactory and comparable to our existing device. AmiCORE is now routinely being used at our blood center for plateletpheresis.

P185 | Evaluation of improved device software for the collection of apheresis platelets: Reinfusion time and loss of red blood cells

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Background: A shorter procedure time for plateletpheresis is in the interest of both the donor and the blood center. To reduce the overall procedure time, Fresenius Kabi developed new software (version 2.1) for platelet collection with the AmiCORE Apheresis System to reduce the time required for reinfusion at the end of a collection. In addition, the changes to reduce reinfusion time were devised to minimize any impact on the residual red blood cell (rRBC) volume lost in the kit following the disconnection of the donor from the device. To ensure that the new software performed as intended, a pilot study was conducted to determine the reinfusion time and rRBC volume at the completion of platelet collection procedures.

Aims: To determine the reinfusion time and the residual RBC volume loss at the end of a collection procedure with the AmiCORE device using new software version 2.1.

Methods: The device was programmed to run as a plasma procedure (i.e., plasma as platelet storage fluid). Residual red blood cell volume (rRBC) (N = 22): The rRBC volume was measured in the disposable kits after 22 procedures with pooled human donor whole blood (4-5 units/pool). The hematocrit (HCT) was adjusted to 35, 45 and 55%, with anticoagulant ratios at 7:1, 9:1 and 12:1 (WB:AC). After the blood supply had been disconnected, residual blood in the disposable kit was drained by pushing it out of the kit with a tubing stripper, and by forcing it out of the cassette pathways with an air-filled syringe. The recovered blood was collected, the HCT measured, and the residual red blood cell volume calculated. The results were analyzed with 95% confidence level for 90% of the population. Reinfusion time (N = 10): Bagged bovine blood was used for a total of 10 pilot procedures, most of which were single dose donations. Donor characteristics (height, weight, HCT, platelet pre-count, target yield, target storage volume) similar to a previous customer comparator study were entered into the device to mimic a diverse donor pool, with the

HCT of the bovine blood adjusted by removing or adding plasma. A software 2.1 verification build was used, with the subsequent final build containing changes unrelated to reinfusion time.

Results: The mean residual RBC volume in the kit at the conclusion of a procedure was 16.1 mL, meeting a limit of 30 mL with a 95/90 tolerance interval (upper bound of tolerance limit: 21.5 mL). The average reinfusion time of the 10 procedures was 3:20 (min:sec) with a standard deviation of 0:23. The new software shortened the collection procedure by reducing the reinfusion time by more than 3 min, compared to previous data achieved with the AmiCORE device in the customer comparator study. It is believed that the use of bovine blood for this part of this study had no influence on the reinfusion time.

Summary / Conclusions: The new software version 2.1 met the criteria for residual red blood cell volume. Red cell loss was maintained at an average of 16.1 mL per procedure and below the previous study average of ≈ 19 mL. Returning blood from the disposable kits to the donor at the end of collection procedures will benefit the donor by reducing iron loss (Infanti, Transfusion and Apheresis Science, 2023) and by reducing the risk of leukopenia (Kaufman, Blood, 2019). The new software also successfully reduced the time required for the reinfusion at the end of platelet collection procedures, reducing the overall time required for platelet collection, which is a benefit to the donor and the blood center.

P186 | Optimizing blood donation workflow—a study on integrating donor registration and hemoglobin testing for improved pre-donation efficiency

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Background: Efficiency optimization is pivotal in the Blood Bank to enhance participation and maximize resources. A continuous evaluation of the pre-donation workflow at all collection sites aims to improve the blood donor experience. This study investigates the impact of integrating hemoglobin testing with blood donor registration, compared with the traditional approach of separate stations where donors progress sequentially through stations, including donor registration, hemoglobin testing with weight-taking, and a medical interview. We propose combining blood donor registration and hemoglobin testing into a single station to enhance operational efficiency and the donor experience.

Aims: The proposed process, combining registration and Hb testing with weight-taking, was piloted at a designated blood donation center. A comparative analysis assessed processing time and operational efficiency.

Methods: The processing time for each donation case during the pre-donation phase was continuously monitored from July to December 2023 after the station combination, compared to the ones before the changes. The number of staff assigned to the process was considered to study manpower efficiency. An online survey invited 200 repeat blood donors to share their experience

with the streamlined process upon completion of donation in parallel with the study.

Results: The average processing time for pre-donation per donor decreased by 9 min from July to December 2023, representing a 24% time savings. The workload initially handled by five staff members was successfully managed by four, reducing manpower by 20%. Survey results from 91% of the 200 participants expressed satisfaction with the combined process, and an overwhelming 97% favoured making the new setup permanent.

Summary / Conclusions: This study underscores the importance of streamlining procedures in healthcare settings to enhance accessibility and effectiveness in blood donation initiatives. Results indicate a reduction in processing time and manpower costs, enhancing operational efficiency with the combined approach. Donors reported greater convenience and a positive experience with the integrated process, potentially increasing the rate of recurring donations. Blood bank plans to implement the integrated process across all sites as part of continuous improvement, bringing benefits to the organization and all blood donors.

P187 | The first experiences in the Republic of Srpska about donor thrombocytophoresis

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Background: One healthy person donates blood through an automatic blood cell separator with the procedure of donor thrombocytophoresis. The separator is programmed to separate and collect the desired number of platelets while simultaneously returning all other blood components.

Aims: The objective is to present data on collected apheresis platelets at the Institute for Transfusion Medicine of the Republika Srpska in the period from February 2021 to January 2024.

Methods: Using sets (LRS, Platelet/Plasma) for single use, the separation of apheresis platelets was performed on the TRIMA ACCEL 7 automatic blood cell separator, enabling the collection of leukoreduced platelets.

Results: During the analysed period, a total of 247 donor thrombocytophoresis procedures were conducted involving 62 voluntary donors. The resulting platelet concentrates obtained from these procedures had a yield ranging from 3 to 4.5×10^{11} . Within the legally prescribed period of every 15 days, the apheresis procedure was performed once in 28 donors, which accounts for 46.67% of the total donors. Additionally, the procedure was performed more than once in 34 donors, representing 54.33% of the total donors. Out of the total number of performed apheresis procedures, 231 (93.5%) resulted in the separation of two units of platelets, while 16 (6.5%) procedures resulted in the separation of only one unit. The average age of the donors was 35 years. Before the procedure, the average haematocrit value was

43.04%, which decreased slightly to 42.58% after the procedure. The average hemoglobin value was 150.85 g/L. The platelet count before the procedure was $229.15 \times 10^9/L$, which decreased to $190.84 \times 10^9/L$ after the procedure. The processed volume during the procedure was 2525.4 mL, and 273.12 mL of anticoagulant (AC) was used. The procedure had an average duration of 43 min. On average, 383.3 mL of the final product was separated, which had leukocytes below 1×10^6 . We successfully pathogenically inactivated 54 platelet concentrates, which accounted for 21.86% of the total collected. These platelet concentrates were collected in nutrient solution (SSP+) and had a volume of over 300 mL. The pathogenic inactivation process was carried out using the INT100 INTERCEPT Illuminator.

Summary / Conclusions: Platelet concentrates obtained through the donor thrombocytapheresis procedure meet all quality requirements as per the latest recommendations from the Council of Europe. Considering the great clinical importance of apheresis platelets, our plan is to increase the number of apheresis procedures and expand our existing register of voluntary platelet donors.

P188 | Automated methods for collection of blood components - Polish experience during a fifteen-year period (2008–2022)

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Background: One of the fundamentals of proper functioning of the healthcare system is adequate supply of blood and blood components which has lately become a challenge for the blood transfusion service in many countries, Poland included. Supply of certain blood components may be supported by the use of automated methods of blood collection (apheresis).

Aims: The study aim was to assess the extent to which automated methods were used for collection of blood components in Poland in the period 2008–2022.

Methods: Retrospective analysis of data collected by 21 Polish Regional Blood Transfusion Centers.

Results: In the period 2008–2022, the method of apheresis was used mainly to obtain plasma and platelet concentrate (PC), rarely red blood cell concentrate (RBC) and granulocyte concentrate (GC). Occasionally two blood components were simultaneously collected by apheresis - most often PC and plasma, less frequently PC and red blood cell concentrate (RBC). The total of apheresis donations averaged from 50,669 to 125,793 per year (highest number in 2021), i.e. 4.27%–9.40% of all donations, including: Plasma donations: 19 865 - 84 679 per year, with a marked upward trend; the highest number of donations was recorded in 2021. PC donations: 12 969 - 35 735 per year, highest number in 2014; starting from 2015, the number of donations of PC alone decreased, while the number of combined donations of PC and plasma increased (11 778 - 28 966 per year, the highest

number in 2019). GC donations: sporadically (50 - 210 per year, highest number in 2009). RBC donations: sporadically (6 - 488 per year, highest number in 2012), while the number of combined donations of PC and RBC averaged 14 - 282 per year, the highest number in 2020. Like in the previous years, during the review period, whole blood was still most frequently collected (an average of 1,143,582 donations per year, from 1,016,411 in 2008 to 1,251,517 in 2022).

Summary / Conclusions: Worth noting is that automated methods (apheresis) are still used in Poland to a relatively small extent. In 2022 they accounted for only 8.87% of all donations, which is less than in 2021 (9.40%).

P189 | Platelet donation using the apheresis technique

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Background: Platelet donation by apheresis is a useful specific blood components collection method. This technique involves connecting the donor to a machine that cyclically withdraws a small quantity of whole blood, separates the various components, retains the platelets in a bag and returns the uncollected blood components to the donor.

Aims: Our aim was to study the specific characteristics of platelet donors and donations by apheresis (CPA) and to compare the incidence of discomfort with whole blood donation.

Methods: We conducted a descriptive cross-sectional study based on an analysis of CPA collection forms and donation-related incidents during 2023. We reported particularities of apheresis donors and CPA donations as well as collection incidents.

Results: Among the 155 candidates who applied for platelets apheresis, 106 were declared fit to donate, i.e. 68.38%. Donors were exclusively male. The average age of donors was 35 (24–55) with 76% under 40. All donors were related to patients. The donors were in 51.9% civilian and in 48.1% military. Were collected 195 CPA (1.83 CPA/donor). Collection procedures lasted an average of 72 min. We recorded 04 sampling incidents i.e. 3.76% (2 haematomas and 2 vagal discomfort) which forced us to stop the procedure. Discontinuation of the apheresis donation procedure is much less frequent than that of whole blood donation (5.5%). The haematology department received the majority of CPA destined to 45 patients with an average of 4.28 CPA/patient, followed by the gynaecology department (07 patients), the intensive care department and pneumology department (05 patients each) and lastly the urology department. (02 patients). All of these patients were hospitalized at the main military training hospital in Tunis.

Summary / Conclusions: collection of platelets by apheresis offers an opportunity to have a high quality blood compound satisfying patients. Platelets needs requiring specific collection measures including donor care (selection, information, sampling, ...).

P190 | Implementation of donor thrombocytapheresis in the Institute for Blood Transfusion in Montenegro

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Background: Donor thrombocytapheresis refers to the collection of platelets using automatics cell separators from voluntary blood donors. By the procedure of the thrombocytapheresis a therapeutic dose of platelets is obtained from one donor, which it would normally require 6 to 8 blood donors. The therapeutic dose of platelets obtained in this way, reduces the possibility of alloimmunization to thrombocyte and leucocyte antigens. It also reduces the occurrence of refractoriness to platelets, as well as the possibility of transmission of infectious agents. The selection of platelet donors is carried out in a similar way as a selection of donors of classic blood donation. There are identical requirements for blood donors. They include a questionnaire for blood donors, a physical examination by a doctor and an anamnesis. The anamnesis includes personal and family anamnesis about the tendency to bleed, as well as the data of taking antiplatelet drugs. It is recommended that the donor has donated the blood once in the classic way, that he/she is over 18 years old, but not more than 60 and that he/she is over 60 kg. Pre-donation testing of donors include: Testing blood to determine the value of hemoglobin (Hgb > 125 for woman and 135 g/L for man). The value of platelet must be over 150×10^9 . The determination of blood group in the ABO and Rh system. Screening for irregular antibodies. The determination of markers for transmissible diseases (HIV, HCV, HBV and TPHA).

Aims: The presentation of the results of implementation of donor thrombocytapheresis introduced in the Institute of Blood Transfusion of Montenegro.

Methods: The procedure of donor thrombocytapheresis is performed by the using the cell separator. This separator uses centrifugation to collect selected component (platelet) and return the rest red blood cells and blood plasma to the donor and uses one venous access with discontinuous process of taking and returning the blood, but with a continuous blood flow through the centrifuge. The machine uses micro-processing technology to separate the blood cells. Special sets for one use are used with the use of anticoagulants. Our institute has a team of educated and trained doctors and technicians. They are educated for this procedure. The implementation of this method includes: register of voluntary platelet donors, protocols of determining criteria of temporary or permanent rejection of donors. It includes questionnaires, brochures, posters as well as media support.

Results: In the period from 18.04.2022 to 29.01.2024. 164 donors thrombocytapheresis were performed. Among them 65 donors had A blood type, 38 – blood group O, 30 – blood group B, 29 – blood group AB. The register of voluntary platelet donors has been formed, which currently has 190 candidates. Among the negative reactions of thrombocytapheresis we can report the occurrence of hematoma with two

donors. The procedure was interrupted. We can report also the occurrence of perioral paraesthesia in the procedure with one donor. In this case oral calcium was prescribed and there weren't any vasovagal reaction.

Summary / Conclusions: The crucial importance of the implementation of donor thrombocytapheresis is for adequate care of hematological, oncological and surgical patients in Montenegro. It is also necessary to work on the motivation and registration of new platelet donors, as well as the expansion of registered new donors.

P191 | Trends of increasing platelet count as an indicator of impending anemia in regular plateletpheresis donors—a study from tertiary care oncology centre in India

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Background: Regular plateletpheresis donors are at risk of gradual depletion of iron stores from minimal blood loss during each donation. Impending anemia can cause reactive thrombocytosis, hence monitoring CBC may be helpful, to detect it.

Aims: To demonstrate the potential relationship between reactive thrombocytosis and impending anemia in regular voluntary plateletpheresis donors.

Methods: A retrospective analysis of CBC data was conducted in 70 regular voluntary male plateletpheresis donors over a two-year period. As a part of routine testing CBC is done once in 30 days. The first CBC measurement was considered as baseline and trends of Hb, Hct and platelet (PLT) counts were observed. Donors were characterized into subgroups based on donation frequency. Group I (40-48 donations in 24 months; $n = 19$); Group II (20-39 donations in 12-24 months; $n = 31$) and Group III (6-19 donations in 6-12 months; $n = 20$). These subgroups underwent CBC analysis for 25, 12, 6 times respectively. (Table 1)

Results: The mean age of donors was 40 years; (range: 22-55 years). Group I and II showed statistically significant correlation between decreasing Hb, Hct and increasing PLT count in relation to the number of donations ($p < 0.001$) (Table 2). Among these donors, 21 had Hb levels < 12.5 g/dl on 1-6 occasions (average:3), leading to temporary deferral until the levels rose > 12.5 g/dl. A significant decrease in Hb and Hct from baseline was observed during third and sixth testing

P191 Table 1: Donor Categorization

Category	No. of donations	Frequency of donations
Group I ($n = 19$)	40-48	Twice in a month
Group II ($n = 31$)	20-39	Once in a month
Group III ($n = 20$)	6-19	Once in two months

P191 Table 2: Correlation of Platelet count with Hb and Hct in different categories

Donor categorization (n = 70)	Variable name	Mean (Range)	Correlation coefficient (r)	p-value
Group I (n = 19)	Mean Hb (g/dl)	13.74 (11–16.8)	–0.92	<0.001
	Mean Plt ($\times 10^3$ /ul)	317.6 (165–519)		
	Mean Hct (%)	41.83 (34.9–50.5)	–0.804	<0.001
	Mean Plt ($\times 10^3$ /ul)	317.6 (165–519)		
Group II (n = 31)	Mean Hb (g/dl)	13.68 (11–16.1)	–0.944	<0.001
	Mean Plt ($\times 10^3$ /ul)	314.11 (156–498)		
	Mean Hct (%)	41.13 (36.2–45.8)	–0.925	<0.001
	Mean Plt ($\times 10^3$ /ul)	314.11 (156–498)		
Group III (n = 20)	Mean Hb (g/dl)	14.12 (12.5–17.1)	–0.628	0.182
	Mean Plt ($\times 10^3$ /ul)	281.28 (173–448)		
	Mean Hct (%)	42.46 (37.1–53.3)	–0.692	0.128
	Mean Plt ($\times 10^3$ /ul)	281.28 (173–448)		

interval for Group I and II respectively, both coinciding with more than six donations.

Summary / Conclusions: The study suggests, monitoring the CBC trends in regular plateletpheresis donors should be done to detect impending anemia. Prolonging inter-donation interval can mitigate the declining Hb levels and periodic iron studies every sixth donation can provide valuable insights. The increasing platelet count thus, emerges as a potential indicator of anemia.

Blood donation—donor adverse events

P192 | Incidence of adverse events during platelet apheresis collections in single and double collections

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Background: The Transfusion Center of the Community of Valencia (TCCV) implemented the Trima[®] Accel Apheresis device in 2021. First analysis of procedural data after 6 months of routine use showed that some donors who qualify to donate double platelet products (6×10^{11}) were donating per default singles (3×10^{11}). Reasons given

by the operators for not choosing the proposed double procedures was the duration of procedure.

Aims: To compare the experience of same apheresis donors when donating singles and double platelet products.

Methods: Procedural data from the Trima devices were captured using the Cadence System; operational data collected at three sites were managed with TOMES which established a bi-directional connectivity of Trima devices and the blood bank information system (BBIS). Donors were selected for analysis if they donated at least once a single and once a double platelet product in the period between 01-01-2022 and 31-12-2023. Average duration time was measured for each donor individually for single and double procedures. The difference in procedural time between doubles and singles was calculated for each donor individually and the average difference in time between doubles and singles was calculated using the donors individual average values. TOMES connected to Trima Accel was used to capture adverse reactions (AR) at the end of every procedure.

Results: A total of 214 donors were identified who donated single and subsequently double platelet products: 180 males and 34 females. A total of 1421 procedures were analyzed: 1237 with male donors and 184 with female donors. In this cohort average platelet yield was 3.17×10^{11} and 6.08×10^{11} for single and double products, respectively. Procedures resulting in single platelet products ($n = 765$) took in average 47 min, those resulting in double products 66 min. The average difference between double and single procedures was 22 min. Rinseback, which is the return of the blood from the channel back to the donor was completed in 97.8% of single and 99.5% of double procedures ($p < 0.05$). Adverse reactions (AR), such as dizziness, nausea and vomiting and loss of conscience happened in 1.3%, 0.26% and 0.13%, respectively of single procedures and 3.7%, 0.3% and 0% of double procedures. Citrate reaction, as paresthesia and tetany were reported in 5.1% and 0.26% respectively of single collections and in 13.1% and 0% of double collections. Differences in rates of dizziness and paresthesia were significant between single and double procedures ($p < 0.05$). Only two events of paresthesia

led to the interruption of the procedure: one in a single and one in a double collection. Moreover, paresthesia was generally very mild; 98.8% of the double procedures were completed and 86% of donors with reported paresthesia did return to donate again. Prophylactic administration of calcium was mostly therapeutically performed (6.9%).

Summary / Conclusions: Bi-direction connectivity of the Trima devices with the BBIS enabled this blood center to evaluate outcomes of apheresis collections. Reported ARs, such as paresthesia were very mild, since most procedures were completed with rinseback and most donors did return to donate another time. Discomfort in donors with recurring paresthesia symptoms may be prevented in the future with the prophylactic calcium administration.

P193 | Profile of the donor with an adverse donation reaction in the Oporto Blood and Transplantation Center

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Background: The notification of incidents related to blood donation in the Portuguese Hemovigilance System (PHVS) is a legal requirement. Portuguese hemovigilance has been active since 2008, after the publication of decree-law n°267/2007. It covers the entire transfusion chain, from donor to recipient, and also the collection of data information relating to the activity of blood services. Blood donation is usually considered a safe process, with a relatively low rate of adverse reactions. However, these can negatively influence donor recruitment and loyalty to the cause.

Aims: The main objective of this work will be to establish the profile of donors with an adverse donation reaction (ADR) at the Oporto Blood and Transplantation Center (OBTC).

Methods: Notifications of ADR in blood donors were analyzed at the OBTC in the period between January 1, 2019 and December 31, 2022. These data were obtained from computer records relating to the OBTC at the PHVS. Subsequently, a database was built in a spread sheet program, where a cross-sectional study was carried out, observable through descriptive records. The following variables were studied: age; sex; hemoglobin value; weight; donation date; standard donation (SD) and cellular components; number of previous donations; recurrence; moment of ADR detection; severity of ADR; donation complications; need for medication; type of evolution and loss of consciousness.

Results: During the period of the study, 1097 ADR's were registered in 283,081 donations at the OBTC (0.4% of donors). 70.46% occurred in females and 29.54% in males. 97.99% were in standard donations, 1.91% in single-component apheresis donations and 0.09% in multi-component apheresis donations. 82.59% occurred in Mobile Blood Collection Session (MBCS), 15.50% occurred in Blood Collection Session in our own facilities (BCS) and 1.91% in mobile unit (MU). 97.45% of ADR's were non-serious and 2.55% serious (according to Hemovigilance criteria). 57.79% of donors required medication to reverse and 42.21% did not. 49.50% of ADR's progressed quickly, 48.68% progressed slowly and 1.64% required EMA (external medical assistance). In 42.11% of

donors, ADR occurred after needle removal and in 29.99% from introduction to removal of the needle. 20.15% of ADR's occurred at the meal location and 6.93% outside the donation site. 7.93% were relapses and 92.07% were single episodes. Regarding age, weight, hemoglobin value and number of previous donations, it was found that the donor with ADR was on average 33 years old, weighed 67 kg, had 14.6 g/dl of hemoglobin and had 5 previous donations. Complications resulting from the donation: immediate vasovagal reaction (IVVR) 57.06%; Immediate vasovagal reaction with loss of consciousness (IVVLC) 16.96%; delayed vasovagal reaction (DVVR) 3.01%; Painful arm 1.46%; Hematoma 17.5%; Post-donation hemorrhage 2.92%; Other 1.09%.

Summary / Conclusions: A higher frequency of ADR's was found in standard blood donations, female, 33 years old, 67 kg, hemoglobin value 14.6 g/dl, with 5 previous donations, without recurrence and more frequently in Mobile Blood Collection Session. Higher incidence of immediate VVR, occurring after removal of the needle, with rapid evolution, whose reaction was not serious but required the use of medication. The implementation of PHVS facilitates registration, allowing more reliable data to be obtained in a timely manner, allowing measures to be taken to minimize these situations.

P194 | Using Estimated Blood Volume (EBV) to predict the risk of vasovagal reactions in whole blood donors in Singapore

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Background: The maximum amount of blood collected from whole blood donors in Singapore is set at 10.5 ml/kg to prevent volume loss induced vasovagal reactions. Even with this restriction, the volume collected may exceed 15% of the donor's estimated blood volume (EBV) which is equivalent to Grade 2 shock or higher and result in vasovagal reaction. Vasovagal reaction (VVR) is the most common adverse reaction reported in blood donors in Singapore and it is important to prevent VVRs and identify donors who are at risk of such adverse reaction. To prevent VVRs, some blood centers in the UK and USA use the EBV of the donor as an additional donation acceptance criteria and donors with an EBV of <3.5L are deferred from donation.

Aims: To aid in our decision-making on inclusion of estimated blood volume (EBV) $\geq 3.5L$ as a donation acceptance criteria, we determined if male and female whole blood donors in Singapore have a higher risk for vasovagal reaction (VVR) if their EBV is <3.5L.

Methods: The donor history assessment questionnaire of all whole blood donors who donated from October 16, 2023 to January 16, 2024 at a fixed blood collection site and mobile drive were reviewed. Deidentified data on the age, sex, weight, height were collected and donors who experienced a VVR were identified. The estimated blood volume was computed using Naedler's formula. The VVR rates and risk ratio were calculated in male and female donors with an EBV below and above 3.5L.

Results: A total of 6753 whole blood donors (57.2% male and 42.8% females) were included. The overall vasovagal reaction rate is 3.5%

P194 Table 1: Data on the EBV or male and female donors

	EBV<3.5L	EBV≥3.5L
Females w/ VVR	63	72
Males w/ VVR	0	98
Females w/o VVR	246	2507
Males w/o VVR	9	3857

(223 cases), all reactions were categorized as mild and there were more female donors ($n = 135$) who experienced VVR compared to males ($n = 98$). Additional information can be seen in table 1. The risk ratio for VVR in female donors with an EBV <3.5L is approximately 7.53 times higher compared to those with an EBV ≥3.5L (p -value is < 0.00001. Significant at $p < 0.05$). The risk ratio for males is zero.

Summary / Conclusions: Although most of the VVRs in blood donors are mild and self-limiting, the experience itself may discourage future donations. Hence, measures to identify donors at higher risk are important for prevention. We found that female donors with EBV <3.5L are at higher risk for VVRs compared to those with an EBV of ≥ 3.5L. As such, deferral of donors with EBV <3.5L may be considered to prevent potential VVRs in female blood donors. However, deferral of such female donors is not ideal since it may affect the blood supply. We need to evaluate additional risk factors in female donors with EBV <3.5L (e.g. identify certain age groups with higher risk) to further refine the criteria and find a better approach to safeguard the health of donors at risk of VVRs without affecting the blood supply.

P195 | Convulsive and other severe donor reactions—analysis of 27 cases of severe adverse donor reactions

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Background: Blood donation is generally a well-tolerated altruistic event, but, occasionally, adverse reactions of variable severity occur during or on completion of collection. While mild donor reactions like itching, rashes, mild vaso-vagal attacks, hematoma are mostly well tolerated and generally accepted by donors, severe donor reactions like convulsions, loss of consciousness, severe vomiting, severe tetany, involuntary passage of urine and faeces generally create panic among both donors and the transfusion medicine specialist.

Aims: Aim of this study was understand the causes of severe adverse donor reactions and possible ways to mitigate the same.

Methods: A prospective study was conducted over a period of 2 years and 6 months years from 1st July 2021 to 31st January 2024. Only donors who donated blood in-house on a controlled environment were taken for study. Donors who have donated at blood donation camps were excluded to avoid variations in donor selections and environment etc.

Results: Study consisted of 12,565 whole blood donors (11,716 males and 849 females). Severe donor reactions were noted in 27 (0.21%) of

12,565 total donors. All the donors who suffered severe reactions were males. Donor age range was between 22 years and 40 years with 33 years with an average age. Severe adverse reactions noted were sweating with loss of consciousness-13 (48.1%), Tonic-clonic convulsions -4 (14.8%), Vomiting -3 (11%), Severe tetany-5 (18.5%) and involuntary passage of faeces or urine-2 (7.4%). The average BMI was 21. Prominent vein and muscular physic with an average blood collection rate of 100 mL/min or more could have been the main cause of acute hypovolumic/vasovagal shock, possibly leading on to severe adverse reactions. All the blood donors were asked to drink 500 mL of water prior to donation as a routine and no correlation to time of food intake was noted among the participants with severe adverse events.

Summary / Conclusions: In all these severe adverse donor reactions, two parameters remained strikingly important. (1). All the donors who had severe adverse reactions to blood donation were young males (2). The blood collection flow rate was on an average 100 mL per minute or more was recorded. It is nevertheless desirable to reduce risks to a minimum, a set of advice, particularly in donors with muscular physic and prominent veins, keeping the blood collection rate below 100 mL per minute will surely mitigate this problem. Decreasing the distance between the donor arm and the primary bag or adding a flow rate monitoring device might help.

Limitations of the study: The sample size is small. A multicentric study with much bigger sample size will bring out the correlation between rapid blood collection and severe donor reactions. Further studies can also guide us regarding the maximum volume of blood that could be collected per minute, during whole blood donation.

P196 | Abstract withdrawn

P197 | Adverse reactions to whole blood donation—experience from a regional blood centre in Peshawar, Pakistan

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Background: Donating blood is generally a safe procedure, and donors have excellent tolerance for the process. Nevertheless, undesirable adverse reactions (ARs) of varying degrees may occur for some donors during or after the blood donation process. The recruitment and retention of donors may be negatively impacted by these ARs. Vasovagal reaction (VVR) is the most frequent acute adverse event, which can occur with or without loss of consciousness. VVR may cause an accidental fall that results in injury. According to reports, the frequency of ARs in blood donation ranges from 0.6 to 36%, and VVR makes up about 75% of them. According to earlier research, the following factors mostly influence donation-related VVRs: weight, age, gender, first-time donor status, low body mass index (BMI), high blood pressure, fast heart rate, and insufficient sleep.

Aims: Ensuring the safety of donors is crucial for preserving a sufficient blood supply. The aim of this study was to examine the frequency of adverse reactions in blood donors. Other objectives, in addition to determining the frequency, include finding out if ARs are more common during first-time donations or in family replacement donations and if the volume of blood donated (450 or 500 mL) has an effect on how frequently ARs occur.

Methods: A retrospective cohort analysis of whole blood donors from September to December 2023 was conducted at the Peshawar Regional Blood Centre in Khyber Pakhtunkhwa province, Pakistan. The donors were categorised into two groups: Group 1 included 8871 donors who donated in September and October, while Group 2 included 9230 donors who donated during the months of November and December. There was no change in the standard process for blood donations during the study period except that Group 1 blood donations were collected in 500 ml blood bags (with 70 mL CPDA-1) while Group 2 donations in 450 mL blood bags (with 63 mL CPDA-1). Documentation of any ARs was done as per national quality control guidelines of 2020. Microsoft Excel spreadsheets were used to computerise the data, and SPSS for Windows, version 25.0, was used to do the statistical analysis.

Results: The first-time donors in Groups 1 and 2 were 51.88% ($n = 4,603$) and 45.92% ($n = 4,239$), respectively. Similarly, the frequency of family replacement donors in Groups 1 and 2 was 84.90% ($n = 7,532$) and 81.70% (7541), respectively. The results showed that first-time donors (3.16% vs 1.08%), family replacement donors (1.89% vs 0.97%), and those in Group 1 (2.32% vs 1.49%) exhibited a higher AR rate comparatively ($p < 0.0001$). Common mild ARs were dizziness, nausea, and sweating without loss of consciousness. Moderate to severe ARs observed were short-term loss of consciousness, sweating with loss of consciousness, severe headache, and vomiting.

Summary / Conclusions: Based on the findings of this study, we can sensitise our staff at blood collection sites regarding risk factors for ARs in blood donors to prevent them. Limitations include a single-center study of a shorter duration; under-reporting and/or reporting errors are expected as haemovigilance is still in a nascent stage in the province. A multi-centre study with a larger sample size is recommended.

P198 | Abstract withdrawn

P199 | Adverse reactions to blood donation—phlebitis - a case study

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Background: Blood donors adverse reactions (DAR) are any unintended response in donors, associated with the blood collection. These can be classified as local, directly caused by the venipuncture, or systemic: vasovagal reactions. In terms of temporal relationship, DAR can be classified as immediate or late if they occur respectively inside or outside the collection site. This case study investigated a

delayed local vein inflammation reported by a donor. Phlebitis is a local symptoms complication in which inflammation appears along the path of a vein and can progress to localized infection, usually requires medical attention and the lack of early treatment, may cause a major local complication. Imputability and severity was inferred and the impact on the donor's experience.

Aims: Reflect about the existence of unidentified late adverse reactions, their impact on donor's experience and the role of blood centers nurses in post-donation follow-up.

Methods: Case study with documentary analysis using the blood service's internal records, photos and medical reports provided by the donor. Application and qualitative analysis of a form to assess the knowledge, feelings, emotions and impact of the adverse event on the donor.

Results: For a better understanding of the case, the data will be presented chronologically starting on the date of the blood collection that preceded the DAR until the study results (present). On Dec, 23 2020 a 34 year old male, 4th blood collection, did a whole blood donation at a mobile session, according to him the whole process went well. Seven days after (Dec, 30 2020) he presented arm-pain and redness in the venous tract. Donor searched for treatment and received medical assistance and the diagnosis of superficial vein thrombosis. Anti-inflammatory and anticoagulant therapy was initiated for a week. No contacts were made to the blood center. After 29 months, this donor decided to repeat blood donation. He started to describe the episode and showed concern and anxiety due to the past facts. The case was flagged for study due to its relevance to the donor haemovigilance in general. In June 2023 the donor was contacted and given informed consent for the study. The nurse team drew an original form to assess the blood collection experience and sent it to the donor after ethical authorization. In August 2023 the donor's form was analyzed. The donor was able to identify emotions such as doubt, fear, admiration and presence of sequelae was mentioned as the greatest concern. Regret was shown without questioning the nursing technician's procedure, but assuming that this event caused insecurity and reluctance to repeat the blood donation.

Summary / Conclusions: This case study identified a phlebitis but also the impact of that adverse reaction on the well-being of the donor. Donor haemovigilance should be a systematic monitoring with a view to improve quality and safety to blood donors, so it is essential to put the person at the center of the process, providing education and establishing effective means of communication. Immediate adverse reactions are recognised and responded in the collection site, but late reactions need to be recognized by the donors or community. Nurses have a fundamental role as health educators and in transmitting post-donation information that empowers donors in this context. By better identifying late reactions, the individual donor experience is improved, loyalty is promoted and positive word-of-mouth is facilitated with a real impact on the gain of new donors.

P200 | Adverse effects in blood donors and room for improvement—past period review**Z Bolta**¹¹*Transfusion centre, General hospital Celje, Celje, Slovenia*

Background: Adverse effects in blood donors are, though in majority minor and unfrequent, still important and in many ways preventable. Transfusion centre, General hospital Celje performs up to 9000 whole blood draws annually, 45% at field draws. In poster we review past data in adverse reaction occurrence and experience in managing them at our establishment.

Aims: Additional to the nationally and internationally accepted recommendations considered before donations, we show the standard preventive measures and informing taken before blood donation, including informed consent, respecting the in house hospital policy. We also present decades long standing educational program efforts as performed by blood establishment regularly, focused on new blood donors in high schools. In new donors we present dedicated quality control marker followed in new donors as represented with trends.

Methods: We compare the type of adverse effect from systemic fainting and convulsions through local physical injury by haemathoma or direct injury by venepuncture analysing the frequency and trends through past period. Outcome of different adverse effects is shown systematically as regularly performed in annual hemovigilance reports. Separately we analyze serious adverse reactions as reported to the national competent authority by type and frequency.

Results: Special effort is taken to perform the follow up and additional diagnostics of the affected blood donors by counseling and organizing when needed. We present typical path taken in case of neuropathy. Besides the interventions taken intra donation we present continuous education of the draw team to offer best possible care. We also present the nationally accepted way of prevention and managing acute post donation adverse effects through surveillance and help during refreshment meal postdonation and organization of work free day for donors.

Summary / Conclusions: Thorough care for blood donors is of paramount importance for our blood establishment, by constantly analyzing and improving our primary goal is to offer best possible multilevel care for blood donors.

P201 | Analysis of serious adverse reactions in donors in Regional Blood Donor Center in 2023**A Łaba**¹, R Klupieć¹¹*Regional Blood Donor Center, Poznań, Poland*

Background: In accordance with the national regulations every serious adverse reaction related to donation of blood or its components must be reported to the supervising authority in Poland i.e. to Institute of Hematology and Transfusiology in Warsaw within 24 h. Adverse reactions in donors can be related to donating whole blood or its components by means of apheresis. Adverse reactions can be divided in

3 categories depending on the grade of intensity of symptoms and their severity: from grade 1 (mild) to grade 3 (severe). Serious adverse reactions are reactions of grade 3 that require indisputable treatment and/or hospitalization. Immediate vasovagal reactions are the most common adverse reactions in donors and they account for over 80% of all adverse reactions in donors. Immediate reactions occur shortly after donation of blood or its components, still in the care of the medical personnel.

Aims: The aim of the thesis is to analyse serious adverse reactions that occurred in donors in Regional Blood Donor Center in 2023.

Methods: The analysis has been carried out using rapid alert notifications about suspected serious adverse reactions related to donation of blood or its components; confirmatory reports regarding serious adverse reactions related to donation of blood or its components; blood donor serious adverse event report forms; blood donor questionnaires; medical history regarding donors and their donations in the system Bank Krwi (system in operation in Regional Blood Donor Center in Poznań).

Results: In 2023 three serious adverse reactions occurred in Regional Blood Donor Center in Poznań, which accounts for 0.77% of all adverse reactions in that year. All reactions took place after donation of whole blood and were the immediate vasovagal reactions. Two reactions occurred in the satellite branch, one in the main seat. One serious adverse reaction occurred in first-time donor, two reactions in repeat donors with no history of previous adverse reactions. Donors were approved for the donation by the physician after medical interview, analysis of the laboratory test results and medical examination. Blood pressure of all donors was normal and in the medical interview they confirmed proper preparation for the donation (low-fat meal and good hydration). Two donors were released home after short stay in the E.R, whereas 1 donor was admitted to hospital after diagnostic tests in the E.R. Two out of three donors confirmed full convalescence after the event, one donor reported mild consequences of the serious adverse reaction which manifested in some pain.

Summary / Conclusions: Donating blood and its components is a safe medical procedure. However, it can still result in adverse reactions, including serious ones. Constant education regarding proper conduct before and after donation targeted at all groups donors: first-time and repeat donors is essential in order to minimize the risk of occurrence of serious adverse reactions. The whole personnel must be trained on a regular basis (including every single step from the registration to the final blood collection) and it is also crucial for them to be aware of how important it is to follow current procedures.

Blood donation—blood donor biorepositories and public health research

P202 | Abstract withdrawn

P203 | Enhanced SARS-CoV-2 reinfection detection using expanded dynamic range anti-nucleocapsid antibody testing in repeat blood donors

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Background: Serosurveillance of blood donors has contributed to SARS-CoV-2 public health surveillance in many countries. Currently, most individuals have experienced at least one SARS-CoV-2 infection, and case reporting has declined, making reinfection (RI) detection an important public health surveillance tool. Robust RI detection is required in the context of existing immunity in those previously vaccinated and/or with one or more prior infections. A previously reported method for monitoring RI incidence based on longitudinal testing of blood donors and anti-nucleocapsid (anti-N) antibody (Ab) boosting was refined to address reduced sensitivity in cases where the pre-RI anti-N reactivity was high.

Aims: To assess the performance of anti-N Ab testing for detection of SARS-CoV-2 RI with and without dilutional testing.

Methods: Using an anti-N total immunoglobulin (Ig) assay, Ab boosting was measured by the ratio of reactivity in subsequent specimens from a donor, to determine whether an RI occurred in the inter-test interval. Optimal Ab boosting thresholds were identified using receiver operating characteristic curve analysis in a longitudinal dataset of previously infected donors. RI cases were donors reporting a swab-confirmed infection ≥ 90 days after a prior infection, defined as a swab-confirmed infection or anti-N seroconversion. Controls were donors with first infections identified serologically in 2020, when RI was rare. Two thresholds were derived for evaluation: weighting sensitivity and specificity equally, and weighting specificity higher. Using a dilutional algorithm, longitudinal samples from RI cases were further tested to evaluate the performance of expanded dynamic range testing with the Ortho VITROS anti-N total Ig assay. Samples were initially tested undiluted, reflexed to testing in 1:20 dilution if the signal to cutoff ratio (S/CO) ≥ 100 , and further diluted 1:400 if dilutional S/CO ≥ 2000 . Sensitivity was estimated, stratified by pre-RI reactivity using the undiluted results alone, and with dilutional results.

Results: Optimal post-RI to pre-RI reactivity ratio thresholds were ≥ 1.4 , using equal weighting, and ≥ 2.1 , using specificity weighting (which maintained specificity $\geq 99\%$). Median observed anti-N reactivity ratios were significantly higher when dilutional results were used (table). Using the ≥ 1.4 criterion, sensitivity declined to 51.3% in the pre-RI S/CO > 150 group, and when using the ≥ 2.1 criterion, sensitivity declined to 42.2% in the > 100 -150 group and to 0.0% in the > 150 group (table). In contrast, sensitivities of 89.7% and 76.9% were observed when using dilutional data for the two criteria, respectively, in the > 150 group. Overall sensitivity when using ≥ 2.1 improved from 71.8% to 85.5% with dilutional testing.

Summary / Conclusions: Expanded dynamic range anti-N Ab testing is critical to maintain sensitivity of serological detection of RI where pre-RI reactivity is high and is especially important for detection of multiple RIs. This approach requires a longitudinal cohort such as repeat blood donors. Testing approaches, assays, and RI detection criteria may need to be adjusted with changes in the immunologic landscape.

P203 - Table 1

Pre-RI S/CO (N)	Median ratio Neat Dilutions	Ratio ≥ 1.4 (Sp = 96%)	Ratio ≥ 2.1 (Sp = 99%)
		Sensitivity (%) Neat Dilutions	Sensitivity (%) Neat Dilutions
All (436)	4.7 34.3	84.4 88.8	71.8 85.1
≤ 50 (256)	13.6 118.9	88.3 88.3	87.1 87.1
> 50 -100 (77)	2.9 30.3	87.0 87.0	81.8 87.0
> 100 -150 (64)	1.9 22.1	85.9 92.2	42.2 79.7
> 150 (39)	1.5 13.4	51.3 89.7	0.0 76.9

P204 | Impact of sampling intervals on the assessment of Omicron infection rate by ratio-based anti-N signal analysisR Bazin¹, A Lewin², J Perreault¹, Y Grégoire¹, C Renaud², M Germain¹¹Medical Affairs and Innovation, Héma-Québec, Québec, ²Medical Affairs and Innovation, Héma-Québec, Montréal, Canada

Background: The ratio-based analytical approach for SARS-CoV-2 anti-nucleocapsid (N) antibody results is a new, highly sensitive approach that successfully addresses the issue of vaccination in SARS-CoV-2 serosurveys. Compared with conventional methods using a single absorbance threshold for seropositivity determination, this ratio-based approach significantly improves the identification of vaccinated individuals with a recent history of SARS-CoV-2 infection (95.2% vs. 63.3%). The ratio-based approach relies on longitudinally collected samples and determines seropositivity by an anti-N absorbance ratio greater than 1.5 between two consecutive samples. In addition, the approach effectively detects SARS-CoV-2 reinfections, defined as a sequence of anti-N ratios ≥ 1.5 followed by at least one ratio < 1.5 and then returning to ≥ 1.5 . Nevertheless, the influence of the interval between consecutive samples on the sensitivity of the approach remains to be elucidated.

Aims: Determine the impact of different intervals between consecutive samples on the identification of new infections and reinfections using the ratio-based analytical approach.

Methods: A cohort of 254 plasma donors was followed throughout the first 18 months of the Omicron wave (December 2021 to July 2023). Anti-N levels were measured in samples collected in December 2021 and every 3 months thereafter, using our previously described in-house anti-N total Ig ELISA. We then used the ratio-based analytical approach to determine the number of infections and reinfections, analyzing the samples separated by a 3-month interval. A similar assessment was carried out with samples separated by an interval of 6 or 9 months.

Results: Using the ratio-based approach with samples taken at 3-month intervals, the cumulative infection rate was 115.7%, including a reinfection rate of 28.3%. Increasing the interval to 6 months, the estimated cumulative rate was 96.5%, with a reinfection rate of 11.4%. When samples were spaced 9 months apart, the cumulative infection rate fell to 77.6%, without the possibility to identify reinfections, since only two periods were evaluated. In comparison, the cumulative rates assessed using the ratio-based approach with samples collected at the start of the Omicron wave (December 2021) and at the end of the study period (July 2023), or using the conventional approach applied on the July 2023 samples, were 63.8% and 38.6% respectively. On the other hand, if we consider the number of individuals infected (once, twice or more during the Omicron wave) instead of the total number of infections, the cumulative infection rate was 87.4% (3-month interval), 85.0% (6-month interval), 77.6% (9-month interval), 63.8% (overall 18-month period) and 38.6% for the conventional approach.

Summary / Conclusions: This study shows that intervals of 3 to 6 months between consecutive samples provide a valid estimate of the number of individuals infected during the 18-month study period,

using the ratio-based analytical approach. Longer intervals between samples are associated with a decrease in infection detection sensitivity. In addition, the short interval of 3 months makes it possible to effectively identify cases of reinfection and their evolution over time, whereas longer intervals are not appropriate for this assessment.

P205 | Incidence of SARS-CoV-2 during the Omicron wave—results of a longitudinal serosurvey in Québec, CanadaR Bazin¹, A Lewin², M Germain¹, Y Grégoire¹, G De Serres³, C Renaud²¹Medical Affairs and Innovation, Héma-Québec, Québec, ²Medical Affairs and Innovation, Héma-Québec, Montréal, ³Institut national de santé publique du Québec, Québec, Canada

Background: Conventional serological approaches using a single blood specimen lack sensitivity for the detection of recent SARS-CoV-2 infections in vaccinated individuals, as these individuals exhibit a blunted anti-nucleocapsid (N) response. This limitation was recently addressed by the development of a "ratio-based analytical approach", which compares longitudinally collected specimens.

Aims: Estimate the incidence of SARS-CoV-2 infection and reinfection in Québec (Canada) during the Omicron wave using the ratio-based analytical approach.

Methods: Consenting plasma donors were included if they donated plasma before December 15, 2021 and during six consecutive periods of 2-3 months between December 15, 2021 and July 7, 2023 (study period). Anti-N were measured using a previously described in-house ELISA. Anti-N seroconversion was characterized by a ratio of ≥ 1.5 between the optical density of two consecutive samples.

Results: Among the 254 participating donors, the adjusted risk (95% CI) of infection ranged between 18.1% (13.2%–23.0%) and 24.2% (18.8%–29.7%) over Periods 1-5 and fell to 7.9% (4.9%–11.0%) during Period 6. During the study period, the risk of infection decreased among donors aged < 60 (Period 1 = 31.6%, Period 5 = 4.4%), but increased among those aged ≥ 70 (Period 1 = 0.3%, Period 6 = 10.3%). Throughout the study period, 72 (28.3%) possible reinfections occurred (i.e., two seroconversion events in a single donor). Overall, 87.4% (95% CI = 82.7%–91.2%) were infected by SARS-CoV-2 at least once during the study period.

Summary / Conclusions: The vast majority of the Québec population have been infected during the Omicron wave. This longitudinal survey demonstrates the usefulness of the "ratio-based approach" for identifying both new infections and reinfections within a vaccinated population.

P206 | Abstract withdrawn

Blood product / components— blood processing, storage and release

P207 | Implementation of the automatic blood bag processing
system (BBPS) at Sanquin Blood Bank

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Background: Within Sanquin Blood Bank in the Netherlands 400.000 red blood cells are released by operators in the processing department. Before releasing these products, the crossmatching tube (cross tube) of the blood bag is segmented into 8 sections, folded and attached to the blood bag with a rubber band as per the request of our hospitals. These manual procedures contribute to onset of repetitive strain injury.

Aims: The aims of the project were to automate processing, to become more efficient, to reduce repetitive manual actions, to become more consistent and to completely eliminate the risk of human error. After separating whole blood into its constituents, the BBPS should process the red blood cells as follows: Strip the cross matching tube, making sure it is filled homogeneously. Seal the cross matching tube into separate segments. Bundle the segments (zigzag) and fasten to the back of the blood bag. Release/label the bag using our Blood Bank Information System (eProgesa). Sort the blood bags according to the attached labels.

Methods: The following steps were taken to design and build the machine according to our specifications and Good Practice Guidelines. User Requirement Specification (URS). Design of the machine and building at the supplier (DEC group). Documentation of the design, building, change in process. Interfacing the machine with eProgesa. Factory acceptance

testing (FAT). Installation at Sanquin location Amsterdam. In-process Testing and Site Acceptance Test (SAT). Operator training and awareness.

Results: We designed a trolley which can contain 91 bags of red blood cells as input to the BBPS. Robot Arm 1 of the BBPS takes a bag from the trolley and presents it to a scanner which verifies the bag type. The bag is then placed upon a conveyor belt where the cross matching tube will be stripped with mechanically assisted rollers and sealing of the segments takes place. The cross matching tube will be automatically folded in a zig zag manner and the bundle will be fastened to the back of the blood bag using special tape. The bag continues on the conveyor after the bundle has been attached. Robot Arm 2 takes each bag to a scanner which images the unique information on the blood bag. eProgesa uses the scanned information to generate a label using a zebra printer. The new label is attached to the blood bag and released/validated. Robot Arm 2 also sorts the blood bags based on labelling control result and blood group. If the bag has not been fully validated it is directed to a “work in progress” bin. In the below table the time saved using the BBPS is summarized. The process time is given in products per min, hours per day and hours per year. These calculations are based on average of 1 FTE and processing of 1600 red blood cell bags per day.

Summary / Conclusions: We reached our goal and implemented an automatic process for stripping, segmenting, bundling the crossmatching tube and releasing red blood cell bags and reduced manually repetitive movements in this process.

P207 – Table 1

Manual Process	Products/min	Hours/day	Hours/Year
Segment cross matching tube and bundling	0.5	13.3	3.200
Releasing red blood bags	0.3	8	1.920
Total time manually	0.8	21.3	5.120
Automated Process	Products/min	Hours/day	Hours/Year
BBPS	0.42	11.2	2.688

P208 | Buffycat pooled cold stored platelets as compared to conventional room temperature platelets—an in-vitro study of platelet metabolism and function

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Background: Recent literature states that cold-stored platelets have a longer shelf life, better hemostatic function and lesser chances of bacterial contamination as compared to room temperature stored platelets.

Aims: This study was done to assess platelet metabolism and function of cold stored platelets with and without platelet additive solution for up to 21 days as compared to conventional room temperature platelets.

Methods: We pooled 16 ABO compatible Buffycoats to prepare four final buffycoat platelet bags: part A, B, C and D. Part A & B were suspended in plasma, and part C & D were suspended in PAS. Part A and C were kept at Room temperature for five days in an agitator, and Part B and D were kept at 4°C without agitation for 21 days. The Final platelet product A and C (Stored at 22°C) were serially sampled on Day 1, 3, 5, and B and D (Stored at 4°C) were sampled on Day 1, 3, 5, 7, 10, 14, 21. Each sample was tested for parameters like platelet count, WBC count, Swirling, pH, Glucose, Lactate, CD-62, IL-6, TNF- α , Aggregation studies with platelet agonists: ADP and Epinephrine, Clot strength tested by SonoClot® (Sienco®, Inc, Arvada, CO, USA) and sterility by culture on Day 5 for Part A & C and Day 10 and 21 for part B&D.

Results: The cold-stored platelets showed the disappearance of swirling within 1-2 h of placing at cold temperature. The mean platelet counts ($\times 10^{11}$) on Day 1 of storage in parts A, B, C, D were 2.6 ± 0.5 , 2.6 ± 0.4 , 2 ± 0.4 , 2.1 ± 0.5 , respectively. There was no significant fall in room temperature platelets over storage for five days, but the fall was significant ($p = 0.04$) in the cold-stored platelets on day 14 and day 21, with a mean platelet count ($\times 10^{11}$) decreasing from 2.6 to 1.5 and 1.3, respectively. The mean pH in all parts was maintained above 6.4 throughout the storage. The percent aggregation to ADP in Parts A, B, C, D was 66 ± 17 , 57 ± 11 , 56 ± 20 , 53 ± 20 on Day1

which steadily decreased over storage. The decrease of aggregation to ADP in room-temperature platelets was significantly more than in cold-stored platelets. The platelet function by Sonoclot was maintained above 3.0 in all parts stored at room temperature and cold temperature until the last day of storage. The marker of platelet activation (CD-62) increased in all parts during storage, but the increase in room-temperature platelets was steeper than in cold-stored platelets. Activation of cold-stored platelets is less when kept in PAS than when kept in plasma.

Summary / Conclusions: Our study concludes that cold-stored platelets maintain invitro functional viability similar to or even better than room temperature platelets till day 10 of storage both in plasma and PAS. PAS further helps in slowing the platelet storage lesions, and we were able to demonstrate acceptable in-vitro functional viability of cold-stored platelets upto 14 days.

P209 | Maintaining the sterility of welded tubing using a cordless sealer

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Background: The MacoSeal LIGHT cordless sealer allows the sealing of PVC tubing by high-frequency welding of the walls together and the separation of the 2 parts of tubing along the welding zone.

Aims: The objective of the study is to evaluate the maintenance of the sterility of the liquid present inside the tubing when the area of the tube where the welding is carried out is artificially contaminated by a microorganism.

Methods: Systems composed of tubing connected to 2 bags and a filling site were produced and sterilized using 2 methods: steam and beta sterilization. Two types of PVC tubing were tested: 4.1 and 4.5 mm external diameters. The systems were pre-filled with culture medium and the external surface of a portion of the test tubing was contaminated with a reference microbial strain, namely *Bacillus subtilis* (ATCC 6633) or *Staphylococcus aureus* (ATCC 6538) at a concentration of approximately 10^6 colony forming units (CFU). After making the disconnections, the systems obtained were then incubated at a controlled temperature for 7 days in order to detect contamination of the liquid inside them.

P209 Table 1. Results obtained for each condition tested (bacterial strain, tubing size and sterilization). The numeration of inoculum shows the quantity of bacteria inoculated. Compliant positive and negative controls validate the experiments.

Bacterial strain (number of CFU per device)	<i>S. Aureus</i>		<i>B. Subtilis</i>	
Tube diameter: inside \times outside (mm)	3.4 \times 4.5	3 \times 4.1	3.4 \times 4.5	3 \times 4.1
Sterilization	beta	steam	beta	steam
Bacterial growth/ number of trials	0/30	0/30	0/30	0/30
Negative control (bacterial growth/ number of trials)	0/3	0/3	0/3	0/3
Positive control (bacterial growth/ number of trials)	1/1	1/1	1/1	1/1
Compliance percentage (%)	100	100	100	100

Results: The configurations tested for each bacterial strain showed an absence of contamination of the culture medium after sealing and disconnecting the bags (Table 1). Inoculum control tests showed a quantity of bacteria inoculated between $9.5 \cdot 10^5$ and $6.2 \cdot 10^6$ CFU.

Summary / Conclusions: External contamination of beta or steam sterilized tubes by bacterial strains does not lead to contamination of the liquid inside the tube after sealing with the MacoSeal LIGHT.

P210 | Lipaemic plasma—an objective non-invasive photometric method to judge plasma turbidity and its association with red cell stability

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Background: According to the European Pharmacopeia, plasma must have a clear to slightly turbid appearance, without any signs of haemolysis. To judge this, plasma units are visually inspected and non-transparent units are rejected. Moreover, there exists conflicting data on the quality of red cell concentrates (RCC) during storage from donations with fatty plasma. The visual inspection for judgement of plasma turbidity is a subjective method, which might lead to over- or under-rejection of blood products. A quantitative measurement of the turbidity of plasma would result in more objective judgement of plasma units.

Aims: To evaluate: (1). an objective non-invasive photometric method for judgement of turbid plasma units and (2). the role of lipemic plasma on red cell stability.

Methods: A total of 365 recovered plasma units, both clear and turbid were visually inspected for turbidity and analysed for light reflection (L^* value, Konica Minolta CM-5 spectrophotometer) and triglyceride (TG) levels. Leukoreduced RCCs in SAGM, prepared from lipemic whole blood, were stored for 6 weeks and analysed for quality parameters.

Results: A correlation ($R^2 = 0.703$, $p < 0.0001$) was found between TG levels and L^* values. Plasma with $TG \geq 2.5$ mmol/L showed a L^* value >50 and $>90\%$ specificity and sensitivity (Table 1). 89% of the plasma with $L^* < 50$ were visual clear and 98% of the plasmas with $L^* > 50$ were turbid. Of those plasma's that were not visually clear but had a L^* value below 50, the triglycerides indeed were found to be significantly lower (3.2 mmol/L) versus 5.7 mmol/L in the $L^* > 50$ group ($p < 0.0001$). Therefore, RCCs from lipemic donations were divided in 4 groups based on the L^* value (Table 2). RCC quality parameters between the groups were found to be similar, except for haemolysis. The group with an L^* value above 68 showed significantly higher haemolysis levels and more units with haemolysis $>0.8\%$ as compared to the other groups.

Summary / Conclusions: The non-invasive photometric analysis of plasma turbidity correlated with visual inspection and plasma TG levels. Implementation of this method would standardize judgement of turbid plasma and rejection from processing. Visual judgement of turbidity, can be replaced by an objective non-invasive photometric measurement. The light reflection (L^*) showed a good correlation with TG levels in the plasma. Measurement of L^* values of plasma may be helpful to identify WB donations with high TG levels and higher risk for increased haemolysis during RCC storage.

P210 Table 1: The plasma L^* value and triglyceride levels for clear versus lipemic plasmas

	$L^* < 50$ (n = 177)	$L^* > 50$ (n = 188)
Percentage visual turbid	11%	98%
L^* value	45.2 ± 1.9	59.1 ± 5.3
Triglycerides, mmol/L	1.4 ± 0.9	5.7 ± 2.4

P210 Table 2: Quality parameters of stored RCCs, sorted by L^* value of the corresponding plasma (mean \pm SD, # $p < 0.05$ vs $L^* > 63$).

Plasma L^* value	<58 (n = 33)	58-63 (n = 78)	63-68 (n = 53)	>68 (n = 32)
Haemolysis, %	$0.30 \pm 0.15\#$	$0.35 \pm 0.17\#$	0.45 ± 0.41	0.50 ± 0.31
Percentage RCCs with haemolysis $>0.8\%$	0.0	2.6	9.4	15.6

P211 | Automated labelling and verification of blood componentsC Boljesic¹, D Marinakis²¹Manufacturing & Logistics, Australian Red Cross Lifeblood, Brisbane,²Manufacturing & Logistics, Australian Red Cross Lifeblood, Melbourne, Australia

Background: Australian Red Cross Lifeblood labels more than one million blood components for transfusion each year manually. As demand for blood components increases, so too does the manual effort of labelling and the risk of potential mislabelling events. Information contained on these labels include blood component composition and indications for use and storage. This information is used by hospitals to determine donor-to-patient compatibility and transfusion dosage.

Aims: The purpose of the Automated Labelling and Verification (ALiVe) system is to reduce manual handling and improve the integrity of the labelling manufacturing process and reduce the risk of mislabelling events.

Methods: Australian Red Cross Lifeblood contracted Bosch Australia Manufacturing Solutions to develop a “first of its kind” automated machine to label and verify manufacturing labels applied to blood components using robotic automation and system control. Several design iterations were proposed before Lifeblood agreed on the one to proceed with. The design was a simple, innovative solution to meet Lifeblood's labelling needs for refrigerated, frozen and room temperature stored blood components including Red Cells, Fresh Frozen Plasma and Platelets.

Results: The ALiVe machine is interfaced to the National Blood Management System to generate manufacturing labels. Using sophisticated robotics and vision technology, the machine scans, labels, and verifies information contained on the label to ensure that the right label has been applied to the right blood component. The label verification process checks for label defects such as text misalignment, incorrect volumes, and the status of the irradiation label. It can sort and segregate blood components that fail label verification and segregate blood components labelled with rare phenotypes. In addition, the ALiVe has a frost removal function to remove frost from fresh frozen plasma. This is done by applying a temperature-controlled rubber block against the surface of the blood component so that the barcodes can be read by the vision system and the label can adhere. Lifeblood has undertaken extensive validation of these functions to ensure the ALiVe machine meets regulatory requirements and that it is fit for purpose.

Summary / Conclusions: The ALiVe is the first piece of automation of its kind to support the manufacturing teams at Lifeblood with performing the labelling of clinical blood components. Automation is pivotal to Lifeblood's future and transformation journey. It enables Lifeblood to respond to change and supports Lifeblood's ability to improve healthcare across Australia. The Automated Labelling and Verification system: Reduces the risk of potential labelling errors and enhances Lifeblood's ability to deliver high quality, safe blood products. Helps us better respond to customer needs, manage growth, and

prepare us for the future. Reduces the possibility of injury and improve the safety of Lifeblood's people by automating highly repetitive tasks.

P212 | Young RBC subpopulation from frequent senior blood donors has lower hemoglobin-oxygen affinityM Yazdanbakhsh^{1,2}, O Mykhailova², J P Acker^{1,2}¹Laboratory Medicine and Pathology, University of Alberta, ²Innovation and Portfolio Management, Canadian Blood Services, Edmonton, Canada

Background: Red Cell Concentrates (RCCs) contain cells at different stages of their life cycle. The RBC aging process is associated with a reduction of mean corpuscular volume (MCV) and an increase of mean corpuscular hemoglobin concentration (MCHC) due to water loss resulting in a lower surface-to-volume ratio and higher density in old RBCs (O-RBCs). p50 refers to the partial pressure of oxygen (pO₂) in blood at which hemoglobin is 50% saturated with oxygen. Frequent blood donations may alter the quality of blood components by modulating RBC characteristics. As it has been shown that young RBCs (Y-RBCs) have lower Hb-O₂ affinity than old RBCs (O-RBCs), we predict that in frequent donors, there will be a higher portion of Y-RBCs that could affect the Hb-O₂ affinity.

Aims: Evaluate hemoglobin-oxygen affinity of the subpopulation of “young” and “old” RBCs during hypothermic storage as a function of the blood donor age and frequency of blood donation.

Methods: RCCs were collected from healthy frequent teenagers (more than 3 times donation per year) ($n = 5$), non-frequent teenagers (1 donation per year) ($n = 5$), frequent seniors ($n = 5$) and non-frequent seniors ($n = 5$). Samples were percoll-density separated into portions of less dense / recently matured (Y-RBCs) and dense/senescent (O-RBCs). Changes in MCV, MCHC, and p50 were assessed on days 5, 14, 28, and 42 of storage using established methods in our group.

Results: The lowest Hb-Oxygen affinity (highest p50) was related to the Y-RBCs from frequent senior blood donors over all storage periods (21.72 ± 3.01 mmHg). Also, there was a significant difference between Y-RBCs non-frequent seniors (20.89 ± 1.82 mmHg) and non-frequent teenage (16.82 ± 1.10 mmHg) donors on day 42 of storage in all RBC age groups ($p = 0.0038$). The highest Hb-oxygen affinity (lowest p50) was related to the O-RBCs from non-frequent teenage blood donors compared to all other blood groups (17.50 ± 1.90 mmHg) ($p = 0.0013$). The MCHC was significantly higher in non-frequent donors compared to frequent donors ($p = 0.008$). Also, non-frequent teen donors had the highest MCV compared to all other groups in all storage periods ($p = 0.007$).

Summary / Conclusions: Y-RBCs of frequent senior donors have lower Hb-O₂ affinity suggesting more efficient oxygen release post-donation at the baseline timepoint, compared to Y-RBCs of frequent and non-frequent teenage donors. The frequency of blood donations might impact the distribution of Y-RBCs, by inducing Y-RBCs release to blood circulation and influencing the hemoglobin-oxygen affinity of red blood cells. This, in turn, could affect the effectiveness of blood

transfusions, leading to higher oxygen release in recipients receiving blood from frequent donors.

P213 | Abstract withdrawn

P214 | A multicentre European validation study of hypoxic red blood cells

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Background: During storage of red blood cells (RBCs), oxidative damage from exposure to oxygen, in addition to metabolic impairments, can result in a decreased quality of the RBC. Hypoxic storage, where the oxygen content of RBC units is reduced to <20% saturation of O₂ (SO₂) prior to refrigeration and maintained throughout storage, reduces the accumulation of oxidative and metabolic storage lesions, preserves RBC deformability, and improves oxygen off-loading. In preparation for clinical investigations with patients requiring RBC transfusion, validation studies of the *in vitro* performance of a system used to process and hypoxically store RBCs – CPD/PAGGSM Leukocytes-Reduced (LR), O₂/CO₂ Reduced – were executed at regional blood banks in Germany, Italy, Norway and Switzerland.

Aims: To evaluate CPD/PAGGSM LR, O₂/CO₂ Reduced RBCs stored for 42 days after pre-storage O₂/CO₂ reduction and ensure that the product met acceptance criteria in each of the included regions.

Methods: Informed consent was obtained from all blood donors. Each whole blood unit collected generated one unit of O₂/CO₂ reduced LR-RBC. Whole blood was collected and processed using multiple modalities across centres: an automated blood processing system, top-top and top-bottom methodologies. Whole blood processing, including leukocyte depletion, could be performed shortly after collection, within 24 h at ambient hold (20–24°C). All units in the study were processed using the CPD/PAGGSM LR, O₂/CO₂ Reduced RBC storage system and stored for 42 days at 2–6°C. The key endpoint-associated study acceptance criteria at 42 days for all included regional blood banks were:

P214 Table 1: Validation results from regional blood banks

	Baseline	Day 21	Day 42
HCT, mean (SD), %			
Germany	61 (2.40)	62 (2.80)*	63 (2.30)
Italy	61 (2.56)	66 (8.10)	63 (5.59)
Norway ^a	57 (1.69)	–	58 (2.63)
Switzerland	60 (2.20)	–	64 (3.40)
Haemolysis, mean (SD), %			
Germany	0.13 (0.02)	0.19 (0.04) *	0.25 (0.07)
Italy	0.10 (0.03)	0.20 (0.09)	0.25 (0.09)
Norway ^a	0.10 (0.03)	–	0.20 (0.05)
Switzerland	0.10 (0.02) [†]	–	0.41 (0.19) ^b
Hb, mean (SD), g/dL			
Germany	19.6 (0.97)	19.5 (0.93) *	19.6 (0.85)
Italy	20.3 (0.90)	21.8 (2.8)	20.6 (1.80)
Norway ^a	19.0 (0.81)	–	19.0 (1.04)
Switzerland	19.3 (0.90)	–	19.6 (0.90)
Blood culture			
Germany	Sterile	Sterile*	Sterile
Italy	Sterile	–	Sterile
Norway ^a	–	–	–
Switzerland	–	–	–

Note: Germany, N = 26; Italy, N = 30; Norway, N = 33 (Bergen) and N = 21 (Oslo); Switzerland, N = 31.

* Day 23.

[†] N = 26 (bags handled in the pilot unit).

^a Average of results from two blood banks in Norway: Oslo and Bergen.

^b One red cell concentrate had a haemolysis higher than 0.8% (0.81%) at the end of storage but this result remains within the validation acceptance criteria (a minimum of 90% of units tested should meet the required value).

total haematocrit (HCT) >50% and haemolysis at Day 42 of <0.8%. Additional assessments included haemoglobin (Hb) levels and the sterility of the hypoxically stored blood (German and Italian centres only).

Results: Blood units were evaluated after processing with CPD/PAGGSM LR, O₂/CO₂ Reduced storage system, and after 21 and 42 days of storage. Throughout storage to Day 42, SO₂ was maintained at approximately 20%. The most relevant findings are shown in Table 1.

Summary / Conclusions: Hypoxic RBCs were successfully validated irrespective of the different modalities which were used to collect and process the RBCs. These results indicate that RBCs processed and stored under hypoxic conditions satisfy acceptance criteria for transfusion into patients in four European countries.

P215 | A novel viscoelastic testing platform for the assessment of haemostatic function in stored platelets

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Background: A novel point-of-care viscoelastic testing platform (Quantra) utilises SEER (Sonic Estimation of Elasticity via Resonance) sonorheometry to measure changes in the haemostatic properties of whole blood. One of the novel features of this device is that it directly quantifies the platelet contribution to clot stiffness (PCS), accounting for both the platelet count and their ability to aggregate, contract, and contribute to clot strengthening. As such, it may allow for a more comprehensive understanding of how storage-specific changes (time and temperature) in platelets contribute to the mechanics of clot formation, in the context of an *in vitro* whole blood model of transfusion.

Aims: To assess the haemostatic properties of platelets stored under different conditions (temperature and duration), using a novel viscoelastic platform.

Methods: Apheresis platelet donations were stored under the following conditions: room-temperature (20–24°C; RT), refrigerated (2–6°C; cold), and cryopreserved (–80°C with 5–6% DMSO; frozen). RT and cold platelets were tested over a 21-day period, while frozen platelets were examined immediately after thawing and reconstitution in freshly thawed plasma. Platelet samples were reconstituted with red cells from fresh red cell concentrates and thawed fresh frozen plasma (haematocrit 40%; platelet volume 20%) as a model of platelet transfusion. Four key parameters were measured using a Quantra Hemostasis analyser and associated QPlus cartridges (HemoSonics LLC, a Stago Group company): CT (clotting time); CS (clot stiffness); FCS (fibrinogen contribution to clot stiffness); PCS (platelet contribution to clot stiffness). Standard platelet quality markers were assessed by haematology analyser, pH meter and flow cytometry.

Results: CT was faster in cold platelets (day 21: 162 ± 7 seconds), compared to RT platelets (day 21: 176 ± 9 s; $p = 0.0123$). Frozen platelets displayed the fastest CT (122 ± 7 s). The CS was higher in RT platelets,

compared to cold platelets at all time-points (Table 1). Interestingly, the CS of frozen platelets was better than RT platelets at day 21 and cold platelets from day 14. The platelet (PCS) and fibrinogen (FCS) contributions to clot stiffness followed a similar trend as the CS (Table 1 and data not shown). Platelet number and phenotype (CD61, CD62P, PAC-1, bound fibrinogen) changed as a result of temperature and storage duration, and these parameters were more strongly related to the clot stiffness parameters (CS and PCS), rather than the time to clot formation (CT). Specifically, the platelet concentration was strongly correlated to the PCS for RT platelets ($r = 0.905$). However, this relationship was reduced in cold ($r = 0.551$) and frozen platelets ($r = 0.487$). The abundance of CD61 (MFI) strongly correlated to PCS ($r = 0.820$) for cold platelets, while CD62P (%) was the most strongly correlated parameter with PCS in frozen platelets ($r = 0.650$).

Summary / Conclusions: The Quantra analyser is suitable for the haemostatic assessment of platelets stored under different conditions. In addition, it provides a novel opportunity to interrogate the relationships between the storage-induced platelet changes and their effect on haemostatic function.

P216 | How to achieve a “God speed” platelet electronic matching for iPTR patients—a single-center experience

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Background: Transfusion of HLA-compatible platelets is an effective means to resolve the immune platelet transfusion refractoriness (iPTR). Compared with the “wet” serological cross-matching technique, the “dry” HLA electronic cross-matching has many advantages such as no need to collect donor and patient samples every time, a large number of donors database can be matched, a higher degree of compatibility between donors and recipients, and so on. But the most serious problem faced by virtual matching is the probable delays in transfusion for emergency patients due to uncertainty of recruiting compatible donors.

Aims: To shorten the waiting time of iPTR patients with virtual matching by implement several measures and adjusting the matching process.

Methods: The following strategies have been implemented at Blood Center of Zhejiang Province, China, to reduce the waiting time of virtual matching: Increase the donor number of HLA database, especially active donors who donate apheresis platelets more than 3 times per year. For the emergency patients, Luminex-SSO HLA medium-resolution genotyping was used to replace PCR-SBT or NGS high-resolution genotyping. The primary source of virtual matching platelets has changed from donor recruitment to physical platelets in inventory. The scope of physical platelets for virtual matching has been expanded from the testing bank to the collecting, the testing and the qualified banks. When compatible platelets are not found in the physical blood products, a “fishnet” electronic-interception is carried out in the collection process. All work are managed by information system, including automatic searching in donor database and physical products, automatic intercepting of target donors, automatic

P215 Table 1.

CS (hPa)	RT	Cold	Frozen
Day 7	15.0 ± 1.6	11.0 ± 1.8*	-
Day 14	12.4 ± 2.2	6.7 ± 2.1*	-
Day 21	6.8 ± 3.1	3.4 ± 1.4*	-
Thawed	-	-	8.3 ± 2.1*#^
PCS (hPa)	RT	Cold	Frozen
Day 7	13.8 ± 1.4	10.1 ± 1.7*	-
Day 14	11.2 ± 2.0	5.8 ± 2.4*	-
Day 21	6.0 ± 2.9	3.0 ± 1.2*	-
Thawed	-	-	7.2 ± 2.0*#^

* $p < 0.05$ compared to RT; # $p < 0.05$ compared to D14 cold; ^ $p < 0.05$ compared D21 cold.

locking of compatible platelets, automatic generating and issuing of matching electronic-reports.

Results: By the end of 2023, the HLA database of our blood center reached 25,007 donors, of which 2588 donors donated 21,857 bags of apheresis platelets in 2023 (57.4% of all platelets). Among them, 1925 active donors donated 20,940 bags (55.0% of all platelets). The experiment time of HLA genotyping and antibody identification for emergency patients was reduced from 6 days to 2 days. From Sep 2020 to Feb 2024, a total of 1463 virtual matches were completed. The waiting days of patients at three different periods (recruitment mainly, testing bank mainly, and expansion of inventory scope) was 5.4 ± 5.4 d (median 4d), 3.8 ± 3.1 d (median 3d), and 1.2 ± 2.5 d (median 0d), respectively. After using all optimization measures from Apr 2023, a total of 702 virtual matches have been completed. Of these, 417 matching platelets (59.4%) were from the same day's inventory, 104 (14.8%) were from platelets intercepted the next day, and 181 (25.8%) were from platelets recruited and donated on the appointment date.

Summary / Conclusions: A number of innovative measures in the whole process of matching platelets have greatly reduced the waiting time of patients, and most of the virtual matching platelets are issued to hospitals on the same day or the next day at a "god speed", meeting the urgent transfusion of clinical iPTR patients.

P217 | Abstract withdrawn

P218 | Reduction of platelet count and their activation potential during cold storage of whole blood

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Background: A beneficial role for the early haemostatic resuscitation of patients with massive bleeding is well established. Balanced transfusion of platelets and plasma containing clotting factors along with red blood cells is the requirement. Providing preparedness with these short-lived blood components for the emergency medical services, especially pre-hospital, is logistically very challenging. Another drawback is the significant volume of additive solutions that are used for preservation of cellular concentrates after plasma separation. Therefore, there is a need for a single bag containing all the components and minimum of additive solutions. Leukocyte-depleted whole blood has been suggested to preserve enough haemostatic activity for up to 3 weeks of cold storage, providing easy access to a balanced haemostatic product.

Aims: To evaluate the kinetics of platelet counts and their activation potential during 3 weeks of cold storage of leukocyte-depleted whole blood.

Methods: Whole blood from ten blood group O donors was included in the study after written consent. The blood was filtered through a

platelet-sparing filter and stored at $+2-6^{\circ}\text{C}$ for 22 days. Samples were drawn after filtration (day 0), and on days 2, 8, 15, 22. Flow cytometry was used to assess activated integrin $\alpha\text{IIb}\beta 3$ and p-selectin expression on platelets before and after activation with two thrombin-receptor activating peptides (SFLRN and AYPGKF). Platelet activation potential was calculated as the ratio of mean fluorescence (MFR) measured after stimulation and on resting platelets for each marker. Phosphatidylserine (PS) expression was detected by annexin V staining. Platelets from four concentrates pooled from five buffy coat each, stored at room temperature for 8 days were included as control.

Results: A significant reduction of platelet count from $153 \times 10^9/\text{L}$ (95% CI 96.6-242.7) at day 0 to $49 \times 10^9/\text{L}$ (95% CI 37.8-64.3) at day 8 was observed. The platelet count was stable the next two weeks. PS expression was undetectable on the surface of resting platelets at day 0, but increased under the storage and reached 12% and 33% on day 8 and 22, respectively. In parallel, an increased number of CD41 positive fragments was observed. Activated integrin $\alpha\text{IIb}\beta 3$ MFR decreased from 15 to 2 between day 0 and day 8, and p-selectin MFR from 129 to 4. A larger remaining activation potential was seen in platelets originating from the buffy coat. At day 8 MFR was 3 and 20 for integrin $\alpha\text{IIb}\beta 3$ and p-selectin, respectively, and only 3% of the platelets expressed PS.

Summary / Conclusions: In cold stored whole blood, the most prominent changes were found between day 0 and day 8 with significantly reduced platelet counts and activation capacity of the remaining platelets, and a concomitant increase in percentage of PS positive platelets and CD41 positive fragments. How these changes affect the haemostatic activity of the cold stored whole blood remains to be tested.

P219 | Preserved agonist-induced activation response during 7-day storage of platelets formulated in PAS-E

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Background: Donation, processing, and transport steps are known to affect platelet activation during storage, a known factor of platelet storage lesion. Upon activation, platelets lose their discoid morphology and expression levels of various proteins are altered. The changes finally give rise to conformational changes in the GPIIb/IIIa complex, expose more binding sites for adhesive proteins and result in platelet aggregates. Retained ability to respond to agonist-induced activation aims to predict preserved *in vivo* function. While activation can have varying causes, thrombin is known to be a key regulator of platelet activation in response to vascular injury and functions via two receptors, PAR1 and PAR4. TRAP-6 is a peptide fragment of PAR1 that also acts as a PAR1 agonist, with the benefit of being slightly less potent than thrombin, allowing for a higher resolution assay better able to discern differences during storage.

Aims: Evaluate the effect of storage time on activation ability upon TRAP-6 agonist engagement of platelets formulated in PAS-E and correlate this data to other platelet characteristics during storage as part of a shelf-life study.

P219 Table 1

Mean \pm stdev	Day 1	Day 6	Day 8
Activated, pct	10.64 \pm 3.13	23.96 \pm 3.92	30.57 \pm 4.86
Agonist activated, pct	92.96 \pm 2.79	91.33 \pm 4.56	89.33 \pm 5.16
Activation level, MFI	23206 \pm 1827	33293 \pm 4624	34506 \pm 3377
Agonist activation level, MFI	129 731 \pm 16 091	131 623 \pm 17 228	121 179 \pm 17 678
Platelet concentration ($\times 10^9$ /l)	1015 \pm 122	990 \pm 136	1019 \pm 182
PDW (fL)	9.99 \pm 0.84	9.25 \pm 0.77	9.09 \pm 0.68
MPV (fL)	9.76 \pm 0.44	9.43 \pm 0.43	9.38 \pm 0.37
glucose (mmol/l)	8.94 \pm 0.46	5.93 \pm 0.68	4.36 \pm 0.82

Methods: The study used 22 buffy coat platelets (BC-PC). BC-PCs were prepared from pools of 4 BCs, excess from routine production, using a combination of centrifugation (Hettich GmbH) and automated separation (Fresenius Kabi, G5), then formulated in PAS-E (T-PAS+, Terumo) and stored for up to 7 days at +20–24°C with constant gentle agitation. BC-PCs were rested for 1 hr post-preparation prior to initial sampling, then further sampled on days 6 and 8 to reflect a full day 5 or 7 shelf-life, respectively. Activation, defined as CD41+CD62P+ platelets, was assayed by flow cytometry (DxFlex, Beckman-Coulter) both directly from storage and following treatment with TRAP-6 (12 μ M, 20 min, RT). This measurement was supplemented with various other platelet characteristics and quality markers, such as PDW, MPV, glucose/lactate, pH, pO₂+CO₂, and compared to other existing in-house data.

Results: Table 1 summarises the characteristics of BC-PCs throughout 7-day storage. Notably the activation capability upon agonist treatment is retained, though activation levels vary. Platelet concentration remains equal, with minimal changes to platelet size.

Summary / Conclusions: Resting level platelet activation level increases significantly during storage. However, platelets retain their agonist-mediated activation ability throughout seven-day storage, though the level of activation in the same cell subset slightly decreases. This may correlate with an increased presence of minor aggregates seen in the platelet units, which might require further clinical evaluation.

have a period of non-agitation between the end of the collection and reception for processing at the production service.

Aims: To evaluate the effect on APC quality with a period without agitation of 16 h after collection in the French blood establishment (EFS) Grand Est (GEST).

Methods: Two APC were collected on the same apheresis machine, they were pooled and split in order to obtain two identical APC. One bag was stored with agitation (agitated APC) after collection while the other one was stored without agitation (non-agitated APC). 16 h after collection, both APC were treated for pathogen inactivation (IA) and then stored under slow and continuous agitation between +20°C/+24°C until 7 days (D) of storage. Visual aspect, platelet count, glucose, swirling index were measured from collection (D0) to day 7 (D7).

Results: 18 APC were collected on 3 different apheresis systems (Terumo, Fresenius and Haemonics) ($n = 9$): 8 APC in Nancy, 6 in Strasbourg and 4 in Reims. They were pooled two by two and split. The evolution over time of the different biological parameters tested was identical between the non-stirred APC and the stirred APC (Table 1).

Summary / Conclusions: No agitation during the first 16 h between apheresis platelet collection and the start of the production process does not affect APC storage quality. The characteristics of APC collected in EFS GEST with or without agitation are consistent and equivalent. APC can thus be stored and transported from collection sites to the two production sites without carrying out continuous agitation within the first 16 hours post-collection.

P220 | No agitation during 16 hours after apheresis platelet concentrates collection does not affect their quality

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Background: Slow and continuous agitation of platelet concentrates (PC) enhances gas exchanges and maintains platelets in suspension in the bag. However, due to the production system organization and transport logistic, it may be possible to have agitation interruptions during PC production and conservation stages. Apheresis PC (APC) collected from remote sites that do not have agitators or supplies outside the opening period of the production platform. They can thus

P220 Table 1. Biological parameters on collected APC (Mean \pm SD) (N = 9).

	Agitated (A) / Non-agitated (nA)	D1	D5	D7
pH	A	7.20 \pm 0.05	6.82 \pm 0.14	6.80 \pm 0.14
	nA	7.14 \pm 0.09	6.83 \pm 0.11	6.80 \pm 0.11
Glucose (mM)	A	6.5 \pm 1.5	2.7 \pm 2.4	0.4 \pm 1.2
	nA	6.2 \pm 1.6	2.8 \pm 2.2	0.4 \pm 1.2
Platelet count (G/L)	A	1505 \pm 108	1251 \pm 97	1225 \pm 173
	nA	1516 \pm 108	1214 \pm 97	1266 \pm 173
Swirling index	A	+++	+++	+++
	nA	+++	+++	+++

P221 | Practical and in-process testing of a new sterile connecting device with different PVC tubings

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Background: In the preparation process of pooled platelets, tubing of buffy coats are connected to the tubing of a pooling system with a sterile connecting device. These devices make 'one-to-one' connections, however for the preparation of one pool, five to six connections have to be made. The Maconnect (Macopharma) enables 6 connections at a time and has the opportunity to decrease the number of single-use wafers, process time and also workload by prevention of repetitive strain injury (RSI).

Aims: To investigate quality of sterile docks made with Maconnect in currently used tubing, and more specifically in non-DEHP PVC tubing.

Methods: The configuration 'dry-dry' was tested with (a) tubing of the currently used whole blood collection system (CQC2988, Fresenius, DEHP-PVC) and (b) tubing of a non-DEHP-PVC system connected to the pigtail of the pooling system we use (EQ06AOE, Fresenius vs. GT526NL, Fresenius, both PVC-TOTM). Connections were judged for ease of opening using a scoring system, and for integrity by pressurizing the tubing with 3 bar, while submerged in a water bath. The configuration 'dry-wet', as applied in the pooling process, was (c) simulated by connecting the pigtail of GT526NL to waterfilled bags and tubing (representing forces of 6-40 N) and (d) tested in-process by the Production Department. Integrity of connections was assessed by a leakage check under gravity of the filled bags. Integrity of the in-process prepared platelet pools was also checked with routine bacterial screening (BacT/Alert). Simulation tests (a, b, c) were performed in 10-fold, resulting in 60 docks each, and 100 pools were prepared during the in-process test, resulting in 600 docks.

Results: To operate the Maconnect was considered as easy and user-friendly, however, to reset the alignment of the clamps they have to be closed again. Almost all (99%) connections were judged as easy to open within one second, and were smooth at the inside. No failure of integrity was found in any tubing that was pressurized or weighted. Also, and more important, bacterial tests of platelet pools were all negative.

Summary / Conclusions: Sterile connections in PVC tubing, DEHP or non-DEHP plasticized, made with the Maconnect were easy to open, smooth and tight. Closing the clamps for re-alignment may be considered as a disadvantage, but still means a reduction in clamp-closing of 67% in the BC pooling process (RSI prevention).

P222 | Comparison of different methods for platelet count

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Background: During the validation of the Blood Bank (BB) mode of the hematology analyzer XN-1000 (Sysmex) for blood components quality control (QC), a significant 17% average increase in platelet count was observed when comparing impedance-based (PLT-I) to flow cytometry-based (PLT-F) measurements. Platelet count is a critical parameter for platelet products QC, which can directly impact in platelet products manufacturing. For this reason we had the need to verify the results obtained with the PLT-F channel, to ensure accurate platelet concentrate dosing. Taking into account that for cell count, flow cytometry is a gold standard technique, in this work we have compared PLT-I and PLT-F counts with two different flow cytometry techniques, one using fluorescent microbeads used to determine absolute counts, and the other by counting absolute cell numbers directly (per unit sample volume).

Aims: Compare 4 different platelet count methods to ensure the platelet dose of our platelets concentrates for transfusion.

Methods: The comparison involved two platelet product types: platelet pools from 4 or 5 units of whole blood buffy coats ($n = 31$). Platelet counts were evaluated using four techniques: Impedance with Sysmex XN-1000 CBC mode (PLT-I), Flow cytometry with Sysmex XN-1000 PLT-F channel utilizing a specific fluorescent RNA dye (PLT-F), Flow cytometry with CD41a mAb-stained samples and counting with FlowCount microbeads on a Beckman Coulter Navios Ex flow cytometer (FC-Microbeads), and Flow cytometry with CD41a mAb-stained samples and absolute counting using Miltenyi MACSQuant 10 (FC-AC). Platelets were identified in flow cytometry by CD41a expression.

Results: The study confirmed an 18.4% average increase in platelet count between PLT-I and PLT-F ($p = 0.0137$). Additionally, comparing PLT-F with flow cytometry using microbeads showed a 5.3% average difference with an $r^2 = 0.8887$ ($p = 0.4684$). Furthermore, platelet count using FC-AC was only 2.7% below PLT-F with and a $r^2 = 0.8995$ ($p = 0.6272$).

Summary / Conclusions: The results demonstrate that impedance underestimates platelet count in platelet products, with PLT-I consistently lower than the other three methods. Moreover, PLT-F yielded similar results to two different flow cytometry techniques specifically labelling platelets using the CD41a marker. In conclusion, PLT-F emerges as a useful and accurate automated analysis for ensuring the platelet count in transfusable platelet concentrates.

P223 | Performance validation of an automated blood processing system in blood service of Singapore

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Background: The Blood Services Group (BSG) in Singapore secures the national blood supply, ensuring all patients in Singapore have access to adequate and safe blood. BSG currently processes whole blood (WB) collected in Top and Bottom Blood Collection Bag into blood components with semi-automated buffy coat method using automated extractors and convention centrifuges. The pooled platelet is processed using TASCI PL System (Terumo Automated Centrifuge & Separator Integration System for platelet). BSG is looking for a fully automate blood component processing to further improve productivity and efficiency. Therefore, a performance validation of TERUMOBCT REVEOS Automated Blood Processing System (Reveos System) is conducted to access its performance and the suitability for use in BSG.

Aims: The study aims to evaluate the quality of blood components derived from WB collected in Reveos blood collection sets and processed using REVEOS System and study the throughput and efficiency of Reveos System in comparison to the current system in use by BSG.

Methods: 62 units of WB collected using Reveos NLR set (non-leucocyte reduced) and Reveos LR EXT set (leucocyte reduced) were processed using Reveos System. Pooled platelet units were prepared using Platelet Pooling set. Standard quality control (QC) parameters of blood components processed by Reveos System were monitored based on AABB and CE standards. The processing steps and time of Reveos System and the current processing system were recorded and measured. The throughput and turnaround times (TAT) of both systems were calculated for comparison based on an average weekday and weekend processing loads with the consideration of manpower and number of equipment required.

Results: Red cell concentrate (RCC), leucocyte reduced RCC, plasma and pooled platelet processed from NRL set, LR EXT set and platelet pooling set were all within QC parameters criteria based on AABB and CE standards except that 2 of 29 units of leucocyte reduced RCC failed to meet the CE standard for residual white blood cell counts. The leucocyte reduced RCC yielded an overall passing rate of 93%, slightly below the requirement of 95%.

It was estimated to take 6 hrs 43 mins to process 300 units of WB (weekday average workload) and 13 h 26 mins to process 600 units (weekend average workload) with 2 operators and 8 units of Reveos System. The TAT was 7% longer than the current processing system but there was a 50% savings in manpower and 58% fewer equipment needed. Pooled platelet processing comparing to the current method, there was a 10% time savings in TAT for an average production of 50 units, but 9% more time was required in processing a max production of 100 units. Similarly, there were a 67% savings in manpower and 75% fewer equipment required.

Summary / Conclusions: Blood components, processed using Reveos System, were all within the QC requirements in BSG as AABB Standard is used for acceptance. QC failure for leucocyte reduced RCC may be due to the small sample size. Adjustment of separation setting in Reveos System for optimisation will be required for adoption. While the TAT of both systems was insignificant, there were significant savings in manpower and equipment with the Reveos System compared to the current processing systems. This reduction in equipment required could translate to time and cost saving in maintaining all equipment for the processing. Cost benefit analysis shall be studied.

P224 | Blood donors with a result of positive sickle cell test at a blood bank in Medellín, Colombia

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Background: Sickle cell disease is a group of disorders where hemoglobin is affected, driving red blood cells (RBCs) to adopt a "sickle" shape due to the formation of Hemoglobin S (Hb S), produced by a mutation in an amino acid in the globin chain gene. Sickle cell trait (SCT) is a condition in which the patient has only one copy of this gene affected, leading to a benign condition of this pathology. In Colombia, up to 10% of SCT has been reported in areas where the black race predominates, such as in the Chocó, Antioquia, Valle del Cauca and Nariño. Some countries do not accept donors who suffer from sickle cell anemia but accept people who have SCT; the transfusion of RBC from these donors is carried out without problems in most cases, it is only restricted in intrauterine or neonatal transfusion, due to the risks that these patients could suffer. The use of filtered components has shown some benefits during transfusion; therefore, it has become important to assess the parameters that can negatively affect the filtration process. The deficiencies in filtration have been attributed to the in vitro polymerization of Hb S, partially or completely obstructing the filter, causing poor recovery of RBCs and non-elimination of leukocytes (LC), given by the reduction of the LC binding sites in the filter, creating channels that allow LCs to pass easily.

Aims: Determine the number of donors who presented SCT detected due to complications when filtering blood components.

Methods: A descriptive observational study was performed. The information used was taken from donations that presented complications

during filtration, this included filtration before storage (Pre-S) of whole blood and after storage (Pos-S) of RBCs, for which a sickle cell test was performed. The description of the characteristics of the population was performed with absolute (n) and relative frequencies (%) and measures of central tendency.

Results: A total of 17 donors were gathered in whom a problem occurred during filtration (7 Post-S; 10 Pre-S) between the years 2021 and 2024. Of these donors, 58.8% were men and 42.2% were women. The average age and hemoglobin content were 38 years ± 11.4 and 14.9 g/dL ± 1.4 respectively. Ten donors had a positive sickle cell test, and filtration complications occurred in 58.8% in Pre-S.

Summary / Conclusions: This study generates the necessity for an active search of donors who may have SCT, since some tools can be implemented to avoid the filtration of units in this type of population because the transfusion of units with SCT can lead to the formation of irregular antibodies, or may have phenotypes necessary for patients with atypical antibodies; It is also important to characterize them and inform them of their status, because under normal conditions, people who have SCT do not polymerize Hb S, with the exception of minor sickle cell anemia, observed in the hyperosmotic and low pH environment of the renal medulla. In this study, a proportion of donors with a negative sickle cell test of 42.2% was found. In these donors, filtration difficulties could have been due to filter quality defects, the presence of fibrin or micro clots. Finally, the filtration process of blood components must be optimized; since the transfusion of this type of components can prevent events such as non-hemolytic febrile transfusion reactions, cytomegalovirus infection and alloimmunization to human leukocyte antigens (HLA).

P225 | Obtaining granulocytes for transfusion from buffy-coat pool residues

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Background: Granulocyte (GC) transfusions can be used as supportive therapy in patients with life-threatening neutropenia caused by bone marrow failure or in patients with neutrophil dysfunction. There are two methods for collecting GC for transfusion: a pooled, buffy-coat (BC) derived component, and by apheresis of GC, usually collected after stimulation of donors with steroids and granulocyte colony stimulating factor (G-CSF). In some countries, this practice is not considered ethical for unrelated/non-directed donation. There is limited published literature on the *in vivo* efficacy of whole blood (WB) derived GC concentrates. However, this practice is included in Guide to the Preparation, Use and Quality Assurance of Blood Components, 21st ed, published by EDQM in 2023, and in Clinical Guidelines for the Use of

Granulocyte Transfusions, prepared by the Granulocyte Transfusion Working Group, published by the NHS in 2006 and revised in 2021.

Aims: Our Blood Centre is required by a hospital of our area to process this product containing GC, to be transfused to a 18-year old female patient weighing 40 kg, diagnosed with aplastic anemia and life-threatening invasive pulmonary aspergillosis related to persistent agranulocytosis, who requires allogenic bone marrow transplant as only way to survive, what is nowadays delayed. To date, the patient receives 2 GC transfusions per week, obtained by apheresis from her father, after stimulation. She receives each time 0.5×10^9 granulocytes/kg (weekly dose of 40×10^9 granulocytes). However, GC are obtained increasingly less, related to the donor's loss of response to stimulation. Sometimes, the patient has been transfused with 8 buffy-coats from our Centre. The objective is preparation of GC concentrates through the 8-BC pool residues, obtained in production of inactivated platelet pool BC derived. After the separation of the platelet concentrates, the obtained residues are known to have a high count of GC, and can be transfused to the patient, after demonstrating their complying with the current Guides.

Methods: The Blood Centre produces bags containing a residue of a 8-BC pool after having removed the platelet concentrate, that will be after inactivated. Each BC is obtained from a single WB donation of 450 mL, having a volume of 42ml. The product should contain $>5 \times 10^9$ GC/unit, must be irradiated before transfusion due to high content of lymphocytes, and must be ABO-compatible for transfusion, that should be done before midnight the day following donation.

Results: 16 pool residues are obtained from 8-BC pools on day +1, obtaining the following average results: Volume 231 ml. Hemoglobin/unit 39.8 g (Average hemoglobin content in RBC concentrates, 50.80 g). Hematocrit 52.6%. Leukocyte count /unit 16.5×10^9 . Granulocyte count/unit 6.9×10^9 . Platelet count/unit 1.37×10^{11} (Average platelet count in platelet concentrates, 3.18×10^{11}).

Summary / Conclusions: - GC concentrates meet the quality standards (average GC count $>5 \times 10^9$ /unit). Transfusion of a GC concentrate also represents a contribution of RBC and platelets. Each unit represents 78.35% of a RBC concentrate, and 43.08% of a platelet concentrate. So, RBC and platelet transfusion requirements may be reduced during treatment. Patient will need 5.79 residues per week to achieve the same dose she is currently receiving. Periodic checks to the patient are recommended. A procedure must be established to meet the needs/risks of potential patients who may require the administration of this component.

P226 | The evolving landscape of blood collection centers—challenges in processing of whole blood units after donor pool diversification

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Background: The recent migration movements in European countries have been posing several challenges towards the transfusion of patients from a different ethnic background. It is thus desired that

P226 Table 1 Results from all units of packed red blood cells that could not be leukoreduced.

Year	n of prolonged/ unsuccessful leukoreduction (% of all collected whole blood units)
2010	3 (0.013%)
2011	13 (0.056%)
2012	22 (0.091%)
2013	11 (0.048%)
2014	9 (0.043%)
2015	2 (0.011%)
2016	2 (0.011%)
2017	5 (0.028%)
2018	6 (0.034%)
2019	5 (0.028%)
2020	0 (0.000%)
2021	1 (0.006%)
2022	12 (0.068%)
2023	12 (0.069%)
n of units (total) from prolonged/unsuccessful leukoreduction	103
n of donors (total) of prolonged/unsuccessful leukoreduction	69

P226 Table 2 - Characterization of the donors

Sex (%)	
Male	Female
47%	53%
Ancestry/Ethnic background (%)	
Portuguese/Caucasian	71.0%
Brazilian	18.8%
African	8.7%
Venezuelan	1.4%
Identification of Hb variants	
HbS heterozygosity	22.0%
Other Hb variants (not specified)	1.4%

blood banks diverse their donor pool. On the other hand, it is known that some hemoglobinopathies are widely prevalent in some regions of world.

Aims: Our aim was to analyze the incidence of hemoglobin variants in whole blood donations whose packed red cell units (pRBC) could not be leukoreduced.

Methods: We performed a retrospective analysis from 2010 to 2023 of all the pRBC that could not be leukoreduced.

Results: From 2010 to 2023 we observed that we could not leukoreduce 103 pRBC corresponding to 69 whole blood donors (Tables 1 and 2).

In the face of a prolonged or impossible leukoreduction, the donor would be called up to the blood bank to review medical records and to collect a sample for hemoglobin (Hb) variants screening. Initially it was done through Hb solubility test and Hb electrophoresis, later through high performance liquid chromatography. Donors who returned to our blood bank and were confirmed to be carriers of HbS or other variant were informed of their status and referred to a specialized Hematologist. In those who had adequate venous accesses, they were offered the possibility of apheresis platelet donation.

Summary / Conclusions: The presence of HbS precludes adequate leukoreduction of pRBC. Even though a careful donor history is performed before donation, sometimes carriers of hemoglobinopathies are not aware of their status. To convey international recommendations, units that present HbS should not be used for transfusion, since some RBC may undergo sickling. The trend towards increasing the diagnosis of hemoglobinopathies is in line with recent mass migration movements from high prevalence countries. The numbers decreased during the COVID-19 pandemic. Study of Hb variants is always helpful both for donors' global health and to maintain quality parameters in blood collection centers.

P227 | Impact of storage conditions in pediatric platelet concentrates

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Background: Production of paediatric platelet concentrates (PPC) consists on transferring a wished-for volume of platelet solution from an apheresis platelet concentrate (APC) to a transfer bag. This transfer may either be realized in advance—and the PPC is then stored in similar conditions as the mother-APC (i.e. in polyolefin storage bags suitable for gas exchanges); or may be realized extemporaneously, just before transfusion. In this latter situation, paediatric PVC transfer bags may be used. Etablissement Français du Sang Grand Est practices both processings

Aims: Over a period of 7 days, study the impact of storage bag type on standard quality parameters in PPC produced from pathogen-reduced APC.

Methods: One APC (control condition A) was pathogen-reduced with amotosalen/UVA at Day 1 (D1) after collection with amotosalen/UVA, then used at D2 for the preparation of two PPC (~70mL each). The first PPC was stored in a 1L polyolefin bag(condition B) and the second PPC in a 150mL PVC bag (condition C). All three PPC were stored during 7 days under agitation, between +20°C and +24°C. The following biological parameters were analyzed at D2, D4 and D7: swirling, platelet count, mean platelet volume, blood gas, pH, glucose, soluble p-selectin, lactate, LDH.

Results: Control APC and PPC B respect regulatory requirements (volume, pH, platelet count) while 1/12 of PPC C units had a pH lower than 6,4 since D4. Almost all biological parameters demonstrate a strong decrease in PC quality after two days of storage (D4) when

platelets are stored in PVC bags. Platelet clusters were observed in all and only PVC bags from D4, correlating with a decrease in platelet swirling. PPC storage in polyolefin results in a slight decrease of PC quality, possibly due to the bigger volume content of the bag (1L for 70mL of PC) compared to the mother-APC. LDH and p-selectin show similar concentrations in APC A and PPC C, while PPC B show a slight LDH and p-selectin increase.

Summary / Conclusions: PPC prepared in PVC bags must be delivered without delay and platelets should not be stored in PVC bags over a few hours. If PPC are prepared in advance, they should be prepared in polyolefin bags that permit gas exchange and are suitable for PPC storage during 7 days. Blood establishments may adapt their PPC production to organization and economic considerations, as polyolefin bags are 2.5 times more expensive than PVC ones.

P228 | Erythrocyte microvesicles stored under blood bank conditions—imaging and analysis by flow cytometry and atomic force microscopy

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Background: The membrane vesicles are a heterogeneous group of different sizes, origin, biological and physical properties, mechanism and source of formation. During blood banking, erythrocyte vesicles formation and membrane remodeling was observed. Phosphatidylserine is exposed in the outer bilayer of the cell membrane, and is important during red blood cell aging and microvesicle release. Despite many studies, the exact and complete mechanism and components of the vesicle involved process formation remain only partially established. Observations have shown a link between elevated number of vesicles released from blood cells during storage of blood concentrates and post-transfusion complications.

Aims: Quantitative and qualitative analysis of erythrocyte microvesicles isolated from blood units (5th, 20th, 40th day of storage) with flow cytometry and atomic force microscopy.

Methods: Microvesicles released from human erythrocytes were isolated and characterized. These structures were tested by flow cytometry and atomic force microscopy. Cytometric analysis was performed using the annexin V-FITC, anti-CD235a-PE and anti-CD47-PE antibodies. Mix of fluorescent calibrated beads of varied diameters allowed us to determine the size of microvesicles in flow cytometer. The counting beads were used to measure the absolute number of microvesicles per 1 μ L. The topographical images of erythrocyte microvesicles were recorded using AFM technique to observe morphological and structural features of these structures.

Results: Analysis of microvesicles using flow cytometry and calibration beads allowed the identification of erythrocytes microvesicles with diameter's of about 0.5 μ m, that were positive for phosphatidylserine and CD235a. High resolution AFM images revealed the

appearance of vesicles of a size of around 0.2 μ m or more. We observed a statistically significant increase in the number of erythrocyte microvesicles in the "old" (40th day of storage) samples, as compared to the erythrocyte microvesicles in the "fresh" (5th day of storage) samples. We didn't find any statistically significant differences in the expression of CD235a comparing vesicles from samples stored for 5, 20 and 40 days. Microvesicles on day 5 had elevated expression levels of CD47 molecule, as compared with MV on day 40.

Summary / Conclusions: The results of our study reveal a relationship between the storage time of erythrocyte concentrates, the amount of released microvesicles and the expression of phagocytosis regulator-CD47 molecule. Increase of the circulating microvesicles number, topography and shape changes depend also on the time of blood storage.

P229 | Seasonal variation of donors platelet count in France. Impact in platelet concentrate production

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Background: In France, donor platelet count has been measured for each blood donation since 2008. In the same period, platelet content in apheresis platelet concentrate (APC) and buffy-coat platelet concentrate (BCPC) is systematically determined before delivery and labeled on the bag.

Aims: Study the seasonal platelet count in donors in the different French region and evaluate potential impact of this in platelet content in APC and BCPC. Then, determine and describe a method who permit to obtain a constant platelet content in BCPC production.

Methods: Mean of platelet count in donors was calculated monthly in the four regions in metropolitan France (East, South, West, North) and in Reunion island during several years (2017 to 2023). Mean of platelet content in BCPC an APC monthly produced in facilities of Strasbourg, Nancy, Besançon, Marseille, Toulouse and Saint-Denis de La Réunion was determined. Production of BCPC: pooling of buffy-coats is performed randomly or in function of donors platelet count.

Results: Mean of donor platelet counts during May to September (warm period in the northern hemisphere) were lower than other months in each region (Table 1 shows 2022. Other data were not show). In Reunion island, a French department located in southern hemisphere, mean of donor platelet counts were higher during May to September (cold period) than other months. Consequences of these variations are a decrease of buffy-coat number with high platelet count $>275.10^9/L$ during warm period ($<27\%$) in comparison to cold period ($>34\%$). Simultaneously, number of Buffy coat with low platelet count $<225.10^9/L$ increase during warm period ($>34\%$) in

P229 Table 1: Mean of donor platelet counts per month ($10^9/L$) (Mean \pm SD) in 2022.

N > 40 000	French Regions	Months											
		Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec
2022	East	272	271	271	271	269	269	269	266	269	273	274	275
	South	274	272	274	274	271	268	264	264	269	274	276	276
	West	273	273	274	274	270	269	267	267	269	271	273	274
	North	274	270	269	269	263	258	256	256	260	266	266	270
	Reunion Island	237	234	234	231	237	238	241	241	237	234	235	235

P229 Table 2: Buffy coat frequency in function of platelet count (2022 data) (%)

	Low platelet count <225.10 ⁹ /L	Medium platelet count 225-275 10 ⁹ /L	High platelet count >275.10 ⁹ /L
Warm period (May to September)	>34%	35-38%	27%
Cold period (October to April)	<28%	35-38%	>34%

comparison to cold period (<28%) (Table 2). Platelet content in APC and random BCPC follow these seasonal variations: mean platelet content during warm period (APC: $4.5.10^{11}$; BCPC: $6.0.10^{11}$) was less than mean of platelet content during cold period (APC: $5.0.10^{11}$; BCPC: $6.6.10^{11}$). However, when BCPC is produced by pooling buffy coat in function of donors platelet count, mean of platelet count was constant during the year. There is no variation in platelet content.

Summary / Conclusions: Seasonal variation of platelet count in blood donors has been observed in France with higher platelet count during cold period. APC and BCPC processes has to be adapted to avoid seasonal variation of platelet content in the platelet concentrate and to permit platelet delivery with right dose.

P230 | Evaluation of leucoreduction time for red blood cells processed at Lisbon Blood and Transplantation Center—Algarve Delegation (LLA)—IPST

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Background: In Portugal, since January 1st, 1999, universal leucoreduction of cellular blood components became mandatory. At LLA-IPST, blood components are obtained from Whole Blood (WB) collected in Fresenius Kabi Compoflow® Top and Bottom bags

(T&B), with a volume between 450 ± 50 ml. The reference of this manufacturer is between 20 and 30 min for the leucoreduction time (LT).

Aims: This study was developed to verify if the LT of Red Blood Cells (RBC), processed at LLA-IPST, meets the manufacturer's specifications.

Methods: From November 2015 to December 2023, the LT of 9565 RBC with volume ≥ 230 ml, was monitored using timers. In the same period, residual leucocytes (RL) were determined in 2511 RBC using flow cytometry. Also, haematological QC was performed on 2843 RBC utilizing a haematology analyser.

Results: Regarding LT evaluation, approximately 9.5% of the WB donations made in the Algarve between 2015 and 2023 were analysed. The average volume of the analysed RBC was 263.4 ± 16.52 ml. It was found that the average LT was 21 ± 3.91 min, with a statistical mode of 20 min. Out of the analysed RBC, 3516 (36.8%) had an LT between 12 and 19 min, 5902 (61.7%) had an LT between 20 and 30 min and 147 (1.5%) had an LT between 31 and 47 min. Out of the analysed RBC, 1028 had an LT between 12 and 16 minutes (average volume was 250.2 ± 14.9 ml), 4693 had an LT between 17 and 21 minutes (average volume was 258.5 ± 13.9 ml), 3008 had an LT between 22 and 26 min (average volume was 270.9 ± 14.1 ml), 721 had an LT between 27 and 31 min (average volume was 279.1 ± 14.9 ml), 97 had an LT between 32 and 36 min (average volume was 287.2 ± 14.4 ml) and 17 had an LT between 37 and 41 min (average volume was 282.4 ± 12.3 ml). Only 1 RBC, with 276 ml, present an LT between 41 and 47 min. The 18 RBC with a LT higher than 37 min didn't correspond to any other event such as previous leucoreduction failure/blocking, presence of haemoglobin S or RL out of specifications. From the 2511 RBC (28.2% of RBC production) subjected to RL, approximately 0.08% showed values above specifications. The 2843 RBC (32% of RBC production) subjected to haematologic QC, had haematocrit within requirements and 0.18% had haemoglobin below specifications.

Summary / Conclusions: In this study, 61.7% of the RBC LT matched the manufacturer's specifications. Theoretically, the volume is one factor that can affect the performance of leucoreduction filters and all the RBC analysed had a WB collection volume between 450 ± 50 ml. However, it was found that the LT of RBC was shorter if RBC volume was lower. On the other hand, higher LT was associated with

higher RBC volume. The QC results of RBC were according to national specifications and met the state of the art.

P231 | A comparison of manual vs. automated hematocrit measurement in red blood cell concentrates in PAGGSM additive solution

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Background: Upcoming European regulation banning di(2-ethylhexyl) phthalate (DEHP) plasticizer in medical devices has led to the development of alternative blood collection and storage systems. DEHP leeches from bags and stabilizes RBC membranes, and its removal could impact red blood cell (RBC) component *in vitro* quality. To mitigate these potential impacts, additive solutions (AS) that better preserve RBC concentrates (RCC) during storage are being explored. One promising plasticizer/AS combination is DEHT (di (2-ethylhexyl) terephthalate)/PAGGSM. RBC swelling upon addition of PAGGSM, an isotonic storage solution, has been shown to differ compared to hypertonic solutions such as SAGM (Zehnder, *Vox Sang*, 2008), which could impact hematocrit (Hct) measurement.

Aims: To compare Hct of RCCs in DEHT-PAGGSM measured by an automated hematology analyzer or manually in a microhematocrit centrifuge.

Methods: Whole blood (WB) was collected into prototype 475 mL PVC-DEHT top/bottom WB collection sets (Macopharma REF PRORQT4-B) anticoagulated with CPD. RBCs were separated from WB using one of two semi-automated component production processes: A “warm” process in which WB units were kept at room temperature and RCC were separated 18–24 h post-collection; or a

“cold” process in which WB units were placed in refrigerated storage (1–6°C) within 24 h of the stop bleed time and RCC were separated 42–48 h post-collection. RCC in PAGGSM were stored refrigerated and sampled on day (D) 1 (warm process) or 2 (cold process), D28, D36 and D43. Hct was measured using an automated Sysmex XN-1000 and a microhematocrit centrifuge (Hettich). Hemolysis was calculated using automated and manual Hct values, total hemoglobin (Hb; Sysmex XN-1000) and plasma Hb (HemoCue Plasma/Low).

Results: Automated Hct values were slightly, but statistically significantly, lower than manual Hct values on D1 or D2 (Table 1). For warm process RCCs, automated values continued to underestimate Hct at D28, but on D36 and D43 there were no statistically significant differences between the methods. For cold process RCCs, the automated method overestimated Hct at D36 and D43. The lack of concordance between automated and manual Hct values at D1 or D2 did not significantly impact determination of hemolysis (warm process RCCs: 0.05 ± 0.02 % when automated or manual Hct was used; cold process RCCs: 0.02 ± 0.02 % when automated or manual Hct was used).

Summary / Conclusions: Automated methods use electronic impedance to calculate mean cell volume and hence Hct. Manual Hct measures the volume of packed RBCs in suspension which we postulate provides a more accurate assessment of osmotic changes. Although small, potential bias in Hct determination depending on the method, which we show varies during storage, should be considered by blood operators assessing the impact of alternative plasticizers/AS, particularly to ensure calculation of hemolysis is not impacted.

P232 | Determination of optimal pool of buffy-coat for preparation of adult-dose pooled-platelet concentrates based on in-vitro parameters single-center study from India

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P231 - Table 1: Comparison of Hct (L/L) in RCCs in PAGGSM measured by Sysmex XN-1000 (Auto) or using a microhematocrit centrifuge (Man). Mean (\pm SD) are shown. Paired *t*-tests were used to calculate statistical significance between Auto and Man Hct

	Warm (n = 15)	p-value	Cold (n = 16)	p-value
D1 or 2 Auto	0.62 (\pm 0.02)	<0.0001	0.60 (\pm 0.02)	<0.0001
D1 or 2 Man	0.65 (\pm 0.02)		0.63 (\pm 0.02)	
D28 Auto	0.61 (\pm 0.03)	0.0001	0.60 (\pm 0.03)	0.0860
D28 Man	0.62 (\pm 0.02)		0.60 (\pm 0.02)	
D36 Auto	0.62 (\pm 0.03)	0.8692	0.60 (\pm 0.03)	0.0073
D36 Man	0.62 (\pm 0.02)		0.59 (\pm 0.02)	
D43 Auto	0.61 (\pm 0.03)	0.7281	0.60 (\pm 0.03)	0.0012
D43 Man	0.61 (\pm 0.03)		0.58 (\pm 0.02)	

Background: In India, recent amendments in the Drugs & Cosmetics Act, 2020 has allowed, for the first-time, pooling of whole blood derived platelet concentrate. Internationally, 4-6 buffy-coats (BC) are pooled and processed into pooled-platelet concentrates (PPC). There are a couple of studies from India which have evaluated pooled-platelets. None of these studies have evaluated an optimum number of BC to be pooled (4, 5, or 6) to match the quality standards applicable to single dose platelet concentrates (SDPC) prepared from apheresis, for which BC pools (PPC) could serve as an alternative.

Aims: To establish the optimal number of BC to be pooled to prepare PPC that would conform to quality standards of SDPC including total platelet of 3×10^{11} per unit.

P232 Table 1: Groups I, II and III in comparison with SDPC (group IV) on various study parameters. (*- *p*-value is significant, <0.05)

	Group I Mean ± SD	Group II Mean ± SD	Group III Mean ± SD	Group IV Mean ± SD
Platelet Count	239 ± 38 × 10 ⁹ (<i>p</i> -value = 0.001*)	313 ± 30 × 10 ⁹ (<i>p</i> -value = 0.994)	348 ± 35 × 10 ⁹ (<i>p</i> -value = 0.032*)	309 ± 72 × 10 ⁹
pH	7.2 ± 0.1 (<i>p</i> -value = 0.997)	7.2 ± 0.1 (<i>p</i> -value = 0.984)	7.1 ± 0.1 (<i>p</i> -value = 0.842)	7.2 ± 0.1
pO ₂	173.3 ± 38.9 (<i>p</i> -value = 1.000)	162.3 ± 31.3 (<i>p</i> -value = 0.958)	191.7 ± 39 (<i>p</i> -value = 0.684)	171.4 ± 51.3
pCO ₂	37.9 ± 8.9 (<i>p</i> -value = 1.000)	36.6 ± 10.2 (<i>p</i> -value = 0.980)	39.2 ± 8.1 (<i>p</i> -value = 0.995)	38.2 ± 10.5
Lactate	8.8 ± 3.2 (<i>p</i> -value = 0.823)	9.4 ± 3.2 (<i>p</i> -value = 0.967)	10 ± 3.1 (<i>p</i> -value = 1.000)	10.1 ± 3.3
Glucose	316.3 ± 30.4 (<i>p</i> -value = 0.961)	326 ± 41.1 (<i>p</i> -value = 0.681)	321.7 ± 31.6 (<i>p</i> -value = 0.836)	308.8 ± 33.6

Methods: BC (55-60mL) were prepared from 450mL whole blood collected in the top-and-bottom quadruple blood bag system Composelect (Fresenius Kabi, Germany). PPC were manufactured by pooling 4, 5, or 6 ABO-matched BC using the Octopus pooling set Compostop (Fresenius Kabi, Germany). Plasma was added to the BC pools to achieve a final PPC volume of 300mL. PPC were subsequently leukoreduced by the in-line leukoreduction filter of the pooling set. The three groups comprising of 10 PPC each—group I (pool of 4 BC), group II (pool of 5 BC) and group III (pool of 6 BC) were compared amongst themselves and SDPC (group IV) on hematological (platelet count), physical (volume, pH, swirling), biochemical (pO₂, pCO₂, glucose and lactate) parameters and sterility. ANOVA test was applied to detect statistical significance (*p*-value < 0.05). The institutional review board approved the study.

Results: Table 1 shows the comparison of the four groups on various study parameters. The four groups had no statistically significant difference in hematological, physical and biochemical parameters. The mean volume was 300mL and all PPC units exhibited swirling. All PPCs were sterile and had a residual leukocyte count of <5 × 10⁶/unit (as per Indian guidelines requirements). Since groups II and III were equivalent or superior to SDPC (group IV) in terms of total platelet count, and because group II had fewer BC (5<6) than group III, it seemed to be the 'optimum' pool.

Summary / Conclusions: This study suggests that pools of five BC (group II) provided optimum adult dose of platelets comparable to SDPC. PPC manufactured from 5 BC can be used as an alternative to SDPC in clinical practice.

P233 | Development and validation of transport boxes for red blood cell concentrates by unmanned automated drone

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Background: A red blood cell concentrates (RBC) transport was executed for the first time in July 2023 with the Helicus company to proceed a fully automated flight without pilot. Dedicated transport boxes were validated 2 RBC transport boxes specifically developed for controlled temperature shipments by drone and allowing fully

P233 Table 1: Temperatures registered during transport for one bag at 30°C

RBC Temperature (°C)	Test 1	Test 2	Test 3	Mean
Directly out of the fridge	4.3	4	4	4.1
Transport departure	5.7	5.7	5.1	5.5
After 4h of transport	5.4	5.4	5.4	5.4
Time to reach 10°C	*	*	*	*
Time to reach 6°C	*	*	*	*
Time to reach 2°C	*	*	*	*

P233 Table 2 Temperatures registered during transport for 5 bags at 30°C

RBC Temperature (°C)	Test 1	Test 2	Test 3	Mean
Directly out of the fridge	4.3	4.3	4	4.2
Transport departure	5.4	5.1	4.5	5
After 4h of transport	5.4	4.8	5.4	5.2
Time to reach 10°C	*	*	*	*
Time to reach 6°C	*	*	*	*
Time to reach 2°C	*	*	*	*

automated operations and compatibility with future Helicus Drone Cargo Ports.

Aims: Develop and validate boxes for drone transport of RBC's as fast as possible on an intervention area or urgently to hospitals and ensure transport temperatures compliant with European Guidelines.

Methods: Triplicates tests were performed by simulating an ambient temperature of +30°C in worst case payload conditions: minimum box content (one RBC bag) and maximum maximum payload (5 RBC bags). Two different box designs were tested. RBC's stored at 4°C ± 2°C before the tests were taken out of the cold room and maintained 10 min at ambient temperature (23°C) before packaging in the transport box to simulate routine operations. Transport boxes were placed in a thermostatic chamber for 4 hours. Acceptance criteria were set according to EDQM guideline, RBC shipped between + 2°C and + 6°C. Temperature of components should never be below + 1°C nor exceed + 10°C during a maximum transit time of 24 h.

Results: For both minimum payload and maximum payload, respectively 1 and 5 RBC, shipped at ambient temperature of +30°C of the boxes maintained shipment temperature between 2 and 6°C.

Summary / Conclusions: The transport boxes tested for drone transport of RBC's complied with EDQM requirements and maintained the content between 2 and 6°C during 4h. Additional experiments are required to simulate additional ambient temperatures conditions and widen the operating range of the drone in all realistic flight conditions.

P234 | Field study of the implementation of a blood processing device in Midlands KZN

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Background: The South African National Blood Service's Processing Center in Kwa-Zulu Natal (KZN) currently processes an average of 11334 units of Whole Blood (WB) per month and supplies a product mix across a large geographical footprint to all blood banks in KZN. The current processing center experiences bottlenecks in production and an increased Turn Around Time (TAT) for processing blood which impacts availability and service delivery to patients in distant hospitals. There is a need to investigate the possibility of decentralising the processing center and creating an inventory hub in Midlands KZN to improve service delivery to the surrounding areas.

Aims: To validate the automated Reveos processing instrument for processing WB into Red cells (RCC), Plasma (FFP) and an Interim Platelet unit (IPU) to manufacture pooled platelets (PP). Furthermore, to test the system in the field.

Methods: A location was identified that was strategically located with minimal setup costs. Stakeholder engagement to share information, Standard Operating Procedures were written and a new product ID for Reveos Products was created. For the validation, eight WB donations were processed into RCC, FFP and IPU. The IPU was stored on a platelet agitator and 5 IPU's were pooled with Platelet Additive Solution (PAS) to make a PP. The volume and factor VIII were measured on the FFP; the white cell count (WCC) and haematocrit (HCT) on the RCC; the platelet count on the IPU and the platelet count and sterility on the PP. Once validated the instrument was used to routinely process donations between 15/08/23 and 22/09/23. According to the Standards in South Africa, ≥ 80% of the products processed must meet the acceptance criteria for RCC, FFP and PP.

Results: All 8 WB used for validation passed the quality control indicators except one RCC that was not tested and one RCC failed due to the donor having a high WCC. The FFP had an average volume and FVIII count of 262.5ml and 0.85 IU/ml respectively; RCC had an average WCC and HCT of 1.42×10^9 /unit and 0.6 l/l respectively. The IPU's had an average platelet yield index (PYI) of 98×10^9 /unit resulting in an average platelet count in the PP of 1174×10^3 cells/μl.

Summary / Conclusions: The Reveos Field Study in Midlands Pietermaritzburg successfully demonstrated the system's transformative

impact on WB processing. In the field 4343 whole blood donations were processed on the automated Reveos platform. 96.08% was correctly processed. However, 170 WB failed due to a loading error (staff newly trained). This is slightly higher than our manual failures. Automation, the key feature of the Reveos, improved productivity, flexibility, quality assurance and compliance. The impressive PYI data ($\sim 98 \times 10^9$ /unit), indicates the potential to optimize platelet pooling to 4 IPU instead of 5, which will have significant cost and efficiency benefits to SANBS.

P235 | Extending platelet storage time up to ten days with additive qingkailing injection

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Background: The demand for apheresis platelets in hospitals is increasing, yet donations are limited, leading to wastage of unmatched units due to their short shelf life. Numerous platelet additive solutions (PAS) have been developed and approved to extend the average storage time which is five days at room temperature with gentle agitation. Traditional Chinese medicine injections have been purified and designed for detoxification and deoxidation purposes. For instance, Qingkailing injection is commonly used to treat infections, pneumonia, cerebral thrombosis, hemorrhage, and more. Reports suggest that compounds like baicalin, geniposide, and bile acid in Qingkailing injection have functions such as inhibiting platelet activation, protecting against platelet aggregation, and inhibiting cell apoptosis. This solution shows promise as an additive for preserving platelets.

Aims: To explore an alternative approach using intravenous injection in vitro to extend platelet storage time and investigate its function and effectiveness for up to 14 days.

Methods: The estimated percentage of Qingkailing injection in patients' bodies after administration is approximately 1%. Therefore, the in vitro experiments involve collecting 15 samples of mechanically collected platelets (≥ 250 mL/sample), dividing each sample into two portions of 125 mL, and assigning them to the experimental and control groups, respectively. In the experimental group, each portion received approximately 1.25 mL of Qingkailing Injection, resulting in a final concentration of 1%, and were stored under shaking at $22 \pm 2^\circ\text{C}$. The control and experimental groups were subjected to identical storage conditions. Samples from the experimental group were extracted on days 1, 3, 5, 8, 10, and 14 to assess platelet count, pH value, and CD62p expression rate. Samples from the control group were taken for the same tests on days 1, 3, and 5 of storage. Differences in various indicators between the two groups were then compared.

Results: The initial platelet counts collected averaged around 996.17×10^9 / L. Comparing to day 1 (d1), the experimental group's platelet count decreased by 1% to 8% maximum on days 3, 5, 8, 10, and 14. In contrast, for the control group, the platelet counts on days 3 and

5 were 1% to 6% lower respectively compared to day 1. Within 5 days, there was no statistically significant difference in platelet count between the experimental and control groups ($p > 0.05$). The pH values for the experimental group were 7.21, 7.16, 7.03, 6.76, 6.45, and 6.10 on days 1, 3, 5, 8, 10, and 14 respectively. In contrast, for the control group, the pH values were 7.43, 7.06, and 6.63 on days 1, 3, and 5. Regarding CD62p expression rate (%), on day 5, the experimental group and control group showed rates of 70.50 and 83.16 respectively ($p < 0.05$). By day 14, the rate for the experimental group was 82.77 ($p < 0.05$).

Summary / Conclusions: The CD62p expression rate serves as the activation index of platelets, and it was found to be at the same level for day 14 of the experimental group and day 5 of the control group. Upon comparing platelet counts, pH values, and CD62p expression rates within a 14-day period of in vitro preservation, both with and without 1% Qingkailing injection, this additive solution may potentially extend the platelet preservation period at room temperature to at least 10 days. Further studies are underway to assess other relevant parameters and to find out the preserving mechanism.

P236 | Verification of non-DEHP blood bags for an apheresis blood collection system

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Background: The European Union has banned the use of di(2-ethylhexyl) phthalate (DEHP) in blood bags. As such, manufacturers must develop blood bag systems that use alternative plasticizers. One concern with using non-DEHP systems is that the physical properties of the vinyl, such as the ability to withstand centrifugation and temperature fluctuations (freezing/thawing), must be validated to ensure that the bags are suitable for storing blood from collection through distribution.

Aims: The goal of this study was to evaluate the physical properties of non-DEHP blood bags intended for use with the Trima Accel™ Automated Blood Collection System (Terumo BCT, Lakewood, CO). The new non-DEHP red blood cell (RBC) bags are comprised of the proprietary ErythroMate™ vinyl, while the non-DEHP plasma (PLS) bags are comprised of a tris (2-ethylhexyl) trimellitate (TOTM) plasticized PVC. Platelet (PLT) bags have always been comprised of the non-DEHP formulation of butyryl trihexyl citrate.

Methods: All bags were assembled and sterilized per typical protocols for Trima Accel disposables. Bags were tested at initial time ($T = 0$) per the International Organization for Standardization (ISO) standards for apheresis blood bag systems (ISO 3826-4 and 3826-1). Briefly, volume was tested to meet standard practice without overflow. Centrifugation testing consisted of spinning bags at $5000 \times g$ for 10 min at 37°C and 4°C. Internal pressure testing consisted of exposing bags at maximum volume to 50 kPa for 10 min. Tests were required to meet 95% confidence and 95% reliability to be

considered a pass, translating to zero failures in $n = 59$ samples per bag type.

Results: All non-DEHP bags passed volume verification. RBC bags held 632.5 mL (550 mL + 15%). PLT bags held 600 mL + 10% when intended for centrifugation and 1200 mL + 10% when not intended for centrifugation. As PLS bags are available in 3 different volume configurations, 600 mL, 800 mL, and 1000 mL bags were each tested with fresh and frozen plasma. The 600 mL bags held 600 mL + 10% when fresh, 400 mL + 10% when frozen. The 800 mL bags held 800 mL + 10% when fresh, 600 mL + 10% when frozen, and 1000 mL bags held 1000 mL + 10% when fresh, 600 mL + 10% when frozen. Additionally, PLS bags withstood thermal stability testing and did not leak. RBC and PLT bags passed verification tests for centrifugation and showed no evidence of leaking. All RBC, PLT, and PLS bags passed verification of internal pressure and showed no evidence of leaking, meeting the requirement of 95% confidence and 95% reliability.

Summary / Conclusions: All non-DEHP RBC, PLT, and PLS bags at the initial time point ($T = 0$) passed the acceptance criteria and meet the ISO standards for apheresis blood bag systems requirements.

P237 | The effect of vitamin E analogue as an antioxidant on storage of red blood cell

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Background: Despite measures taken to increase the shelf life of red blood cell (RBC) concentrate units outside the body, these units undergo metabolic, structural, biochemical and molecular changes known as "storage lesion" that can reduce the survival and quality of RBCs and lead to severe disorders in patients after transfusion. Oxidative stress plays a major role in the storage lesion that causes oxidation of protein and peroxidation of erythrocyte membrane lipid. The use of natural antioxidants such as vitamin E (α -tocopherol) may help to improve the quality of RBC units by reducing oxidation.

Aims: The aim of this study was to determine the effect of different concentrations in RBC units containing CPDA-A1 stored at 4°C for 35 days.

Methods: Four CPDA1 containing RBC units were selected and each bag was divided into four equal parts. Three bags supplemented with concentrations of 0.125, 0.625 and 3.125 mM of α -tocopherol as test group. One bag supplemented with the solvent (0.5% alcohol) as control group. Test and control units were stored in a refrigerator at 4°C for 35 days. Malondialdehyde (MDA) concentration as the final product of lipid peroxidation, total antioxidant capacity (TAC) and hemolysis index (HI) were measured using commercial kits on days 7, 14, 21, 28 and 35 of storage.

Results: In all three case and control groups, MDA concentration and HI increased and TAC decreased (p -value <0.05). MDA concentration and HI in the 3.125 mM of α -tocopherol group had a lower increase during storage compared to the other cases and control groups. Supplementation of RBC units with α -Tocopherol resulted in significant increase of TAC in all three groups compared to the control group ($p < 0.05$) and had lower reduction during storage.

Summary / Conclusions: Supplementation of RBC units with α -Tocopherol at the onset of storage improves the quality of RBC units by decreasing lipid peroxidation and hemolysis and by increasing TAC. Among the mentioned concentrations, 3.125 mM of α -tocopherol had significantly more antioxidant effect.

P238 | Description of thrombocytopheresis quality on the fifth and seventh day of storage in Central Blood Transfusions Service of Indonesian Red Cross (CBTS IRC)

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Background: Blood services are health efforts to cure disease and restore health that depend on the availability of blood components that are sufficient, safe, easily accessible and affordable. One of the blood components needed is Thrombocytopheresis. In addition to a short life span that is no less important in the storage of Thrombocytes, the quality must be maintained.

Aims: The purpose of this study was to describe the quality of Thrombocytopheresis on the fifth and seventh day of storage.

Methods: The research method used was quantitative descriptive with all Thrombocytopheresis samples using TRIMA, MCS+ and Amicore machines at CBTS – IRC. Research analysis uses descriptive analysis, presenting data in the form of tables and graphs

Results: The results showed that the number of samples was 70 Thrombocytopheresis bags, blood type A Rh Positive 12 bags, B Rh Positive 18 bags, AB Rh Positive 2 bags and O Rh Positive 38 bags. The platelet value content per unit on the fifth day of storage was 100% meets the standard $\geq 2 \times 10^{11}$ per unit with an average value of 2.90 and on the seventh day 100% meets the standard with an average value of 2.86. The content of leukocytes per unit value on the fifth day of storage was 100% meets the standard $\leq 1 \times 10^6$ per unit and on the seventh day 100% meets the standard with an average value of 0.00. The pH value on the fifth day of storage was 100% meets the standard >6.4 with an average value of 7.14 and on the seventh day it was 100% meets the standard with an average value of 7.07.

Summary / Conclusions: The quality of Thrombocytopheresis on the fifth and seventh day of storage at CBTS-IRC still meets the standards

according to MOH Regulatory No. 91 of 2015 Concerning Blood Transfusion Service Standards in Indonesia.

P239 | Abstract withdrawn

P240 | Evaluation of a measuring instrument to support the storage and labelling of fresh frozen plasma

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Background: The inclusion of the Portuguese Institute of Blood and Transplantation—Algarve Delegation, in the National Strategic Program for Human Plasma Fractionation, required planning and implementation of methodologies at the level of processing, storage, and distribution of FFP for the industry, to ensure the production of high-quality plasma.

Aims: Evaluation of a non-contact infrared thermometer for determining plasma temperatures in storage and labelling and establishing the acceptable time interval for FFP labelling.

Methods: The equipment in use was a contact shock freezer fitted with a glycerol dummy bag with a probe inserted inside, whose performance was used as a standard for the infrared thermometer evaluation. Measurements by the infrared thermometer were done by positioning it 5 cm away from the centre of the plasma smooth surfaces. The study was divided into two stages. In Stage 1, the dummy bag was subjected to a 30-min freezing cycle in a fully loaded shock freezer. After completing the cycle, the equipment was opened, the temperature of a dummy bag was measured using an infrared thermometer, and the last record from the probe was taken into consideration. In Stage 2, the dummy bag underwent a 30-min freezing cycle. After completion and equipment opening, the dummy bag was kept connected to the shock freezer and placed on the workbench. Temperatures were taken simultaneously by the above-mentioned methodologies until the dummy bag reached -20°C . Sirius Storage software monitored environment conditions during the two experiments. Regarding eventual differences between the calibrated infrared thermometer and the calibrated probe, specifications from the

P240 Table 1: Temperature of the glycerol dummy bag, immediately after cycle freezing

Probe ($^{\circ}\text{C}$)	Infrared thermometer ($^{\circ}\text{C}$)	Thermal amplitude ($^{\circ}\text{C}$)	Accuracy (manufacturer's specifications)
-42.7	-39.3	3.4	$\pm 4.135^{\circ}\text{C}$ accepted

Environment conditions were temperature 20.30°C and humidity 37.56%.

P240 Table 2: Time interval for attainment of -20°C by both methodologies

Registration time	Probe (°C)	Infrared thermometer (°C)	Accuracy (manufacturer's specifications)	Total Elapsed Time (minutes)
23h49	-40	-36.3	±4.00°C accepted	15
23h52	-35	-31.5	±3.75°C accepted	
23h56	-30	-26.9	±3.50°C accepted	
00h00	-25	-22	±3.25°C accepted	
00h04	-20	-17.8	±3.00°C accepted	

Environment conditions were temperature 20.23°C and humidity 56.19%.

manufacturer were followed. Briefly, the measurement range is -60 to 550°C and the accuracy acceptable is $T_{obj} = -60 \sim 0^\circ\text{C}$: $\pm 0.05/^\circ\text{C}$ (°C).

Results: Stage 1:

Stage 2:

Summary / Conclusions: The thermometer under study was evaluated and accepted for temperature readings within the range inherent to the FFP production/storage process. The probe took 15 min to reach -20°C under the analysed ambient temperature conditions and was roughly coincident to infrared thermometer. Labelling should occur within a maximum of 10 min.

P241 | Statistical analysis of split sample laboratory studies observations from a systematic review of irradiated platelets

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Background: In a systematic review and meta-analysis of irradiated platelets we included laboratory studies which had used split (paired) samples to compare irradiated platelets to non-irradiated controls. With paired samples, statistical analysis can take advantage of the fact that each sample is known to have been identical to its control before intervention. Analysing the difference between each pair removes uncertainty due to population variability and produces a smaller standard deviation (SD) for the mean difference between pairs than the SD for the difference between the two group means. For continuous measures, the mean difference between pairs and its SD is commonly used for paired comparisons, using the t-distribution to construct a confidence interval and the paired t-test for hypothesis testing. A non-parametric alternative is the Wilcoxon signed rank test. Non-parametric analysis is commonly used for small samples which are not normally distributed, although for a paired t-test it is only the difference between pairs that needs to be normally distributed. Non-parametric tests yield results which are difficult to include in a meta-analysis.

Aims: To establish the statistical methods used for the paired data in the studies included in this review, and the availability of results for meta-analysis.

Methods: The protocol was prospectively registered on PROSPERO (CRD42023441930) and complies with PRISMA-P. As part of quality assessment and data extraction, for each included study we checked for a statistical methods section and examined the reporting of results to ascertain whether a paired analysis was used and, if so, whether enough information was reported to include the paired result in a meta-analysis.

Results: The review included 43 laboratory studies: 33 (77%) with full text articles available and 10 abstracts. 26 (79%) of the 33 full texts included a statistical methods section with enough detail to establish what type of analysis, paired or independent, was reported. In addition, one full text and one abstract reported sufficient information elsewhere to establish what method they used. 14 (33%) of the 43 studies used a paired analysis, but only one of these reported enough information to extract the SD of the mean difference. Five others (12%) used a paired t-test but only reported means and SDs per group, with no SD, confidence interval, or p-value for the mean difference. Eight (19%) used a paired non-parametric test, with one of these reporting per group means and SDs. A total of 25 studies (58%) reported per group means and SDs. 18 studies (42%) reported no meta-analysable data.

Summary / Conclusions: Of the 43 included studies, only one-third of them used a paired analysis, and only one of these reported the paired analysis in enough detail to allow the SD of the mean difference to be estimated for meta-analysis. Just over half of the included studies reported enough information to meta-analyse continuous outcomes as if they were measured on independent groups. The standard error of the pooled results based on treating the data as independent groups will be larger than if the paired results were available. This will not introduce any systematic bias, but confidence intervals will be wider than if the paired results were available for inclusion. We encourage researchers using paired samples to use paired analysis, and to report some measure related to the standard error of the mean difference to allow the paired result to be included in systematic reviews.

Blood product / components—blood components

P242 | Abstract withdrawn

P243 | Platelet production—reducing risk in assembling platelet pools from buffy coat with the support of software

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Background: The blood establishment “Officina Trasfusionale di AUSL Romagna” (OT), produces platelet concentrates (PC) by assembling a platelet pool of 4 or 5 buffycoats (BC) using the semi-automatic

TACSI system (TerumoBCT). On average, around 6000 platelet therapies/year are made available for transfusion in the relevant territory with 6096 PCs produced in 2022 and 5556 PCs produced in 2023. Risk analysis of the production process, carried out with FMECA, highlights that the assembly and connection phase of the BCs is a critical activity as it is based on manual skills and subjectivity of the operators and is without computer control.

Aims: increase in the average platelet yield, standardization, traceability, safety, and quality of platelets production from BC pools

Methods: T-Pool Select receives from the BBIS useful information (donation code, blood group, rh, platelet count and donation date) for the selection of valid and available BCs. By setting parameters such as number of BCs per pool, yield minimum, and maximum platelets for pool, the software suggests combinations to obtain the best product. TOMEs Stations, the connection software with TSCDII sterile connectors, tracks the sterile connection process of the BCs assembled in the pool. During assembly, the bar codes of the BCs are scanned by the operator, and T-Pool Select assigns a numbering to each one. The operator then positions each BC in correspondence with the indicated numbering on specially created grids with numbered spaces. After reading all bar codes, available BCs are divided into groups corresponding to the workable pools of 4-5 BCs. The assembled pools are then recorded in the BBIS. Next, TOMEs Stations interfaces with the BBIS and the TSCDII sterile welder to guide the steps of the connection process while tracking operator ID and the lot numbers of kits and solutions in use. Additionally, the software verifies the execution of correct connections by requesting the bar code of each BC.

Results: Prior to software implementation, the production process of platelet pools was very stable with average yields of 3.3×10^{11} (SD 0.4) for 5 BC pools and 2.62×10^{11} (SD = 0.38) for 4 BC pools. After introduction of the new software, the impact of the change control (CC) was verified with an initial analysis of the platelet yields from 500 PCs produced using the T-Pool Select system average yield of PCs produced with T-Pool Select was 3.5×10^{11} (SD = 0.36). In 2023, a total of 5434 PCs obtained from 5 BCs and 122 PCs obtained from 4 BCs were analyzed, all produced with the support of T-Pool Select. The yield of the PCs confirmed the CC data, highlighting an average value of 3.44×10^{11} (SD = 0.36) for the 5 BC pools and 2.96×10^{11} (SD = 0.46) for 4 BC pools.

Summary / Conclusions: After implementation of T-Pool Select and TOMEs Stations, we observed an increase in the average platelet yield and increased standardization, particularly for products consisting of 5 BCs. T-Pool Select and TOMEs Stations provide the operator who manages the assembly of BCs with process support, allowing for an increased level of traceability, safety, and quality of platelets production from BC pools.

P244 | Abstract withdrawn

P245 | Verification of a 6-part differential haematology analyzer at Central Blood Transfusion Services of Indonesian Red Cross

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Background: Prior to the use of new haematology analyzers, it is necessary to conduct an assessment that evaluates stability, precision, linearity, and carry-over within the expected outcomes. The laboratory has the authority to determine the standard and verification limit for this evaluation. An example of a next-generation automated hematology analyzer is the Sysmex XN-1000 (Blood Bank Mode), which provides a comprehensive blood count (CBC) and also offers 6-part white blood cell (WBC) differential counts

Aims: The aim of this study was to perform a comprehensive verification of a 6-part differential haematology analyzer Sysmex XN-1000 Blood bank Mode (Sysmex Corporation, Kobe, Jepang), before routine operation for blood component quality control assessment

Methods: The verification protocol included precision (within- and between-run), which was calculated from observed and manufacturers' recommended values, carryover, and method comparison. Acceptance criteria were based on manufacturer technical specifications (Sysmex) and the MOH of Indonesian Regulatory. Samples from Leucodepleted Packed Red Cell (LDPRC), Platelet Apheresis (PLTAPH), Leucodepleted Fresh Frozen Plasma (LDFFP) were analyzed. A total of 334 samples of all blood components and 40 EDTA samples were used for verification of the analyzer Sysmex XN-1000 (Blood Bank Mode)

Results: The precision of Sysmex XN-1000 (Blood Bank Mode) was evaluated for both within-run and between-run measurements, and all parameters demonstrated acceptable levels. The coefficients of variation were particularly low for mean corpuscular volume (MCV), with values of 0.2% and 0.4% for within-run and between-run measurements, respectively. Furthermore, the estimated bias for all parameters fell within the predetermined acceptance criteria. In addition, the carryover estimates for the different parameters were assessed, and found to be within the manufacturers' specifications, which stipulate a maximum allowable value of 1%. Notably, the platelet count (Plt) exhibited no detectable carryover, while the red blood cell count showed a similarly negligible level of carryover

Summary / Conclusions: In conclusion, the results of this study demonstrate that the Sysmex XN-1000 (Blood Bank Mode) haematology analyzer performs reliably, with satisfactory precision, minimal bias, and negligible carryover effects. These findings confirm that the analytical performance of the Sysmex XN-1000 (Blood Bank Mode) meets predefined quality goals and is suitable for routine operation in laboratory blood component quality control assessment

P246 | Irradiation and washing of red blood cells prepared from whole blood collected in DINCH-PVC and stored in BTHC-PVC storage bags

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Background: Because of its potential toxicity, the European Commission is sunsetting the use of DEHP in Medical Devices. Therefore, alternative plasticizers are investigated for use in blood collection systems. Promising results have been obtained using BTHC-plasticized PVC bags in combination with the PAGGSM red cell additive solution (AS). However, the impact of secondary treatments of non-DEHP-stored red cells, such as irradiation for lymphocyte inactivation and washing to remove certain plasma proteins, on their in vitro quality still require further investigation. As it is known that both irradiation and washing result in faster deterioration of red cell in vitro quality, it is important to also assess the impact of such secondary treatments on non-DEHP stored red cells.

Aims: To investigate the effects of gamma- and X-irradiation and washing on RBC collected and stored in non-DEHP systems.

Methods: Whole blood units (500 ± 50 mL), collected in DINCH-PVC (Fresenius Kabi, GQ422NL) were processed into plasma, buffy coat and RBC after overnight hold at ambient temperature. After adding PAGGSM, RBCs were leukodepleted over the inline filter into the BTHC storage bag and stored at 2-6°C. Study A – Irradiation. At day 14 of storage, 8 pools of 3 RBCs were made. Pools were equally divided over the original BTHC bags. 8 RBCs were used to make paediatric RCCs. From each RBC, 3 paediatric RBCs were prepared: 70-75 mL RBC in 150 mL DINCH bag. At day 14, both whole and paediatric RBCs were either not irradiated, γ-irradiated or X-irradiated. After irradiation, RBC were stored at 2-6°C for up to 28 days after collection. RBCs were sampled and analysed for quality parameters at day 1, before irradiation (day 14) and day 28. Study B – Washing. At day 30 of storage, 12 RBC were washed with PAGGSM, resuspended in 120 mL of PAGGSM and stored at 2-6°C. RBC were sampled and analysed for K⁺, haemolysis and ATP before washing (day 30) and 5 days after washing (day 35).

Results: Study A. Irradiation of RBC in BTHC/PAGGSM resulted in increased free K⁺, lactate and haemolysis and decrease ATP levels, with only minimal differences between γ- and X-irradiation. The results were comparable with those obtained with irradiation of RBCs in DEHP/SAGM (Table 1). Irradiation of paediatric RBC in DINCH/PAGGSM resulted in comparable changes as in standard RBC. Study B. Washing resulted

in lowered free K⁺ levels from 33 ± 1.5 mmol/L before washing (day 30) to 6.6 ± 0.6 mmol/L at day 35. Haemolysis at day 35 amounted to 0.31 ± 0.05% while ATP levels were 4.5 ± 0.5 μmol/g Hb.

Summary / Conclusions: Study A. RBCs prepared from WB collected in a DINCH collection bag, stored in PAGGSM in BTHC (standard RBC) or DINCH paediatric RBC bags showed comparable changes after γ- and X-irradiation as RBCs stored in DEHP and SAGM. Study B. Washing of RBCs stored in BTHC using PAGGSM resulted in efficient removal of free K⁺. At day 35, 5 days after washing, all washed RBCs complied to the requirements for haemolysis (<0.8%) and ATP level (>2.7 μmol/g Hb).

P247 | Enhancing platelet component quality—a comparative analysis in apheresis

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Background: Apheresis-based platelet (PLT) collection offers logistical advantages, addressing targeted PLT needs for refractory patients and providing contingency during PLT pool stock decreases. Various cell separator technologies differ in concurrent product outcomes and in the mechanisms of leukoreduction, which may occur within the separator (Trima Accel[®] and Amicus[®]) or through post-separation leukoreduction filters (MCS[®]+9000), among other aspects.

P247 - Table 1. Platelet and leukocyte contents.

	Trima Accel [®]	Amicus [®]	MCS [®] +9000
Platelet yield (mean ± SD x10E11/U)	4.5 ± 1.55	3.28 ± 0.89	3.31 ± 0.81
Double product (%)	37.8	0	2.7
Adult dose (%)	50.0	76.5	84.3
Pediatric dose (%)	6.9	17.7	7.3
Rejected (%)	5.3	5.9	5.7
Residual leukocytes (mean ± SD x10E6/U)	0.42 ± 1.28	0.83 ± 1.23	3.44 ± 27.67
OOS leukocytes (%)	3.19	17.65	14.27

P246 Table 1. Effect of irradiation (at day 14) on RBC stored in BTHC/PAGGSM and DEHP/SAGM (mean ± SD, n = 8)

Day 28	Plasticizer/AS	K ⁺ (mmol/L)	Hemolysis (%)	ATP (μmol/g Hb)
no irradiation	BTHC/PAGGSM	40 ± 6.0	0.18 ± 0.02	5.2 ± 0.3
γ-irradiation	BTHC/PAGGSM	61 ± 2.6	0.35 ± 0.07	4.7 ± 0.3
	DEHP/SAGM	56 ± 1.1	0.43 ± 0.11	3.6 ± 0.7
X-irradiation	BTHC/PAGGSM	63 ± 1.8	0.39 ± 0.09	4.7 ± 0.3
	DEHP/SAGM	57 ± 1.2	0.50 ± 0.13	3.6 ± 0.8

P247 - Table 2. Leukocyte content in PLTA collected with MCS®+9000

Donation center	n	Leukocytes (mean ± SD × 10E6/U)	OOS (%)
A	90	0.58 ± 2.44	7.9
B	142	0.65 ± 4.17	6.4
C	296	2.10 ± 13.55	11.2
D	31	0.75 ± 2.09	12.9
E	92	4.36 ± 19.96	17.4
F	114	0.81 ± 2.08	12.3
G	87	3.16 ± 7.67	43.7
H	115	0.94 ± 1.96	16.5
I	535	7.52 ± 46.63	17.8
J	159	0.17 ± 1.22	1.3

Aims: Characterize and analyze PLT components obtained through apheresis using diverse cell separators, assessing PLT yields, residual leukocyte contamination, and volume-to-yield ratios with an automated cell counter.

Methods: PLT products were collected by using Trima Accel®, Amicus® and MCS®+9000 in blood donation centers belonging to our regional blood establishment. Plateletpheresis (PLTA) samples were collected through a closed circuit and placed in sera tubes. Cell population counts were performed using the Blood Bank mode of the automated cell counter Sysmex XN-1000, which analyzes platelets and residual leukocytes by flow cytometry with specific fluorescent dyes. PLT and leukocyte concentrations were measured, and the total content per unit (mean ± standard deviation) was calculated. For PLT content (PLTx10E11/U), the following values were considered: ≥4.9 for double products, 2.4-4.9 for adult dose, 1.8-2.3 for pediatric and <1.8 units were rejected. For leukocytes, the number of units within (OK) and outside (OOS) specifications criteria was <1 × 10E6 residual leukocytes/unit.

Results: A total of 1866 PLTA obtained using Trima Accel® (n = 188), Amicus® (n = 17) and MCS®+9000 (n = 1661) presented variations in the collected products, as summarized in Table 1. For MCS®+9000, high variability according to the donation center was observed (Table 2).

Summary / Conclusions: PLTs obtained by apheresis exhibit differential characteristics based on the technology used for their collection. While the PLT content in components obtained with different cell separators analyzed shows acceptable performance averages, the leukodepletion process is crucial to ensure the final quality of each component. PLTA generated by Trima Accel® showed the highest PLT yields, as well as a lower average of residual leukocytes and a lower absolute number of units outside specifications. Conversely, MCS®+9000 generated products with an elevated average of leukocytes and a standard deviation with a wider range that may need to be filtered prior to distribution. The automation of the leukoreduction process appears to produce products that better meet specifications

than manual filtration, which could be affected by the level of training of the personnel obtaining it. Nonetheless, it must be considered the need of obtaining double PLT products or a concurrent plasma according to stock management.

P248 | Towards removal of non-leucocyte depleted blood components from European standards

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Background: Leucocyte depletion of blood components is an effective first-line defence to reduce the risk of transfusion reactions, HLA alloimmunisation and infections including cytomegalovirus. The use of this strategy is increasingly becoming standard practice. A blood component containing less than 1 × 10⁶ leucocytes is considered leucodepleted. The European Directorate for the Quality of Medicines & HealthCare (EDQM) of the Council of Europe *Guide to the preparation, use and quality assurance of blood components* ("the Blood Guide") contains technical guidelines, standards and blood component monographs. It is periodically reviewed and updated to ensure inclusion of current scientific information, set high standards and promote best practice.

Aims: The use of non-leucodepleted blood components in Europe was surveyed, with the aim to consider their removal from future editions of the Blood Guide.

Methods: The most recent EDQM survey for the "Annual report of collection, testing and use of blood and blood components in Europe" included an annex with the aim to collect data on the use of blood components currently included in the Blood Guide monographs. The survey was disseminated in 2023 to the 38 represented member states (MS) of the European Committee on Blood Transfusion (CD-P-TS), of which 34 MS provided answers. Reply options on usage included "yes", "yes, but only as exception", "yes, but considering phasing out" and "no".

Results: The most common red cell component was *Red cells, leucodepleted, in additive solution (AS)*, used in 100% of MS. For non-leucodepleted red cells, *Red cells, non-leucodepleted, in AS, buffy coat removed* was most frequently used (24% of MS; one further MS was

considering phasing out), whereas *Red cells, non-leucodepleted, buffy coat not removed, no AS* was the least used (3%; i.e., one MS). Two additional MS indicated non-leucodepleted red cells were in use, but only as exception. The most common recovered-platelet component was *Platelets, pooled, leucodepleted, in platelet additive solution (PAS)* (used in 79% of MS). For non-leucodepleted recovered platelets, *Single units, in plasma* was most frequently used (18% of MS; in one MS only as exception), while *Platelets, pooled, non-leucodepleted, in plasma* were used least (9%; in two further MS used only as exception). Leucodepleted apheresis platelets were used in the majority of MS (68% plasma/PAS, 59% plasma), but non-leucodepleted were also in use (9% plasma/PAS, 26% plasma). None of the MS were phasing out non-leucodepleted platelets. Plasma does not currently have distinct monographs for leucodepleted components. Overall, 56% of MS had completely removed non-leucodepleted components from their blood supply. 35% of MS still used non-leucodepleted red cells and another 35% still used non-leucodepleted platelets. Of the MS using either non-leucodepleted red cells or platelets, 75% were overlapping. No MS used non-leucodepleted components exclusively.

Summary / Conclusions: More than half of the MS already leucodeplete 100% of their blood supply. Additional MS are considering phasing out non-leucodepleted components, and in some cases, only using them as exception. No MS completely lacks leucodepleting measures. Collectively, the data suggest that leucodepletion is becoming state-of-the-art all across Europe. In line with acknowledged best practice to increase patient safety, this survey provides a solid basis for a future proposition to remove non-leucodepleted components from the Blood Guide.

P249 | Assessing the neutralizing capacity of early pandemic convalescent plasma against emerging SARS-CoV-2 variants

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Background: COVID-19 convalescent plasma (CCP) has shown promise in halting disease progression in COVID-19 patients. However, CCP products are often underutilized and may be stored for extended periods before administration. The efficacy of these CCP products against emerging variants of SARS-CoV-2 that arise later in the pandemic is a topic of concern.

Aims: In this study, we investigated the neutralizing capacity of plasma collected from non-vaccinated donors during the early stages of the pandemic against subsequent emergent variants of SARS-CoV-2.

Methods: Plasma donors, who had experienced COVID-19, were recruited. Levels of anti-SARS-CoV-2 antibodies (including Spike Trimer, Receptor Binding Domain (RBD), S1, S2, and nucleocapsid protein) were

quantified using a multiplex serological assay (MULTICOV AB, NMI, Reutlingen, Germany). The neutralization capacity of RBD antibodies was assessed through an ACE2 binding inhibition assay. ACE2 binding inhibition was measured against wild type virus and variants of concern, including alpha, beta, gamma, delta, omicron BA.4, and omicron BA.5. Friedman test with Dunn's multiple comparisons was employed to compare ACE2 inhibition across different variants with the wild type.

Results: A total of 47 non-vaccinated CCP donors (25 female; 53%) with a median age of 47 years (range 40-60) were included in the analysis. CCP donations were obtained between January 2021 and June 2021. Compared to the wild type (18.2%; 12.9-21.1%), ACE2 binding inhibition was similar against the alpha (17.7%; 13.7-23.4%) and delta (16.1%; 13.2-18.5%) variants ($p > 0.05$). However, ACE2 binding inhibition was significantly lower than that of the wild type against the beta (12.5%; 9.53-15.4%), gamma (10.1%; 6.41-13%), omicron BA.4 (5.22%; 1.99-6.92%), and omicron BA.5 (6.46%; 2.75-8.76%) variants ($p < 0.05$).

Summary / Conclusions: Our findings indicate a potential decrease in the neutralizing efficacy of antibodies present in convalescent plasma from non-vaccinated donors against newly emerging SARS-CoV-2 variants. This emphasizes the critical need to evaluate the neutralization potential of therapeutic products against prevalent variants of SARS-CoV-2 before their clinical application, ensuring optimal clinical effectiveness.

P250 | Long-COVID patients show enhanced formation of COHb and accumulation of toxic porphyrin intermediates in erythrocytes

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Background: The Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was the cause of the global COVID-19 pandemic. Irrespective of the disease severity, a growing number of patients report a delayed state of exhaustion called chronic fatigue syndrome (CFS). At least 10% of all COVID-19 patients experience various persistent or new health complaints after SARS-CoV-2 infection lasting longer than three months. In addition to CFS, Long-COVID patients also suffer frequently from dyspnea and anemia. Recently, we demonstrated that SARS-CoV-2-induced dysregulation in hemoglobin (Hb)- and iron-metabolism contributes to the severe systemic course of COVID-19.

Aims: Since changes in Hb structure may also be significantly involved in the development of Long-COVID symptoms, we put particular emphasis on the analysis of Hb- and iron metabolism of Long-COVID patients compared to healthy donors.

Methods: We performed blood gas analysis of more than 30 Long-Covid-19 and control patients. Furthermore, we measured hematological parameters and Raman spectra of red blood cells (RBCs) from peripheral blood of Long-COVID patients and compared those with healthy donors. Metabolites of the porphyrin metabolism were analyzed in plasma-reduced whole blood samples using mass spectrometry.

Results: Whereas, no differences in the RBC count, Hb content and hematocrit were observed in the blood of Long-COVID patients compared to healthy donors, the amount of COHb was significantly increased. Furthermore, we found significant changes in spin state of the iron in Hb, which were partially comparable to packed RBCs stored for 42 days. In addition, BE- and BI values but not pH were diminished in Long-COVID patients compared to healthy donors, albeit still within the normal range. Lactate levels were also significantly elevated in the blood of Long-COVID patients. Thus, O₂Hb was reduced in Long-COVID patients. Mass spectrometry showed significantly increase in toxic porphyrin intermediates (uroporphyrin III, coproporphyrin I and III) in erythrocytes of Long-COVID patients.

Summary / Conclusions: Our data suggest the formation of a possible secondary coproporphyrin as result of SARS-CoV-2 infection and show drastic changes in Hemoglobin-associated vibrational modes in samples of Long-COVID patients. Together with the diminished Acid-base balance and enhanced formation of COHb, we suggest an impaired erythrocyte functionality in Long-COVID patients, leading to diminished oxygen supply. This in turn could be an explanation for the CFS and dyspnea as well as anemia.

Instructions for use (IFU) from the manufacturer, we aimed to reduce the number of IPU's from seven to six.

Aims: Reduce the number of IPU per platelet pool without decreasing the platelet content below approved numbers.

Methods: Whole blood from 228 blood donors was collected in the Reveos® blood system (Terumo BCT) and divided into three groups, Fresh (14 pools): IPU centrifuged same day as collection, Night (12 pools): IPU centrifuged the day after collection, and Mixed group (12 pools): mixture of Fresh and Night IPU's. Platelet Yield Index (PYI) from Reveos is recommended to be between 400-453, reference value 420. Pooling procedure: 6 IPU's with pooling set and PAS (250 ml SSP +) to (IPU-PC), rest for an hour on the bench, placed on the agitator for one hour and connected to IBS dual storage sets (INTERCEPT® blood system, Cerus). After addition of amotosalen to IPU-PC photochemical treatment with UVA using INT100 (3.9 J/cm²) and transfer into CAD system for 4-16 h and then into the final containers. The pool is then divided into two identical platelet units for transfusion.

Results:

Summary / Conclusions: The use of Reveos platelet pooling system with 6 IPU (fresh, night or mix) and 250 ml SSP+ meets the quality requirements according to EDQM's Guide to the preparation, use and quality assurance of blood components. By decreasing the number of IPU's from 7 to 6, we could see that the number of pools that get stuck in the filter during filtration has decreased. Conclusion are that we can reduce the number of donations necessary per unit, whilst adhering to the manufacturer's Instructions for use (IFU).

P251 | Reducing the number of Interim Platelets Units (IPUs) in the production of double-dose pathogen reduced platelet units

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Background: In our regional blood center, Reveos technology is used for pooled platelet production. Seven Interim platelets units (IPUs) used to be pooled to produce one double-dose of pathogen reduced platelets. As the recent EU directive MDR requires concordance with

P251 - Table 1: Results

Group of pools	Platelet Yield Index	Net weight (g)	WBC < 1 × 10 ⁶	Platelet ≥ 2 × 10 ¹¹
Fresh	419 (390-454)	206 (200-214)	0.00 (0.0-0.0)	2.30 (1.77-2.52)
Night	422 (405-455)	201 (191-207)	0.16 (0.0-0.6)	2.28 (1.95-2.50)
Mix (fresh and night)	417 (402-434)	203 (198-208)	0.03 (0.0-0.1)	2.35 (2.05-2.59)

P252 | Evaluating batch variation in human platelet precipitate and human platelet lysate

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Background: Human platelet precipitate (HPP) is produced as a by-product during human platelet lysate (HPL) manufacturing. It is rich in fibrinogen and growth factors, including transforming growth factor- β 1 (TGF- β 1), platelet-derived growth factor (PDGF), epidermal growth factor (EGF), and fibroblast growth factor (FGF), which have wound healing properties. HPP manufactured under good manufacturing practice conditions is used to make a platelet-derived hydrogel that forms a support matrix for bioengineered human skin equivalents, which can be used for applications such as burns and wound healing.

Aims: The aim of this study was to compare fibrinogen and growth factor concentrations in different batches of HPP and HPL.

Methods: Apheresis platelet concentrates were frozen at -80°C , then thawed at 37°C and centrifuged to remove debris from the supernatant. Following further freeze-thaw steps, the products were centrifuged to separate the HPP from the HPL. Four units each of HPP and HPL were pooled together respectively to produce a single batch, and three separate batches of HPP and HPL were produced. A lower limit of 5 g/L was set for the fibrinogen content of HPP, which was deemed to be the minimum requirement for formation of a stable hydrogel. Fibrinogen was measured using a coagulation analyser, total protein by bicinchoninic acid assay and growth factors (TGF- β 1, PDGF-AB, PDGF-BB, EGF and FGF) by enzyme-linked immunosorbent assay. Mean, standard deviation, ranges and coefficients of variation (CV) were calculated.

Results: The volume of the HPP was very consistent between batches, whilst the HPL volume varied, due to variations in the starting volume of the components. The fibrinogen concentration of the HPP also varied between batches (20% CV; range 9.27–13.73 g/L) compared to the HPL (9% CV; range 1.21–1.46 g/L) due to variations in the fibrinogen content of the starting components. However, all

three batches consistently met the pre-requisite specification 5 g/L for fibrinogen. As with fibrinogen, there was also variation in the growth factor concentrations and total protein in the three batches of HPP and HPL (Table 1). The inter-batch variation differed for each growth factor, with very low (<5%) CV for PDGF-AB and PDGF-BB, low (<15%) CV for EGF and TGF- β 1 and very high (>30%) CV for FGF. The level of correlation between growth factors and the fibrinogen concentrations ranged from $R^2 = 0.2559$ for FGF to $R^2 = 0.9129$ for PDGF-AB. Total protein in HPP and HPL was 67.8 g/L and 57.9 g/L respectively; the inter-batch variability was low (<10% CV).

Summary / Conclusions: There was variation in batches of HPP and HPL, which was largely donor-dependent. However, fibrinogen concentration consistently met the lower limit of 5 g/L set for HPP instead.

P253 | Safety and effectiveness of irradiated platelets for transfusion—systematic review and meta-analysis of in-vitro, in-vivo, and clinical outcomes

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Background: Irradiation of platelet components inactivates residual leucocytes to prevent transfusion-associated graft-versus-host disease (TA-GvHD). It is a widely held belief that the irradiation has no important detrimental effect on platelet quality. Some organisations routinely irradiate all platelets for transfusion, while others irradiate to meet demand. Irradiation of all platelets would simplify stock management, and reduce waste and the risk of issuing non-irradiated platelets when irradiation is indicated.

Aims: To identify any potential harms and quantify the effectiveness of irradiated platelets compared to non-irradiated platelets for transfusion.

Methods: The protocol for this review was prospectively registered on PROSPERO [CRD42023441930], and the review was conducted in accordance with PRISMA guidelines.

Results: Our search identified 3002 references, of which we included 44 studies. Forty-one studies were in-vitro studies, two studies were in-vitro with in-vivo follow-up, and one study reported clinical outcomes in thrombocytopenic patients. x-ray was used exclusively in just three studies, and alongside gamma in one study. Two studies did not report the source of irradiation. The remaining 39 studies used gamma irradiation only. Risk of bias (ROB): We assessed ROB for studies reporting clinical and in-vivo outcomes using ROB 2 (3 studies). We adapted a ROB tool designed for animal studies to assess ROB for the studies reporting in-vitro outcomes (43 studies). For in-vitro, 7 of the 43 studies had high ROB in at least one domain (sample handling, missing outcome data, variation in analysers, manufacturer sponsorship); the remaining 36 studies had low or unclear ROB in all domains. Two of the 3 in-vivo/clinical studies assessed using ROB 2 had some concerns (randomisation process, lack of information throughout) and one was high ROB overall (missing outcome data). No study was low risk in all domains, or unclear in all domains. Intervention effect: Irradiated platelets did not significantly differ

P252 - Table 1: A summary of key parameters for platelet precipitate and platelet lysate

	Platelet Precipitate	Platelet Lysate
Volume (mL)	113.9 \pm 6.1 (5.4)	1113.0 \pm 27.3 (2.5)
Fibrinogen (g/L)	11.2 \pm 2.3 (20.4)	1.33 \pm 0.13 (9.4)
EGF (ng/mL)	2.0 \pm 0.3 (14.9)	1.2 \pm 0.1 (5.1)
FGF (pg/mL)	169.5 \pm 51.1 (30.2)	162.6 \pm 59.0 (36.3)
PDGF-AB (ng/mL)	32.8 \pm 1.2 (3.8)	22.6 \pm 0.4 (1.9)
PDGF-BB (ng/mL)	17.5 \pm 0.4 (2.4)	8.1 \pm 0.9 (10.8)
TGF- β 1 (ng/mL)	136.8 \pm 14.6 (10.6)	44.6 \pm 3.9 (8.6)
Total protein (g/L)	67.8 \pm 4.0 (5.9)	57.9 \pm 2.7 (4.7)

EGF, epidermal growth factor; FGF, fibroblast growth factor; PDGF-AB, platelet-derived growth factor-AB; PDGF-BB, platelet-derived growth factor-BB; TGF- β 1, transforming growth factor β 1.

Data are mean \pm SD (% CV), $n = 3$.

from non-irradiated platelets for most outcomes and timepoints where data were analysable, though the evidence favoured no irradiation (control) for: Platelet count (day 5): 6 studies, 908 samples, Mean Difference (MD) -43.07 [95% CI -76.14, -10.01] $\times 10^9/L$. pH (day 7): 7 studies, 134 samples, MD -0.04 [-0.07, -0.00]. Lactate (day 5): 6 studies, 940 samples, MD 0.34 [0.16, 0.53] mmol/L. Glucose (0-48hrs): 6 studies, 152 samples, MD -0.18 [-0.37, 0.00] mmol/L. P-selectin (day 5): 9 studies, 966 samples, MD 1.58 [0.72, 2.45] %. Annexin V binding (day 5): 2 studies, 24 samples, MD 1.31 [0.06, 2.56] %. And favoured irradiation for Platelet factor 4 (day 7), based on data from a single study (24 samples, MD -1.81 [-3.31, -0.31] mcg/ml).

Summary / Conclusions: The evidence base is limited. Only one study had a sample size large enough to detect small but important differences, and only half the studies could be included in the analysis. Overall, irradiation has little to no effect on most markers of platelet quality and effectiveness. Where there is evidence of detriment from irradiation, differences are small *in-vitro*, and are unlikely to impact clinical outcomes following transfusion. There is very limited evidence for x-ray as a source of irradiation and, given the benefits of using x-ray over gamma irradiation (ease of use and safety requirements), we would welcome further research comparing x-ray to gamma, and x-ray to a non-irradiated control.

P254 | Impact of changing from PVC-DEHP to PVC-DEHT whole blood collection sets on plasma *in vitro* quality

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Background: The phthalate di(2-ethylhexyl) phthalate (DEHP) has long been used as the primary plasticizer in polyvinyl chloride (PVC) whole blood collection sets. However, animal study data suggesting potential toxicity and reproduction concerns have led to regulatory changes being introduced in the

P254 - Table 1: *In vitro* quality of WB-derived FP produced using PVC-DEHT or PVC-DEHP 500 mL top / bottom whole blood collection sets. Table displays results as mean (\pm standard deviation) for $n = 12$ pairs. Statistical significance was determined using paired *t*-tests, except for Prothrombin, for which a non-parametric Wilcoxon matched-pairs test was used.

	DEHP	DEHT	p-value
Volume (mL)	279 (\pm 15)	279 (\pm 14)	0.9544
FVIII (IU/mL)	1.07 (\pm 0.26)	1.07 (\pm 0.25)	0.8800
FVII (IU/mL)	1.31 (\pm 0.10)	1.30 (\pm 0.13)	0.7816
FV (IU/mL)	1.06 (\pm 0.10)	1.06 (\pm 0.07)	>0.9999
Prothrombin (s)	13.23 (\pm 0.28)	13.29 (\pm 0.19)	0.3877
Fibrinogen (g/L)	2.84 (\pm 0.32)	2.86 (\pm 0.31)	0.4783

European Union banning the use of this plasticizer in medical devices. European manufacturers are now focused on the development and regulatory registration of DEHP-free blood bag systems. One PVC plasticizer being investigated as an alternative to DEHP is di (2-ethylhexyl) terephthalate (DEHT). While the impact of DEHP on red blood cells is well known, there is little data to determine if removal of DEHP and replacement with DEHT will have any significant impact on the other whole blood (WB) derived components; specifically, platelet concentrates, plasma, and plasma factors.

Aims: This study aimed to compare the *in vitro* quality of frozen plasma (FP) derived from whole blood collected in top / bottom whole blood collection sets made from PVC-DEHP or PVC-DEHT using a pool and split study design.

Methods: Approximately 480 mL of whole blood was collected into 500 mL PVC-DEHT prototype top/bottom whole blood collection sets (Macopharma REF: PRORQT4-A) in twelve ABO matched pairs. On Day 0 post-collection, the two matched whole blood units were pooled and split evenly back into one of the PVC-DEHT sets and a PVC-DEHP collection set (Macopharma 500 mL top/bottom set REF: LQT710X) with the same configuration and the anticoagulant drained. Whole blood was held overnight at 18–24°C on plastic crates without cooling trays. On Day 1 the paired whole blood units were centrifuged and separated using the same centrifugation (Hettich Roto Silenta; 4898 $\times g$ for 10 min at 21°C) and extraction (Macopress Smart) programs. Plasma units were weighed, then placed in a freezer at $\leq -18^\circ\text{C}$ within 24 h of stop bleed time and were slow frozen. FP was thawed ≥ 30 days post-collection and aliquoted into 2 mL cryotubes, which were stored in a -80°C freezer and thawed immediately prior to testing FVIII, FVII, FV, fibrinogen and prothrombin time (PT; Stago, STA Compact).

Results: *In vitro* quality results for $n = 12$ paired PVC-DEHP and PVC-DEHT FP units are summarized in Table 1. No statistically significant differences in volume, FVIII, FVII, FV, fibrinogen, or PT were observed between PVC-DEHP and PVC-DEHT FP units.

Summary / Conclusions: The replacement of DEHP with DEHT plasticizer in PVC whole blood collection sets does not statistically nor materially impact the *in vitro* quality of the resulting frozen plasma.

P255 | Stability of apheresis plasma treated with amotosalen and UVA stored for 3 years

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Background: In France, the shelf-life of plasma treated with amotosalen and UVA (FFP-A-UVA) for pathogen reduction (INTERCEPT™ Blood System, Cerus) is currently of 12 months from the date of collection.

Aims: In the context of a need to extend the storage time for COVID-19 convalescent specific plasma, the stability of FFP-A-UVA over 36 months was evaluated.

Methods: 34 FFP-A-UVA triplets (3 units of at least 200 mL from the same COVID-19 convalescent apheresis plasma donation of up to

P255 - Table 1: Results of a selection of parameters tested at 1 and 3 years of $\leq -25^{\circ}\text{C}$ storage in FFP-A-UVA

Parameters Tested N = 34 Average \pm Standard Deviation	FFP-A-UVA -1 year: T1	FFP-A-UVA -3 years: T3	% Recovery Significant (S) or non-significant (NS) difference*: T3/T1
PT (%)	95 \pm 9	88 \pm 9	93 \pm 3 / S
APTT (ratio)	0.97 \pm 0.09	0.97 \pm 0.09	100 \pm 3 / NS
Thrombin generation ETP (%)	93.43 \pm 30.56	93.59 \pm 29.50	101 \pm 9 / NS
Fibrinogen (g/L)	2.45 \pm 0.44	2.64 \pm 0.46	108 \pm 4 / S
Factor II (IU/mL)	0.85 \pm 0.11	0.83 \pm 0.10	98 \pm 6 / S
Factor V (IU/mL)	0.88 \pm 0.16	0.87 \pm 0.15	99 \pm 6 / NS
Factor VII (IU/mL)	0.91 \pm 0.24	0.83 \pm 0.20	91 \pm 4 / S
Factor VIII:C (IU/mL)	0.72 \pm 0.23	0.76 \pm 0.20	107 \pm 9 / S
Factor IX (IU/mL)	0.83 \pm 0.16	0.81 \pm 0.15	98 \pm 6 / S
Parameters Tested N = 34 Average \pm Standard Deviation	FFP-A-UVA -1 year: T1	FFP-A-UVA -3 years: T3	% Recovery Significant (S) or non-significant (NS) difference*: T3/T1
Factor X (IU/mL)	0.88 \pm 0.17	0.85 \pm 0.16	97 \pm 7 / S
Factor XI (IU/mL)	0.85 \pm 0.16	0.83 \pm 0.16	98 \pm 6 / S
Von Willebrand Factor VWF:RCo (%)	74 \pm 27	76 \pm 25	103 \pm 7 / NS
Protein S (% activity)	76 \pm 15	70 \pm 15	92 \pm 5 / S
TAT ($\mu\text{g/L}$)	2.2 \pm 0.3	2.5 \pm 0.4	117 \pm 15 / S
C3a ($\mu\text{g/L}$)	92.82 \pm 48.66	102.32 \pm 40.25	122 \pm 33 / NS
C5a ($\mu\text{g/L}$)	14.34 \pm 5.43	11.85 \pm 4.31	83 \pm 8 / S
Anti-SARS CoV-2 ELISA ratio	6.43 \pm 2.82	5.47 \pm 2.95	85 \pm 24 / S

* Student's t-test for paired samples. Significant difference if $p < 0.05$.

650 mL before A-UVA treatment) were selected from the frozen plasmas already stored ($\leq -25^{\circ}\text{C}$) at the EFS (Etablissement Français du Sang). 13 triplets were of group O and 21 of groups non-O. Fast freezing had been carried out after A-UVA treatment within 18 hours of collection. 30 plasma parameters as well as the Anti-SARS Cov-2 ELISA ratio were tested after 1 year (baseline, T1), 2 years (T2) and 3 years (T3).

Results: Table 1 presents the data for a selection of plasma parameters after 1 and 3 years of storage at $\leq -25^{\circ}\text{C}$. The National requirements stipulating that at least 70% of the units should have a FVIII:C concentration ≥ 0.5 IU/mL and a fibrinogen concentration ≥ 2.0 g/L were met at both periods, (FVIII:C 94% at T1 and 97% at T3, Fibrinogen 82% at T1 and 88% at T3). All other plasma factors were within physiological ranges. The ratio of anti-SARS-Cov-2 antibodies showed a moderate decrease.

Summary / Conclusions: The study on the in-vitro quality data of 34 units (triplets) of PFC-IA kept frozen at a temperature $\leq -25^{\circ}\text{C}$ for up to 3 years shows a stability of this plasma with regards of tested parameters. Compliance with the applicable French regulatory requirements as defined by law was confirmed at all periods (T1, T2 (not shown) and T3). The possibility to store plasma for an extended time is of interest in the context of convalescent plasma program.

P256 | Cryopreserved platelets—production, indications, and clinical use over the last 10 years in the Czech Republic

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Background: The short shelf-life of fresh platelets (PLTs) limits their efficient inventory management and availability during a massive transfusion protocol. Insufficient availability can be mitigated by building an inventory of cryopreserved platelets. Frozen PLTs have been produced in the Czech Republic since 2014. Frozen PLTs are currently used in 7 university hospitals with main trauma centers that cover most of the population of the Czech Republic, usually as part of a massive transfusion protocol for polytraumatic patients but also for other indications and their stockpile is part of the state crisis blood policy.

Aims: Describe the production, indications, and experience of clinical use of cryopreserved platelets over the last 10 years in the Czech Republic.

Methods: Platelets are frozen with 6% DMSO at -80°C before freezing. PLTs are concentrated and the supernatant. The shelf life of frozen PLTs is 2 years. Before use thawed PLTs are reconstituted in thawed plasma type AB or PAS and must be transfused for up to 6 hours. In the study, we monitored the number of units of frozen platelets produced and administered to patients with various diagnoses in 7 major trauma centers in the Czech Republic.

Results: A total of 2305 units of frozen platelets were produced. The majority of 1387 units were made from apheresis-collected PLTs, and 918 units of PLTs were obtained from buffy-coat. In Military University Hospital, reconstitution is carried out in AB plasma in other centers in PAS. Over a period of ten years, 1590 TD of thawed platelets were issued for clinical use. Of this number, 58 frozen units were delivered to smaller hospitals. In the main 7 trauma centers, thawed platelets were administered to 443 patients, plus 346 thawed units were issued to other hospitals. The main indication was the administration of thawed platelets to patients with polytrauma (538 units/114 patients), bleeding (278 units/156 patients), and thrombocytopenia (83 units/30 patients). 61 TD frozen units have expired.

Summary / Conclusions: It follows from our practical experience, that frozen platelets are safe and effective and the procedure of thawing and reconstitution of frozen platelets is very simple and fast, and it allows for having quality platelets products when dealing with massive bleedings and other urgent situations. Frozen PLTs are beneficial for civilian as well as military blood banks and all facilities which do not have a permanent, or sufficient stock of fresh platelets available.

P257 | Introduction of amotosalen/UVA pathogen-reduced pooled plasma in a Swedish blood center—impact on production efficiency, clinical availability and cost

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Background: Thawed fresh frozen plasma (FFP) is associated with high wastage rates due to unpredictable clinical demand. Pooled plasma may carry increased risk of transfusion-transmitted infection (TTI); however, treatment with the amotosalen/UVA (INTERCEPT®) pathogen reduction

(PR) system can mitigate this risk. PR-plasma can also be thawed quickly when needed and stored at $2-6^{\circ}\text{C}$ for 5 days. In 2021-2023 a mid-sized blood center in Sweden converted from thawed single-donor whole blood (WB)-derived FFP stored up to 14 days to pooled PR-plasma units that are thawed on demand and proactively protect against TTI.

Aims: To describe the impact of converting from thawed single-donor FFP to frozen-until-needed pooled PR-plasma on clinical availability, outdating rate, and plasma sales for fractionation.

Methods: WB collections, FFP and PR-plasma production, outdating rates, per-patient use, and cost data were exported retrospectively from blood center records in Borås, a city of approximately 114,000 in western Sweden. Descriptive statistics were calculated for 2 periods: 2018-2020, only conventional FFP (P1) and 2021-2023, conversion to pooled PR-plasma (P2). PR-plasma was introduced in March 2021; the conversion to thawed-on-demand PR-plasma was finalized in October 2022. To produce PR-plasma, a pool of 5 thawed ABO identical FFP units (mean volume 260 mL) is pathogen reduced with the INTERCEPT® system before redistribution into 6 PR-plasma units of 200 mL and refrozen. The shelf life of thawed PR-plasma was reduced to 7 days (local variance).

Results: A total of 5183 WB-derived FFP units were produced during the 6-year study period. A total of 2304 PR-plasma were produced in P2. A total of 1,970 were transfused in P1 (100%). In P2, 710 FFP (37.7%) and 1175 PR-Plasma (62.3%) were transfused, respectively. PR-plasma transfusions increased from 13.6% in 2021 to 96.9% in 2023. The number of patients transfused with plasma increased 19% in P2 compared to P1 but the mean number of units per patient declined from 2.4 to 1.8. Thawing time decreased from ~ 17 min for FFP to ~ 7 min for PR-plasma. Despite shorter post-thaw shelf life of PR-plasma, outdating declined 48% in 2023 vs thawed FFP in 2018. The number of FFP units sold for fractionation increased 9% in 2023 vs 2018 while the rate of WB donations was approximately the same.

Summary / Conclusions: Implementation of pooled PR-plasma stored frozen until needed required some additional labor (1 day/week of a technician's time) but contributed to reduced waste despite a shorter shelf life, improved production efficiencies, and increased revenue from plasma sales for fractionation. Efficiencies were attained through pooling of 5 WB-derived FFP units to produce 6 pooled PR-plasma units. Shorter thawing time of PR-plasma with standardized volume of 200 mL is of practical importance for emergency preparedness. The safety of PR-plasma is well documented in the literature; future studies may investigate the clinical advantages of less heterogeneous pooled plasma, e.g., reduced transfusion reactions.

P257 - Table 1

	P1 (2018-2020) Mean (range)	P2 (2021-2023) Mean (range)	Total (2018-2023)	Change 2018 vs 2023
WB collections	8.892 (8.680-9.041)	8.853 (8.363-9.175)	53,235	+0.7%
FFP units produced	921 (770-1060)	807 (752-893)	5183	-26.8%
PR-Plasma units produced	-	2304 (282-1074)	2304	-
FFP / PR-Plasma units transfused	657 (604-741)	628 (593-675)	3855	-16.7%
FFP / PR-Plasma units outdated	233 (201-253)	147 (109-183)	1140	-48.0%
FFP units sold for fractionation	7401 (6946-7923)	7552 (6978-8042)	44,859	+9.0%

P258 | Oxygen saturation levels of red cell concentrates awaiting clinical issue, across 33 days of storage

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Background: Oxidative stress has been proposed to drive the storage lesion of red cell concentrates (RCC) for transfusion. A non-invasive (visible reflectance spectroscopy) 'Oximetry' method of measuring fractional oxygen saturation (%SaO₂) of RCC was recently introduced by Yoshida *et al*, Transfusion (2022). This included a large dataset describing the range of 22-92% SaO₂ in units post-manufacture, and a proposed model for its increase during storage based on a study of 6 RCC pools.

Aims: To measure the %SaO₂ levels of RCC in SAGM, produced for clinical use by NHS Blood and Transplant (NHSBT), and to investigate factors contributing to variation.

Methods: Using an oximeter optimised to perform measurements on blood bags, triplicate readings were made of RCC at days 1 or 2 (*n* = 354) and additionally across the storage duration (*n* = 255) to day 33. An intraclass correlation coefficient (ICC) was calculated to assess the reproducibility of these repeat readings. Data on pack type and component volume were also collected, and associations of these variables with %SaO₂ on days 1-2 were assessed using multivariable regression analysis (MRA). Changes in SaO₂% over time (up to 33 days) were modelled separately, with various polynomial forms considered for the relationship with component age. A second set of RCC (*n* = 32) supplied for research and tested by oximeter were concurrently sampled for direct blood gas analysis to assess agreement between the two methods.

Results: Post-production %SaO₂ in NHSBT components as measured by oximetry, ranged from 30% to 92%. Reproducibility of oximetry readings was high (ICC = 0.973), suggesting that a single measurement may be sufficient in an operational context. In regression analysis, higher component volume was found to be significantly associated with increased %SaO₂ on days 1-2, but there was no association with pack type. However, neither pack type nor volume was associated with %SaO₂ during longer-term storage. Over the longer term, mean SaO₂% first rose and then levelled off at around 10 days, and the optimal model fit was achieved with a square root transformation applied to component age. It was noted that the regression curve resulting from the square root transformation closely mirrored the predictions of Yoshida *et al*, for components tested across the storage duration. Finally, a median absolute difference of 1.8 percentage points was found between %SaO₂ measurements from the blood gas analyser (single reading) and the oximeter (mean of three readings).

Summary / Conclusions: This study confirms earlier observations of the range in %SaO₂ of RCC in SAGM shortly after their production, whilst measurements taken from stock over the entire storage period support the validity of a published model of the evolution in %SaO₂ of RCC. Additional studies are required to further understand the

impact of SaO₂ variations on the overall quality of stored red blood cell components.

P259 | Preliminary study evaluating spray dried UK plasma in a blood bag

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Background: Most military deaths occur within 60 minutes due to catastrophic injury, head injury or major haemorrhage (MH). Early administration of plasma in addition to red cells, reduces mortality from MH. For plasma, this is a logistical challenge due to freeze-thawing/cold chain requirements in austere environments. Dried plasma provides a potential solution; however, commercially available dried plasma products have limitations; primarily glass bottle storage containers and global supply issues. Velico Medical have developed the FrontlineODP™ drying system, to allow blood establishments to produce a dried plasma component in a blood bag from a single unit of plasma. NHS Blood and Transplant (NHSBT) in collaboration with UK Ministry of Defence and Velico Medical aim to assess the suitability of the system for production of a UK-derived dried plasma component.

Aims: This study, the first to produce preliminary coagulation and pH data on UK plasma dried using the FrontlineODP System, aims to confirm the suitability of this system for use in the UK. Changes in pH which occur during spray drying have a detrimental impact on specific coagulation factors; therefore, additional pH data on UK fresh frozen plasma (FFP) was generated to better understand the pH of plasma, along with the impact of the testing / sample preparation method.

Methods: Six units of whole blood derived CPD-anticoagulated plasma from UK donors were sampled and frozen within 27 h of venepuncture. Units were spray dried using the Velico FrontlineODP™ system. After a few weeks of 4°C storage, units were rehydrated with 200 ml of sterile water and sampled. Samples were taken pre and post drying and these were frozen prior to testing. Samples were tested for pH by Velico Medical (Toledo manual) and NHSBT (ABL90 blood gas analyser 37°C and 22°C), and assessed for APTT, PT, fibrinogen (Clauss), FV, FVII, FVIII, FXI, Protein S Activity, von Willebrand factor (vWF) (activity and antigen) by NHSBT (ACLTOP550 Coagulometer). A further six units of UK FFP were thawed and tested for pH (ABL90 blood gas analyser 37°C and 22°C); immediately and following 1 and 2 h storage in a: blood bag, sample tube and following an additional freeze and thaw in a sample tube.

Results: All dried plasma units reconstituted within 5-8 min. The starting plasma mean pH, measured at NHSBT and Velico, was 7.30 and

7.34, respectively. The pH of the subsequent dried plasma was 6.65 (NHSBT data only). All coagulation parameters were within $\pm 20\%$ pre to post drying except APTT (24%), FVIII (-31%) and vWF activity (-40%). vWF antigen decreased by only 5.1%. Further data on pH measurement showed that UK FFP mean pH varied depending on the sample preparation and analysis method; from 7.05 (fresh from bag, 37°C) to 7.31 (frozen/thawed aliquoted sample, 37°C) and 7.24 (fresh from bag, 22°C) to 7.52 (frozen/thawed aliquoted sample, 22°C). pH remained stable in the blood bag over 2 h post thaw (7.05 to 7.07, 37°C).

Summary / Conclusions: Initial data from UK dried plasma was comparable to previously published data generated with US plasma produced using FrontlineODP System and suggests that it may be suitable for clinical use in the UK. Further in vitro and in vivo evaluation of dried plasma is intended by NHSBT, and necessary to fully evaluate the suitability of the FrontlineODP System for clinical use.

P260 | Rapid thawing of FFP with a 45°C dry tempering cycle maintains critical coagulation parameters

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Background: Fresh frozen plasma (FFP), stored below -20°C, must be readily available for cases of massive hemorrhage and complex consumptive coagulopathy. Water baths are widely used to thaw FFP, but the specific heat capacity of water (4.186 J/g°C) makes preheating a time-consuming procedure. Microwave systems are faster, but anxieties persist about focal overheating and denaturation of plasma proteins. A new dry tempering system, the FP-4000 (Hokuyo Denki, Kitamoto, Japan), includes an EXPRESS mode (FP-4000 EXPRESS) that heats FFP by holding the bag between two aluminum panels at 45°C until the product's surface temperature reaches 25°C.

Aims: To validate if FP-4000 EXPRESS heating can quickly thaw FFP without changing its quality, we evaluated thawing times, temperature changes on and in FFP bags, and coagulation parameters of the thawed FFP.

Methods: With IRB approval, FFP obtained from the Japanese Red Cross was pooled and distributed into 120, 240, and 480 mL bags that were stored for about 2 weeks at -30°C and then heated by FP-4000 EXPRESS. Thawing times, bag surface temperatures at the end of thawing, and clotting activities of thawed FFP were examined. In addition, internal bag temperatures were measured using a special thermometer placed in some 120 and 480 mL bags.

Results: Thawing durations of FFP heated by FP-4000 EXPRESS at 45°C were 9'43" \pm 0'51", 15'55" \pm 1'13", and 25'34" \pm 01'01" in

120 mL (N = 8), 240 mL (N = 10), and 480 mL (N = 8) bags, respectively. These times were significantly shorter than those of the FP-4000 normal mode, which heats the bags at 37°C (12'25" \pm 1'30", 21'17" \pm 01'51", 30'50" \pm 02'12", N = 6-10). Mean surface temperatures were 24.1°C, 27.1°C, and 20.7°C on 120, 240, and 480 mL bags at the end of heating in EXPRESS mode. No bags were damaged. Inside temperatures gradually and consistently rose to 26°C and 29°C, respectively, in two 120 mL and two 480 mL bags. After thawing with FP-4000 EXPRESS, international normalized ratios of prothrombin time were 0.921 \pm 0.028, 0.919 \pm 0.024, and 0.930 \pm 0.026, in 120, 240, and 480 mL bags, respectively. Activated partial thromboplastin times, fibrinogen concentrations, and activities of factors II, V, VII, VIII, IX, and X were similarly well-preserved in bags of all volumes after thawing with FP-4000 EXPRESS.

Summary / Conclusions: FP-4000 EXPRESS thawing of FFP can preserve its therapeutic efficacy and save precious minutes when transfusion is urgently needed.

P261 | The impact of blood and fluid warmer use on haemolysis and platelet function in cold stored whole blood

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Background: Blood and fluid warmers (BFW) are used to warm and rapidly transfuse crystalloids and blood products. BFW are mostly used for warming and administration cold stored red cells. There is an increasing interest in transfusing whole blood for patients with life-threatening bleeding. The effect of warming cold stored whole blood using BFW is not well known. The warming procedure might affect platelet count and function and coagulation proteins resulting in decreased haemostatic activity.

Aims: To investigate the impact of blood and fluid warmer use on haemolysis and haemostatic potential of cold stored whole blood.

Methods: A total of thirty (30) whole blood units (500 \pm 50 mL) were leukoreduced using the Imuflex WB-SP system (Terumo BCT) and stored for 21 days at 2-6°C before warming. Whole blood was warmed using Fluidio[®] Airguard system through Trauma Plus Set (The Surgical Company, NL) at either lowest (30 mL/min) or highest (900 mL/min) flow rate and a fixed temperature setting of 39°C. NaCl (0.9%) was used to prime and rinse the set. Blood samples were collected before and immediately after infusion. Samples were tested for haemolysis, platelet count, PT, APTT and thromboelastographic (TEG) parameters.

Results: Warming of cold stored whole blood did not result in increased haemolysis. Mean haemolysis before and after warming was 0.07%, with no differences between the different flow conditions. Warming resulted in a decrease in platelet count which was more pronounced at lowest flow rate (24% loss) as compared to highest flow

rate (11% loss). PT and APTT were slightly increased (<10%) after warming which was most likely due to the dilution of the WB by the priming and rinsing of the disposable. There were only small differences in thromboelastographic parameters before and after warming (Table 1).

P261 - Table 1. TEG parameters after warming

Flow rate (mL/min)	Values after warming (% of before)			
	R	K	Angle	MA
30	84 ± 5.6	104 ± 19	116 ± 23	91 ± 4.0
900	89 ± 5.7	96 ± 18	111 ± 14	95 ± 7.2

Summary / Conclusions: Warming of cold stored whole blood units, using Fluido® AirGuard System, either at lowest (30 mL/min) or at highest flow rate (900 mL/min), did not result in increased haemolysis. Although platelet count decreased after warming, this did not result in a decline of overall whole blood haemostatic potential.

P262 | Descriptive study of the use of cryopreserved platelet concentrates in the hospital setting

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Background: Cryopreserved platelet concentrates (CPCs) are considered a therapeutic alternative to platelet concentrates (PCs), especially in situations with thrombocytopenia accompanied by bleeding. The effectiveness of platelet transfusions are typically evaluated by the post-transfusion corrected count increment (CCI) which is lower with CPCs. Blood and Tissues Bank of Aragón (Spain) has supplied CPCs since 2016, mainly to remote hospitals. PCs are produced from 4 or 5 IPU(Interim Platelet Unit) (Reveos® Terumo BCT) in platelet additive solution, treated with pathogen reduction technology (Mirasol®) Terumo BCT, and cryopreserved using the Valeri method with modifications. A shelf life of one year is assigned.

Aims: The aim of this study is to find out in which clinical indications this type of PCs is being used and how its efficacy is evaluated.

Methods: To this end, a questionnaire with two parts was developed. Part one (7 questions) for the clinicians: age, sex, and ABO-Rh group of the patient, underlying pathology, reason for transfusion, adverse reactions, and verification of efficacy. Part two (3 questions) for the blood establishment: ABO Rh group, estimated count of platelet and remaining shelf life of CPC when it was transfused.

Results: A total of 97 CPCs were transfused to 72 patients in 7 hospitals in 2023. The CPCs had an estimated count of 357×10^9

($412-301 \times 10^9$) platelets and volume 200 mL. The average remaining shelf life was 141 days (288-1 days). The mean age of the patients was 65.4 (99-21), 45 were men (62.5%) and 27 women. The average platelet count before transfusion was $37 \times 10^9/L$ (259-1) $\times 10^9/L$. The transfusions were identical ABO in 47% of the cases. The petitioning service accounted for 90.6% of the transfusions. The most frequent requests for this type of PCs were from Haematology (36%), followed by Emergency (18%), cardiac surgery (10%), intensive care in 7%, surgery (7%) and others (22%). In 66 questionnaires (68% of the total) that were answered by the hospitals completely, the most frequent reason for transfusion was thrombocytopenia with haemorrhage in 37 transfusions (56%) but with the need for surgical intervention in 17 of them (46%), especially catheter insertion or punctures, followed by trauma and endoscopy. In 29 transfusions (44%) there was thrombocytopenia without bleeding but with the need for surgery in 65% of them. Efficacy was proven in 39% of prophylactic transfusions because the bleeding was avoided, 38% because the bleeding improved or stopped (in 4 of these cases with a post platelet count), 20% by a new count of platelets, and 3% not answered. Regarding underlying pathologies, 28 patients (39%) had a neoplasia, surgical cases (19%), sepsis for different reasons (5%), other pathologies (15%) and not information of 16 patients. Unless 10 ml of DMSO was transfused by pool, no adverse reactions were reported by the clinicians. The 85.4% of the transfusions were in remote hospital. The 5% of the CPCs were administrated because they were close to expiration. The patients received more than 1 PC in 26% of the cases (19 patients).

Summary / Conclusions: Cryopreserved platelet concentrates could be an alternative in remote hospital not only for therapeutic transfusions but also for prophylactic transfusion therapy. It is difficult to demonstrate efficacy of CPCs in this situations with a simple post-platelet count.

P263 | Leukoreduced blood components - the strategy for using a new method for blood quality control

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Background: Blood component safety is key in transfusion medicine and blood product manufacturing. Therefore, high standards during the whole process, from donor qualification to quality monitoring of the products, are required. Especially white blood cells (WBC) enumeration is important for process quality control. Until now, haematology analysers could not reach the lower limit of quantification required for this. In France, the residual white blood cells (rWBC) measurements in leukoreduced components are realized by flow cytometry.

Aims: The aim is to search for new alternative to the flow cytometry for enumeration of WBC in leukodepleted components in order to reduce sample manipulation, obtain an automatized system and standardize residual cell analysis. Blood Bank mode implementation on routine might allow the standardization analysis and reduce the time

spend in performing blood components CQ, as well as reduce the economic cost

Methods: Developments enabling cell counts in body fluids have, however, renewed interest in rWBC counting. An assessment of Sysmex XN hematology analyzers with Blood Bank mode to offer automated rWBC enumeration intended for use on blood components was performed in a Quality Control Laboratory in France. Performance characteristics were determined in leukodepleted red blood cell concentrate (RBCC) spiked with WBCs. Four levels of WBC (WBC/ μ L) were tested (6 times each) and results were compared with flow cytometry.

Results: Linearity studies from 1 to 11 rWBCs/ μ L (1; 3; 5; 12) showed good correlation between observed results using the system XN-1000 analyzers with Blood Bank mode and expected results by flow cytometry ($R^2 > 0.96$). Bias analysis between the rWBCs measurements using Blood Bank mode versus flow cytometry gave a maximum value of 19% for a quantitation of about 1 rWBCs/ μ L, (bias = 3; 10; 10% for WBC = 3; 5; 12 rWBCs/ μ L).

Summary / Conclusions: So far, the analyzer XN-1000 with Blood Bank mode seems to demonstrate acceptable performance characteristics for enumeration of rWBCs in RBCC; consequently, additional evaluations for enumeration rWBCs in leukodepleted components as RBCC and platelet concentrates are planned in our study.

P264 | Buffy coat pooled platelet concentrate—a boon or compromise in developing country?

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Background: Various platelet concentrates (PCs) are available such as random donor platelets (RDPs) derived from whole blood, prepared by platelet-rich plasma or buffy-coat (BC) method, and PCs derived from apheresis. Apheresis or single donor (SD) products are preferred over other PCs because they involve lesser donor exposure with a good yield of platelets. The cost and availability of an apheresis facility in the blood center are limiting factors in a developing country and buffy coat pool platelet concentrate (BCP-PCs) may be considered an alternative. This study compares BCP-PCs and SD-PCs for in vitro quality parameters and their in vivo outcomes.

Aims: To compare the quality of BCP-PCs and SD-PCs during storage and to analyze their clinical outcomes after transfusion in Hematology patients.

Methods: This study was a Non-Randomized Trial in which a single group variation of a crossover trial was done. This study was also registered in the Clinical Trial Registry of India. 35 consecutive hematology patients admitted to the Department of Medical Oncology-Hematology requiring more than one adult dose of platelet transfusion during their single admission were enrolled in this study. The order of transfusion was as per the decision of the patient and the

P264 - Table 1

Unit	Mean CCI 1 h	Mean PPR 1 h (%)	Mean CCI 24 h	Mean PPR 24 h (%)
SD-PCs	11274.29	26.27	5757.14	13.43
BCP- PCs	11960	28.10	7254.86	16.97

clinician. The in vitro study was done to compare the various hematological, biochemical and metabolic quality parameters of SD-PCs and BCP-PCs at day 1 & 5 both and in vivo analysis was done to evaluate the count increment, clinical effect of transfusion and analyze transfusion reaction if any. BCP-PC were prepared after pooling 5 ABO identical dry buffy coats prepared from Top & Bottom bags and pooling by train technique and adding a plasma unit in the same. The patient's diagnosis were as follows; 7 with B-ALL, 18 with AML, 3 with APML, 4 with CML, 1 with DLBCL, and 2 with T-ALL.

Results: The median gap between the transfusion of these two products was 72 h (ranging from 36 to 288 h). SD-PCs were transfused to 28 patients as a first transfusion while in 8 patients, BCP-PCs were transfused as a first transfusion. There was no significant difference in the quality parameters of SD-PCs and BCP-PCs except the glucose consumption which was found to be more in SD-PCs products than BCP-PCs. The mean yield of BCP-PCs was 3.4×10^{11} while of SD-PCs was 3.28×10^{11} while WBC contamination of BCP-PCs was 0.57×10^8 /unit while of SD-PCs was 0.52×10^8 /unit. Therapeutic transfusion of BCP-PCs was done in two patients and for SD-PCs were in one patient. Both responded with the transfusion. One allergic transfusion reaction occurred after BCP-PCs transfusion was managed by Inj. Avil which was resolved within half an hour. The following table shows the count increment and recovery after the transfusion.

Summary / Conclusions: There is no significant difference observed in the in vitro quality parameters of both products. The 24-hour CCI is more with BCP-PCs than with SDP. In case of unavailability of SD-PCs, BCP-PCs is a good alternative being similar in vitro and in vivo outcomes with approximately half the cost. Apheresis requires expensive equipment, trained personnel, stringent licensing, and additional space. On the other hand, buffy-coat pooling requires less technical support and easy licensure.

P265 | Red cell concentrates from lipemic whole blood - different levels of hemolysis, different donors

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Background: There are concerns about the in vitro quality of red cell concentrates (RCCs) from lipemic whole blood donations, due to possible negative effects of triglycerides on hemolysis of the red cells.

However, studies have shown only a weak correlation between lipemia and red cell hemolysis during storage, and the mechanism is still unclear.

Aims: To re-investigate hemolysis levels in stored RCCs, obtained from lipemic whole blood.

Methods: After visual judgement of plasmas for being fat, components were included in the study. Corresponding RCCs in SAGM were stored at 2–6°C, sampled and analyzed in week 1, 5 and 6. Based on week 6 hemolysis levels, the group was divided in quartiles. Because studies have also shown an association with donor BMI, and obesity is considered as a low-grade inflammatory state, we hypothesized that high hemolysis is associated with proinflammatory cytokines. Therefore, lipid profile, as well as RANTES (CCL5) and IL-8 were assessed. Quartiles of each parameter were analyzed with ANOVA (GraphPad Instat).

Results: Of the 101 units tested, 3 had a hemolysis at the end of storage of >0.8%, and averages ranged from $0.18 \pm 0.03\%$ in quartile 1 (Q1) to $0.59 \pm 0.15\%$ in Q4. There were no differences in volume, erythrocyte concentration, pH and glycolysis rates between the quartiles. Plasmas of Q4 contained the highest triglyceride levels (Q1: 4.4 ± 1.9 vs. Q4: 7.1 ± 2.3 mmol/L, $p < 0.001$) and lowest levels of HDL-C (Q1: 0.66 ± 0.22 vs. Q4: 0.47 ± 0.10 mmol/L, $p < 0.01$). Also RANTES levels were almost double in Q4 (Q1: 2477 ± 1264 vs. Q4: 4569 ± 1887 pg/mL, $p < 0.001$), but IL-8 levels were not significantly different. Mean age and BMI of donors was significantly higher in Q4 compared to Q1 ($p < 0.05$).

Summary / Conclusions: The association of triglycerides and RANTES with hemolysis was confirmed, but the majority of RCCs from lipemic whole blood had normal hemolysis levels. Measures to discard RCCs from lipemic whole blood may be adapted by distinguishing between low and high triglyceride levels of plasmas using color measurements. After implementation of non-DEHP collection systems, the study will be repeated.

P266 | Abstract withdrawn

P267 | Leukodepleted platelet concentrates from buffy-coat pooling in additive solution—quality evaluation

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Background: The Blood Bank and Tissues of Navarra (BSTN) processes 21,000 whole blood (WB) donations annually to produce leukodepleted red blood cells, plasma, and leukodepleted platelet concentrates (LDPCs). In September 2023, the center implemented a new platelet filter and validated the quality of LDPCs prepared from buffy coat (BC) pooling using platelet additive solution (PAS), as supernatant. An analysis of a week sample, during the months of December of 2023 and January of 2024, was conducted to assess the consistency in quality parameters and to provide evidence for the confidence on the new process.

P267 - Table 1. Results

	Group 1 December 2023	Group 2 January 2024	p-value	EDQM
N	56	54		
Volume (mL)	318 ± 16	318 ± 20	0.853	
Platelet count ($\times 10^{11}$ /unit)	3.47 ± 0.54	3.30 ± 0.55	0.102	>2.0
Leukocytes ($\times 10^6$ /unit)	0.08 ± 0.12	0.10 ± 0.21	0.720	<1.0

Aims: To evaluate the quality of leukodepleted platelet concentrates obtained from BC pooling suspended in PAS-E (SSP+), processed with the Thromboflex[®] TXP system, as support of the implementation for a new platelet filter, analyzing the quality control stability over time.

Methods: The evaluation was performed in accordance with European Directorate for the Quality of Medicines and Healthcare (EDQM, 2023) for LDPCs processing. WB units ($n = 550$, 450 mL) were collected using the Top & Bottom (T&B) configuration, with in-line red cell filter (LQT612U, Macopharma), in December 2023 and January 2024. WB was centrifuged on day 0 at 3,680 rpm for 18 min (Cryofuge 6000i, USA). Blood components were separated with a MacoPress Smarter (MPS), (Macopharma, France). After T&B processing, 5 BCs (ABO match, overnight resting) were pooled (Thromboflex[®] TXP filter system, TRV8006XU, Macopharma), with the multiple sterile connector, (Maconnect, Macopharma) and mixed with 250 mL of PAS-E (Macopharma). BC pools were centrifuged at 1300 rpm \times 6 min and separated using the MPS, to obtain LDPCs ($n = 110$), which were tested for platelet count and leukocyte count (Sysmex XN-10, Japan).

Results: There was no statistically significant variation, in the volume and leukocyte counts, in December 2023 compared to January 2024 ($p > 0.05$) and the implementation of platelet production, with the new filter, was successful and stable.

Summary / Conclusions: Thromboflex[®] TXP system is a method for producing LDPCs from BC pooling that is consistently compliant with the EDQM standards. The quality indicators have demonstrated low rates of variability, making whole blood-sourced, buffy coat pooled platelets, a reliable method to safeguard the blood availability of the blood processing center.

P268 | Analysis of the estimated volume of the platelet concentrates by an automated system

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Background: For the correct compliance of GMP standards in a Blood Establishment it is necessary to trace the different operations carried

P268 - Table 1. Statistical results

	Real Volume (n = 667)	TOMEs Volume (n = 667)	Difference	p
Median	363	398	36	<0.001
Mean	363.85	398.92	35.07	
Standard Deviation	14.78	24.39	21.50	
Max	437	518	160	
Min	316	226	-125	

out on the products and reflect accurately the characteristics of these products.

Aims: To check that the estimated value of volume of platelet concentrates (PC) elaborated in TACSI automated centrifuge (Terumo®) reflects the real value and, consequently, be able to confirm that the information system we used allow us to confirm the correct traceability of data.

Methods: In our Blood Establishment, TOMEs software (Terumo®) is used to register information related to the PC elaboration from 4 Buffy-coats (BCs). TOMEs is able to save information about connections in TSCII sterile connectors (Terumo®) between BCs, PAS and disposables, as well as information about the PC processing in TACSI (Terumo®) devices. These data are transmitted in a bidirectional manner between TACSI device and our IT system (Hematos®) through TOMEs software. Since June 2020, a total of 54,816 PCs were produced. Due to different problems, the final volume corresponding to 735 PCs was not automatically integrated in our IT system. These units were manually weighted (real weight), and volume was included in the IT system. Retrospectively we had been able to recover the TACSI calculated volume from TOMEs. We have compared the calculated volume to the real weight obtained using an electronic scale. Statistical study has been made to determine descriptive statistics (median, mean, maximum, minimum, and standard deviation) and compare means using t-test by means of SPSS software, a p value less than 0.05 was considered statistically significant.

Results: From the 735 PCs: 11 had a 0 mL TACSI-calculated volume; 23 units did not have data in our IT system (fake PCs to make informatic testing), and 34 units have been removed of this study because they had a wrong volume introduced. Finally, from 735 units selected, 667 were analysed. Results are shown in the attached tables.

Summary / Conclusions: Although calculated volumes are statistically different, they differ only in 35.07 mL, a volume that we think does not have a clinical importance. However, we consider, TOMEs may offer a good assistance to the operator and decrease transcription errors in data entry. In addition, TOMEs system allows a correct traceability of the data obtained and allows to work under GMP conditions.

P269 | Optimizing rapid plasma freezing to enhance efficiency in plasma production for medical use—a case study in The Republic of Belarus

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Background: Plasma production for fractionation within the National Blood Service is aimed at developing a national pharmaceutical production including medical products derived from blood plasma, and is an important part of the implementation of the State Program of Actions outlined by the Ministry of Health of Belarus for the substitution of imported medical products. At our State Institution, the production of plasma for fractionation is carried out in compliance with the Quality Management System (QMS) and Good Manufacturing Practice requirements. A key indicator that determines plasma quality and allows analyzing the efficiency of the plasma production process is the activity level of blood clotting factor VIII. It has been proven that the differences in production technology, ranging from plasma preparation methods and anticoagulant types to chosen temperature regimes of freezing, storage and transportation, have significant effect on the main plasma quality parameters. An important aspect in production of plasma that preserves its biological properties is the realization of a rapid freezing process to a temperature of -30°C and below within 60 min from the beginning of freezing.

Aims: The aim of this study is to determine the necessary plasma freezing time and prove that the proposed freezing process is appropriate to be effectively performed within the defined parameters. The expected results are to be achieved under reproducible conditions.

Methods: Following the validation plan outlined within the QMS, the different volumes of plasma were frozen using the CSF61 fast freezer in accordance with the Equipment Operation Manual. Graphs of temperature change inside the simulator bag as a function of freezing time were analyzed after the data was extracted from the CSF61 flash drive.

Results: Freezing process was performed for three plasma container loads of 0.300 L or less (n = 35) and three plasma container loads of 0.600 L (n = 22) over a 60-minute duration under strict process control, using a simulator bag equipped with a control sensor (control container). The evaluation of results revealed that the investigated parameters remained within acceptable limits and met the criteria defined by the validation plan. Quality control assessment of fresh frozen plasma of volume not exceeding 0.300 L (n = 9) and 0.600 L (n = 10) delivered results in terms of the parameter mean factor VIII activity: 89.5 (IU/100 mL), P(71.0<μ<108.0) = 0.95 and 103.3 (IU/100 mL), P(94.6<μ<112.0) = 0.95, respectively. Analyses of temperature change graphs within the simulator bag demonstrated that the time required to reach -30°C inside the control container did not exceed 20 minutes.

Summary / Conclusions: Based on the data gathered from this study, it is recommended that plasma manufacturers for fractionation consider reducing plasma freezing time by utilizing the CSF61 fast freezer. This adjustment is expected to increase equipment throughput, reduce energy costs of operation and optimize the process of plasma freezing to improve the efficiency of plasma production for fractionation.

P270 | Glucose concentration as a quality biomarker in platelet concentrates instrument and method dependent

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Background: In the 21st edition of the European guide to the preparation, use and quality assurance of blood components it is recommended to use pH or glucose concentration as a quality biomarker in platelet concentrates suspended in an additive solution. The rationale for this recommendation is the high buffering capacity in platelet additive solution that will keep the pH constant throughout the storage time. The glucose concentration, however, will decrease during the storage time and a level above the limit of detection is considered acceptable.

Aims: The aim of this study was to compare three different methods and instruments for glucose measurements in platelet concentrates at the end of storage.

Methods: A total of 45 platelet concentrates were included in the study and samples were collected on day eight when the units were outdated. Swirling was documented for all units. Samples were aspirated into a heparinized syringe for measurement in a blood gas instrument (ABL800, Radiometer). Furthermore, samples were collected in FCmix test tubes (Becton Dickinson) for measurement with a routine clinical chemistry instrument (Alinity, Abbott). Lastly glucose was analyzed using a point of care instrument (POC) (HemoCue). Lactate and pH were additionally measured with the blood gas instrument.

Results: The glucose concentration measured with the blood gas instrument and the chemistry instrument gave almost similar results with a median concentration of 0.95 mmol/L. The POC results were significantly different ($p < 0.001$) with a median concentration of 2.4 mmol/L, that is, higher values. In 14 (31%) of the platelet concentrates the glucose level was below the limit of detection when using the blood gas instrument. In 18 (40%) of the units the glucose level was below the limit of detection using the chemistry instrument. Using the POC-instrument all the glucose measurements were detectable. The lactate concentration was >15 mmol/L and the median pH was 7.1 and swirling was observed in all units.

Summary / Conclusions: pH has traditionally been used as a quality marker for platelet concentrates at the end of shelf life, but in concentrates suspended in additive solution glucose is considered as a preferred method. The results from this study comparing three different instruments and methods are not conclusive regarding what

instrument to choose. It is problematic that two methods had a high number of values lower than the detection limit and still acceptable swirling. Further studies are needed to evaluate the best method or the best methods of choice reflecting good quality at the end of storage.

P271 | Performance evaluation of a haematology analyzer on Leucodepleted Packed Red Cell (LDPRC) blood component for quality control assessment at the CBTS of Indonesian Red Cross

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Background: Evaluation of hematology analyzers is an essential step that must be undertaken prior to their operation in laboratories. This verification process encompasses various aspects, including stability, precision, linearity, and carry-over, which need to be assessed across the expected range of results. However, the specific standards and verification limits to be adhered to are ultimately determined by the discretion of the laboratory. Sysmex XN-350 (Low WBC Mode) is a next-generation automated hematology analyzer providing complete blood count (CBC) with 5-part white blood cells (WBC) differential counts

Aims: The purpose of this study was to evaluate the performance of the analyzer to verify blood component quality control assessment using Sysmex XN-350 Low WBC Mode (Sysmex Corporation, Kobe, Japan) automated system, based on the Central Blood Transfusion Services-Indonesian Red Cross established protocol. Furthermore, the evaluation results can verify its utility as a reference equipment analyzer for quality control assessment at other Indonesian Red Cross Blood Center

Methods: A comprehensive evaluation was conducted encompassing specimen stability, precision, linearity, carry-over, method comparison, and workflow to assess the performance of the Sysmex XN-350 (Low WBC Mode) hematology analyzer. This study utilized a total of 196 samples derived from Leucodepleted Packed Red Cell (LDPRC) blood components. Additionally, a subset of 40 EDTA samples were analyzed to compare between the Sysmex XN-350 (Low WBC Mode) with the Sysmex XN-1000 (Blood Bank Mode)

Results: The performance evaluation of the Sysmex XN-350 (Low WBC Mode) hematology analyzer yielded highly satisfactory results. The analyzer exhibited exceptional reproducibility and between-batch precision for key parameters such as white blood cells (WBC), red blood cells (RBC), hemoglobin (HGB), and hematocrit (HCT). Furthermore, it demonstrated good linearity and acceptable carry-over. In comparison to the Sysmex XN-1000 (Blood Bank Mode) analyzer, the correlation coefficients for WBC, RBC, HGB, and HCT ranged from 0.954 to 0.998, indicating a strong agreement between the two

instruments. Moreover, the workflow test results revealed no significant difference between the XN-350 (Low WBC Mode) and the XN-1000 (Blood Bank Mode) ($p = 0.480$), suggesting that both analyzers require a similar amount of time for blood component quality control assessment

Summary / Conclusions: In conclusion, the Sysmex XN-350 (Low WBC Mode) analyzer demonstrated excellent reproducibility and precision for key blood parameters. It also showed strong correlation with the Sysmex XN-1000 (Blood Bank Mode) analyzer and had similar workflow test results. These findings support the utility of the Sysmex XN-350 (Low WBC Mode) analyzer for an accurate and efficient blood component quality control in laboratory settings.

P272 | Effect of acidosis on conformational changes in erythrocytes

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Background: The physiological functioning of the organism is ensured by the balance of homeostasis, hemostasis, and the acid-base state of the blood, while the development of critical conditions is accompanied by a disturbance in oxygenation and quickly leads to shifts in this balance. The acid-base balance is the most sensitive to hypoxia, whose indicators are within a very narrow range of viability. When the oxygenation function of the lungs is impaired and hypoxia occurs, acidosis quickly develops, the negative effects of which include arterial vasodilation, decreased neuronal excitability, insulin resistance, and immune imbalance. Acidosis is a constant companion of hypoxia and reflects the degree of its severity and duration. Changes in the pH of stored red blood cells trigger a response from cells, including red blood cells, in the form of changes in the rate of biochemical and physical processes affecting specific cell function.

Aims: investigate the impact of acidosis on conformational changes in erythrocytes.

Methods: A suspension of erythrocytes from a healthy donor (RBCs suspension) was obtained. Seventy-five microliters of RBCs were added to 1 ml PBS with pH 7.4 and 6.4, which was divided into two parts. One of them was placed in solution No. 1, where pH = 7.4, which corresponds to physiological value, and the other - in solution No. 2, where pH = 6.4, which corresponds to deep acidosis, at which cell death occurs. Suspensions were kept in appropriate solutions at constant air temperature of +24°C. Two study points were determined: 1st point - exposure for 30 min and 2nd point - exposure for 7 h. After the appropriate exposure time, a monolayer of erythrocytes was prepared for study by AFM.

Results: After 30 min of incubation in solution No. 2 (pH = 6.4) there was a statistically significant change in the composition of erythrocytes, compared to the initial indicators and solution No. 1: the total number of discocytes decreased by 1.6 times, and echinocytes

increased by 8 times ($p < 0.0001$). Exposure of erythrocyte suspension for 8 h resulted in changes in the morphologic composition of cells in both groups. In the experiments the dynamics of changes of the values and, and were investigated, which characterize the roughness parameters of erythrocyte membranes and, in general, the properties of its nanosurface. Exposure to a more acidic environment with pH 6.4 for 8 h resulted in significant changes in membrane structure. It was recorded that when exposed to pH 6.4 for 7 h: increased by 1.5 times compared to the control data ($p < 0.001$), increased by 2 times ($p < 0.001$), by 5 times ($p < 0.0001$), by 3.3 times ($p < 0.001$)

Next, the Length of the cytoskeleton pores was calculated, which is the maximum distance between two cross-sectional points. The control values of Length under the conditions of this experiment had values of $0.13 \pm 0.07 \mu\text{m}$, and in solution No. 2 Length of pores increased by 1.5 times ($p < 0.05$). After 24 h in the solution No. 1 there is a tendency to increase the Length of pores. In solution No. 2, compared to the control sample, increased by 2 times ($p < 0.01$).

Summary / Conclusions: Critical acidosis simulated in vitro negatively affects all erythrocyte characteristics: morphological composition, heights, spatial periods, pore number and pore size. The results show that the stability of the second-order nanostructure is an indicator of cytoskeleton stability and reversibility of cell shape.

P273 | Unveiling platelet quality challenges—a comprehensive analysis of flocculation in apheresis-derived products since April 2023

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Background: Since April 2023, platelet concentrates from apheresis have increasingly exhibited flake-like flocculation. The occurrence and intensity of these flakes varied across different sections of the production line.

Aims: We investigated a possible relation between the end-product quality, measured as number of platelets, and characteristics that relate to the blood donation as well as the development of flakes formation across different production stages.

Methods: Conspicuous products were observed over a period of approximately eight months. The affected products were divided into three groups based on the level of flake formation (low = 1, medium = 2, high = 3). We measured thrombocyte cell concentration (Tc) in the affected products at following time points during the production: (a) in the evening upon delivery to the production site, (b) immediately before pathogen inactivation on the following day and (c) after pathogen inactivation in the final product.

In order to find a relevant difference in the process in relation to specification limits, we compared Tc of the end-products with flocculation with the end-products from the regular quality control using scatter plots. Blood donors whose products led to flocculation in 2023 were

compared with donors from an earlier observation period in 2019-2020 in which similar flocculation problems were detected. A simulation was applied to test the expected random pairing with donors who have already generated products with flakes in the earlier observation period. We also aimed to identify the donor and blood collection characteristics related to flocculation. Following characteristics were analyzed: blood donor age, number of donations during the observation period, blood collection duration, blood collection type and volume, blood pressure, weight, height, hematocrit, hemoglobin, platelet percentage, leukocyte percentage, pulse.

Results: The scatterplots show a clear reduction in end-product quality that increases with the degree of flocculation, but most products were still within the specification limits. Importantly, our simulation indicates a strong relationship between the donor and flake formation in the products.

Summary / Conclusions: Due to the observed reduction in quality, all products with flocculation grade 2 and 3 are subjected to additional quality control. The established reference to the blood donor and the type of collection indicates the direction in which further tests are to be investigated.

P274 | Evaluate platelet production from whole blood by the fully automated blood separation system

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Background: The leukocyte reduced (LR) and non-leukocyte reduced (NLR) sets for automated whole blood processing has been validated in Blood Transfusion Centre, Faculty of Medicine, Khon Kaen University, Thailand in 2016 and 2019, respectively. The protocol was implemented and started running until now. The machine can expect platelet yields index (PYI) from interim platelet (IPU), transfusable platelet concentrates (TPU), plasma volume of IPU/TPU, the volume of plasma product, and volume of buffy coat.

Aims: To assess the quality of platelet concentrates from whole blood collections by the fully automated blood separation system.

Methods: We reviewed on the data of platelet yields index (PYI) derived from each unit of transfusable platelet concentrates (TPU) and interim platelet (IPU) from 2021 to 2022 which prepared by the fully automated blood separation system.

Results: The protocol of non-leukocyte reduced (NLR) set was shown the average of PYI derived from transfusable platelet concentrates (TPU) 6.96×10^{10} cells/unit ($N = 2336$) and 8.10×10^{10} cells/unit ($N = 8660$) from interim platelet (IPU) whereas the average of PYI from IPU by the protocol of leukocyte reduced (LR) set was found 6.31×10^{10} cells/unit ($N = 5470$). The PYI less than the standard value (5.5×10^{10} cells/unit) were detected in 595 units (3.6%) from 16,466 blood donors. The white particulate matter, we found 287 units (1.7%) was the key reasons for such incidents, and 308 units (1.9%) have an unexplained the low PYI. Most of the PYI value passed the standard value to 96.4%.

Summary / Conclusions: This study revealed that, the fully automated blood separation system prepared both of TPU and IPU reached the recommended quality of Council of Europe (EU), American Association of Blood Bank (AABB), and National Blood Centre, Thai Red Cross Society.

P275 | Using xenon for red blood cells prolonged storage

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Background: Xenon, an inert gas commonly used in medicine, has been considered as a potential option for prolonged storage of packed red blood cells (pRBCs) under hypoxic conditions. By using inert gases, the storage process is expected to minimize the occurrence of structural and biochemical changes in pRBCs caused by oxidative processes, ultimately leading to a reduction in transfusion-related complications.

Aims: investigate the impact of Xenon on the characteristics of pRBCs during their storage period.

Methods: Leukodepleted pRBCs in air-tight bags containing CPD/SAGM solutions were obtained from Moscow blood transfusion centers. A single standard unit of pRBCs (large bag) had a volume of 450 ml. Each unit of pRBCs was divided into a small 30 ml bags. Xenon was pumped into 45 small bags, and another 45 small bags were stored without adding Xenon (control). Samples from pRBC were stored under normal conditions at $+4^{\circ}\text{C}$ and withdrawn on days 4, 8, 14, 22, 32, and 42. A wide range of methods comprising biochemical analysis, spectroscopy, atomic force microscopy and photography to detect the changes in pRBCs stored in the presence of Xe were employed. Atomic force microscope NTEGRA Prima (NT-MDT Spectrum Instruments, Moscow, Russia) was used to obtain morphology and cytoskeleton images, stiffness in different control days of storage. Unico 2800 digital spectrophotometer (United Products & Instruments, USA) was used for to determine the concentrations of hemoglobin derivatives.

Results: During prolonged storage it was shown that the presence of Xenon led to significant morphological changes in pRBCs, similar to those observed under standard storage conditions. These changes include irreversible cell transformations such as the formation of echinocytes, microspherocytes, and ghosts. Hemolysis during storage of cells with Xenon exceeded the permissible limit by six times. A specific effect of Xenon exposure was the production of deoxyhemoglobin in the cell suspension.

Summary / Conclusions: The main idea of this study was to use Xenon as a protector of red blood cells during storage. Unfortunately, our study did not show any apparent protective effects of Xenon. Although Xenon, as an inert gas, does not cause oxidative stress, changes in the gas composition caused by its addition may create situations that enhance oxidative processes. Perhaps the deciding factor

is the Xenon concentration. Therefore, it is important to conduct further research to determine the optimal Xenon concentration to improve the properties of stored red blood cells.

P276 | Routine use of digital microscope for residual white blood cells counting in a quality control lab

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Background: The ADAM rWBC HT is an automatic fluorescent microscope used for residual white blood cells (rWBC) counting in blood components.

Aims: This study has the objective to evaluate the advantages to use the ADAM rWBC HT compared to the use of cytometry (FACSVia from Becton Dickinson).

Methods: The ADAM rWBC HT is a digital microscope that counts rWBC stained by propidium iodide and detects its fluorescent expression. A mix of blood component and reagent containing propidium iodide and lysis solution (ratio 1:4 for platelet concentrates and red blood cell concentrates, ratio 1:1 for plasma) is deposited on a disposable slide and placed on a rack to be automatically transported to the fluorescent microscope that counts rWBC in a minute. It automatically focuses on the slide and counts the cells from 63 images, then averages out the counting results to increase the accuracy and reliability.

Results: The counting of one sample by cytometry took 108 s on average compared to 69 s with the ADAM rWBC HT. Daily in our lab, we measure 13 samples; the time saving is 16 min per day. Moreover, the start-up time for the cytometer takes 14 min compared to the ADAM that takes 2 min. The controls samples analysis is effective after 5 min for the ADAM compared to 8 min for the cytometer. Finally, there is also a money sparing with the use of ADAM because there are no multiple washing solutions; and one leucocount kit from Becton Dickinson (50 tests) costs same price as one kit (50 tests) for the ADAM.

Summary / Conclusions: The use of ADAM rWBC HT in our quality control lab allowed to save time in routine analysis of approximately 16 min daily compared to the use of cytometry without automatic loader.

P277 | Preparation of eye drops from autologous serum with assessment of effectiveness of eye drops in different eye surface pathologies—pilot project

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Background: A new concept in the field of ophthalmology Autologous serum is the production of eye drops from the patient's own blood.

Eye drops produced from autologous serum effectively alleviate the complaints of patients with dry eye syndrome who are allergic / resistant to commercial drugs, and promote healing after corneal transplantation. Autologous drops are an indispensable therapy for some patients. It is about innovative and safe production in a closed system. The prevalence of dry eye disease is increasing in developed countries.

Aims: The method of examination of transfusion patients, collection, processing and storage as well as the effect of monthly application of autologous serum in the form of 34 vials on the surface of the eye, to compare the symptoms before and after and in general the tolerability of the drops. To investigate the use of autologous serum (AS) eye drops in patients with ocular surface disorders who were refractory to conventional treatments

Methods: The pilot project was implemented at the Institute for Transfusion Medicine of the Federation of Bosnia and Herzegovina, in the Department for Collection and Production of Blood components. We included 10 patients referred by different ophthalmology institutions with different diagnoses. Ophthalmological findings and clinical history were reviewed. A database was created with all relevant patient data. Efficiency was assessed subjectively, and with individual ophthalmological tests. Drops were prepared as autologous serum with a 1:4 dilution of saline as a monthly therapy that included 34 vials and additional vials for microbiological and serological control.

Results: A pilot project on 10 patients included patients with different diagnoses: blepharitis, keratopathy, pseudophakia, Sjogren's sy, glaucoma, erosio cornea, sy dry eye... After the monthly application of the drops, the subjective evaluation of the application was examined and it was noted: reduction of eye pain, increased moisture, better tolerability of the serum compared to commercial drops, feeling of smoothness, the eye looks freshly rested and lubricated. Ophthalmologists report: positive experiences that manifested themselves in the reduction of erosions, epithelization of ulcers, the possibility of final extraction of the therapeutic lens, healing.

Summary / Conclusions: Preparation eye drops from autologous serum on transfusion provides a number of advantages, excludes bacterial contamination, provides a simple and safe system that is CE certified, ensures equal amount of drops, microbiological, mycological and serological testing, sterility and better tolerability. Autologous serum eye drops address deficiencies with their tear-like biochemical features and supply nutritional components, provide nutritional and growth factors that are necessary to maintain epithelial recovery processes and bactericidal components that reduce the risk of contamination and infection.

P278 | Microbiological contamination of donor blood processed before despatch

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Background: Uganda Blood Transfusion Service (UBTS) is mandated to supply the Ugandan population with sufficient and secure blood.

The main supply of blood and blood components for patients and individuals needing transfusions in UBTS is blood donation by unpaid individuals. It has never been taken seriously to consider the risk of microbiological contamination and the prevention of potential contamination during the collection and processing of units in the blood bank and from the blood donor, which puts the recipient of donor blood products at risk. Although sepsis caused by bacterial blood contamination is a relatively uncommon occurrence in transfusion medicine, transfusion-associated sepsis episodes can have disastrous consequences and should be avoided. In most transfusion-related sepsis cases, determining the source of contamination can be difficult. However, by considering isolated species of organisms, information about likely transmission pathways can be learned. Potential sources include donor bacteraemia, intrinsic contamination of the phlebotomy site, contamination of blood containers or needles, and equipment contamination.

Aims: (1) To detect the presence of microbiological contamination of donor blood products notably, whole blood, packed red blood cell concentrates, plasma, platelets before laboratory dispatch in UBTS. (2) To confirm laboratory isolation and determine antimicrobial susceptibility patterns of the microbiological species responsible for contamination in the donor blood. (3) To determine the knowledge, attitudes and practices of laboratory and field staff that handle donor blood units about the risks of possible bacterial contaminations of the blood units.

Methods: This research was cross-sectional study. Following blood collection, blood culture samples were removed from the satellite bags, triple packaged, and returned for examination to Kampala Hospital Microbiology Department within three days of collection. The sample were chosen at random. The stratified random sampling strategy was used to select blood units showing signs of contamination. Using a sterile graduated syringe and needle, 3–8 mls of blood for the study samples were collected aseptically from blood unit bags into Blood culture bottles, triple packaged and transported back to the Microbiology Department for analysis within three days of collection. A sum of 78 samples was collected by field teams, donor rooms, and collection centres.

Results: The cultured samples were 78 in number, of which 75 samples showed no growth of bacteria or fungal contamination. Three (3) samples showed microbial growth. The microbial organisms that were isolated and identified were *Staphylococcus epidermidis* (2 counts) and *Staphylococcus saprophyticus* (1 count). Ten (10) antimicrobials were set against the microbial isolated, only four (4) were sensitive, five (5) were resistant and Only one (1) antimicrobial showed intermediate susceptibility.

Summary / Conclusions: Blood is not totally free from contamination especially bacterial contamination, even confirming from the personnel practices. Platelets that are prone to contamination due to their storage conditions. The isolation of *s. saprophyticus* in donor blood products is an indication of presence of pathogenic micro bacteria among donor blood products. The results of the antimicrobial susceptibility tests (AST) showed that there is 66.66% resistance of microorganism's isolated in the donor blood.

P279 | Reducing of the number of Buffy Coats (BC) in the automated pooling system used in the preparation of double-dose pathogen reduced platelet units

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Background: In our blood center, Terumo Automated Centrifuge & Separator Integration (TACSI[®] System, Terumo BCT Lakewood, USA) technology is used for pooled platelet production. Seven Interim platelets units (IPUs) used to be pooled and connected to Intercept Blood System (IBS, Cerus Corporation, Concord, USA) to produce one double-dose of pathogen reduced platelets. As the recent EU directive MDR requires concordance with Instructions for use (IFU) from the manufacturer, we aimed to reduce the number of buffy coats (BC) from seven to six.

Aims: Reduce the number of buffy coats per platelet pool without decreasing the platelet content below approved numbers.

Methods: Whole blood (450 mL), collected on day 0 into citrate-phosphate-dextrose anticoagulant using a quintuple top and bottom bag (LQT 614E, Macopharma, Mouvoux, Franc) 1 092 blood donors centrifuged in Hettich Roto Silenta day after collection and the three components (plasma, red blood cells and buffy coat) were separated using a semiautomated press, Macopress Smart (Macopharma, Delcon, Italy) Pooling: 6 BC by TSCD (Terumo BCT) sterile docking device using TACSI[™] PLkit (93000, Terumo BCT) with and platelet additive solution, PAS (280 mL SSP+) The BC:s pools were centrifuged and leucoreduced simultaneously in a TACSI[™] automatic device (Terumo BCT). The final BC-PCs were connected to INTERCEPT[®] blood system (IBS) dual storage sets, INT 2503B (Cerus, Concord, USA). After addition of amotosalen to the six BC-PC were treated with a controlled dose of UVA using INT100 (3.9 J/cm²). After photochemical treatment, the PCs were transferred into CAD system for 4–16 h and then into the final container. The pool is then divided into two identical platelet units for transfusion.

Results:

P279 - Table 1. Results

Platelet units for transfusion	Net weight (g)	WBC < 1×10^6	Platelet $\geq 2 \times 10^{11}$
182 (n)	197 (184-211)	0,007 (0,000-0,011)	2.27 (1.79-2.69)
Approved (%)	100%	100%	95%

Summary / Conclusions: The use of TACSI[™] platelet pooling system with 6 BC and 280 ml SSP+ meets the quality requirements according to EDQM's Guide to the preparation, use and quality assurance of blood components. Conclusion are that we can reduce the number of donations necessary per unit, whilst adhering to the manufacturer's Instructions for use (IFU).

P280 | Evaluation and quality control of labile blood products at the Constantine University Hospital Blood Transfusion Center

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Background: The quality of labile blood products is a requirement for transfusion safety, and conditions transfusion yield and effectiveness.

Aims: To assess and control the quality of labile blood products, and check their compliance with regulatory requirements. Evaluate the effectiveness of the preparation protocol in place and propose appropriate corrective actions.

Methods: We used stripping sampling, an automated blood count machine and a precision balance to determine the main characteristics of labile blood products (Red Blood Cell Concentrates, Standard Platelet Concentrates, and Apheresis Platelet Concentrates) prepared at Constantine University Hospital Blood Transfusion Center, Data were collected and processed using Excel software.

Results: 96.36% of Red Blood Cell Concentrates were within normal limits, with an average of 318 ml. In terms of Hb levels, 88.68% of bags were within normal limits, with an average of 58.04 g. For hematocrit, 76.36% of Red Blood Cells Concentrates were within norms, with an average of 60.4%. For platelets, the volume of 80% of Standard Platelet Concentrates is within the norm with an average of 56 ml, while all Apheresis Platelet Concentrates have a volume within the norm. The average platelet richness of Standard Platelet Concentrates and Apheresis Platelet Concentrates is 0.61×10^{11} and 3.98×10^{11} respectively, with a compliance percentage of 80% for Standard Platelet Concentrates and 100% for Apheresis Platelet Concentrates. 89.10% of Platelet Concentrates have a residual WBC rate within the norms

Summary / Conclusions: Despite the satisfactory compliance rate of Labile Blood Products prepared at the Constantine University Hospital Blood Transfusion Center, regular and periodic quality control is essential to further improve their quality.

P281 | Is donors age involved in the modulation of molecules present in single donor apheresis platelet concentrate and involved in severe adverse reaction—a multiparametric analysis?

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Background: Blood transfusion is a life-saving procedure in which whole blood, or blood components are provided to a patient. Platelet concentrates (PC) may sometimes induce adverse reactions that are occasionally severe. PC comprise active biomolecules such as

cytokines and lipid mediators. Platelet storage lesions are structural and biochemical changes in PC and vary in collection and processing conditions.

Aims: Our objectives are 1/ to discriminates bioactives molecules whose the expression is modulated through physiological age of the PC's donors; 2/ to evaluate this modulation in terms of transfusion adverse reaction.

Methods: The PC left over were analysed by mass spectrometry (for lysophospholipids, sphingomyelin 1 phosphate) and ELISA (for sCD40L, secreted p-selectin, HMGB1 and PF4), and classified with the donor's age in "aged donors" (60-70 years old) and "young donors" (18-30 years old), and the impact on severe reaction after transfusion (with/out).

Results: Platelet Factor 4 expression increased during physiological age, and correlated with physiological age of the donor and the storage time. However, S1P expression decreased during physiological age whereas the expression of HMGB1, sCD40L are not modulated.

Summary / Conclusions: Our multiparametric study revealed some bioactive molecules modulation during the ageing. These molecules have an impact in platelet concentrate transfusion, and will be monitored in the future before transfusion.

P282 | Comparison of Factor VIII Levels in plasma components before freezing by centrifugation and filtration plasmapheresis methods

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Background: Plasma blood components can be produced from whole blood by centrifugation or sedimentation and plasmapheresis. Plasmapheresis is a method of taking blood component that are needed, and the rest components that are no needed are returned to donor's body. In plasmapheresis there are two methods of apheresis to produce blood plasma products, centrifugation and filtration methods. The usual parameters for examination of plasma components in the field of blood transfusion are factor VIII, in order patients who have disorder of blood clotting factors, such as hemophilia.

Aims: To determine the value of factor VIII in plasma components before freezing using method of centrifugation and filtration plasmapheresis.

Methods: Plasmapheresis sampling of 20 samples, with a target volume of 600 mL in each plasmapheresis method. The process of plasmapheresis is divided into two, centrifugation process and the filtration process. Centrifugation process is performed rotation, and separation based on the cell density. The filtration process is the separation of blood cells based on the cell size, with filtration

pore size of 0.65 microns, so only plasma can pass through the semipermeable membrane filter. The examination sample was taken as much as 3 mL to be checked for factor VIII before freezing with semi-automated coagulation analyzer, in a period of less than 1 h. The acceptance standard for factor VIII is ≥ 0.7 IU/mL.

Results: The results the examination obtained are plasmapheresis by centrifugation method with a range of factor VIII levels 0.83 IU/mL, with an average of 0.95 IU/mL. Plasmapheresis with filtration method with factor VIII content range 0.92–1.03 IU/mL, with an average of 0.96 IU/mL. The median for both plasmapheresis method was 0.98 IU/mL. 20 samples of plasmapheresis centrifugation method and 20 samples of plasmapheresis filtration in accordance with the standard of acceptability ≥ 0.7 IU/mL.

Summary / Conclusions: Based on the results of examination, all sample of plasmapheresis centrifugation and filtration methods in accordance with acceptable standard ≥ 0.7 IU/mL, with the median of both plasmapheresis method is 0.98 IU/mL, and the average level of factor VIII by centrifugation method is 0.95 IU/mL, and the average level of factor VIII by filtration method is 0.96 IU/mL.

P283 | Abstract withdrawn

P284 | Increased potassium release in X-ray irradiated deglycerolised red blood cells

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Background: Transfusion-associated graft-versus-host disease (TA-GVHD) is a rare, commonly fatal complication of transfusion, which is preventable by irradiation of blood components. The guidelines for the prevention of TA-GVHD vary depending on the jurisdiction, with a lack of consensus as to whether irradiation of deglycerolised red blood cells (RBCs) is necessary for those patients at risk. Irradiation of RBCs exacerbates the storage lesion, in particular the release of extracellular potassium (K^+), and often leads to a reduced component shelf-life. The current shelf-life in Australia for deglycerolised RBCs stored in saline-adenosine-glucose-mannitol (SAGM) additive solution is 24 h. X-ray irradiation for all blood components, including deglycerolised RBCs, has recently been introduced at our facilities.

Aims: The aim of this study was to evaluate the quality of x-ray irradiated deglycerolised RBCs resuspended in SAGM additive solution and determine a suitable shelf-life.

Methods: Twenty-one pairs of ABO/RhD matched leukodepleted RBCs were pooled and split into two equivalent components. RBCs were glycerolised with 40% glycerol using an ACP-215 cell washer and frozen at -80°C . Units were thawed in a 42°C water bath and

deglycerolised using an ACP-215 cell washer, then resuspended in SAGM. One of each pair was x-ray irradiated immediately after deglycerolisation, whilst the other was untreated (control). Post-deglycerolisation RBCs were stored at $2-6^\circ\text{C}$. RBCs were sampled pre-freeze and immediately after irradiation and then at 24-, 48- and 72-h post-irradiation. Samples were tested for quality indicators including RBC indices, metabolic and biochemical parameters. Irradiated and control RBCs were compared over the storage period using repeated measures ANOVA; $p < 0.05$ was considered significant.

Results: Extracellular K^+ was significantly higher in the x-ray irradiated RBCs compared to control RBCs at 24-hours post-irradiation (6.26 ± 0.95 and 3.21 ± 1.01 mmol/unit respectively; $p < 0.0001$) and was 1.7-fold higher by 72-h post-irradiation. Haemolysis was also higher in x-ray irradiated deglycerolised RBCs ($0.35 \pm 0.14\%$) compared to control RBCs ($0.32 \pm 0.05\%$) after 24 h of storage (current expiry), and rapidly increased to $0.52 \pm 0.12\%$ and $0.45 \pm 0.09\%$ respectively by 72 h post-irradiation ($p = 0.0772$). There were no significant differences between X-ray irradiated and control RBC groups for volume, haemoglobin (Hb), haematocrit, or supernatant Hb immediately post-thaw ($p = 0.6425$, $p = 0.2314$, $p = 0.0609$ and $p = 0.3951$ respectively). The adenosine triphosphate concentration in both x-ray irradiated and control RBCs was lower after deglycerolisation, but they were not significantly different from each other ($p = 0.9046$). The number of microparticles (MPs) in RBCs increased gradually during storage, with a trend towards higher MP release in x-ray irradiated RBC at 72 h post-irradiation ($p = 0.0870$).

Summary / Conclusions: X-ray irradiation had a significant effect on K^+ release from deglycerolised RBCs during storage. Whilst K^+ , and to a lesser extent; haemolysis, increased during storage following X-irradiation, the levels at 24 h were acceptable, as were the other quality parameters. Deglycerolised RBCs have a shelf-life of 24 h when resuspended in SAGM, and this is also a suitable shelf-life for x-ray irradiated deglycerolised RBCs.

Blood product / components—plasma derived products

P285 | Abstract withdrawn

P286 | Immunoglobulin treatment and survival in patients with chronic lymphocytic leukaemia—data-linkage cohort study in Victoria, Australia

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Background: Patients with chronic lymphocytic leukaemia (CLL) are at increased risk of infections, which account for approximately 30% of

P286 - Table 1. Key characteristics and outcomes of patients with CLL during follow up.

	Never treated with Ig	Treated with Ig	p-value
N patients, n (%)	7694 (84.4)	1422 (15.6)	
Follow-up per patient (year), mean (SD)	4.9 (4.0)	7.1 (4.0)	<0.001
Anticancer treatment admissions per year, mean (SD)	0.5 (1.4)	1.4 (1.7)	<0.001
Infection-related hospital admissions per year, mean (SD)	0.4 (1.2)	1.1 (2.0)	<0.001
Infection-related ICU admissions per year, mean (SD)	0.0 (0.4)	0.1 (0.4)	<0.001
Died, n (%)	2639 (34.3)	694 (48.8)	<0.001

deaths. Prophylactic immunoglobulin (Ig) treatment is common in these patients to prevent infections, but the real-world use of Ig and its association with clinical outcomes remains unclear.

Aims: This study aimed to explore the association between Ig treatment and overall survival in patients with CLL.

Methods: Retrospective data-linkage longitudinal study of patients diagnosed with CLL from 2007 to 2021 in Victoria, Australia. Data from the Victorian Cancer Registry and Death registry were linked with hospital admissions by the Centre for Victorian Data Linkages. Patient characteristics, anticancer treatment, and clinical outcomes were compared in patients who had never received Ig treatment and those who had at least one Ig treatment episode. Kaplan-Meier survival analysis was used to estimate survival outcomes, and Cox survival analysis to assess the impact of Ig treatment on overall survival after controlling for prognostic factors.

Results: A total of 9116 patients with a diagnosis of CLL were included in this analysis. The median follow-up was 5.3 years, ranging from 1 day to 14.7 years, and 66.8% of patients had at least one hospital admission. Over the follow-up period, 15.5% of patients received Ig, 36.1% had anticancer treatment, 43.3% experienced an infection leading to hospitalization, 9.2% were admitted to the intensive care unit (ICU) due to infection, and 36.6% died. Patients who received Ig at any time were significantly different to those who had never received Ig in terms of baseline characteristics (e.g., age, comorbidities), treatment and clinical outcomes during the follow up period (Table 1). The median time to death was 10.8 years. The number of admissions for Ig treatment during follow up was not significantly associated with overall survival (HR = 0.98; 95% CI 0.97, 1.00) after adjustment for prognostic factors. Hospital and ICU admissions due to infections during the follow-up period were associated with a significantly increased risk of death (HR = 1.09, 95% CI 1.08, 1.10; HR = 1.39, 95% CI 1.33, 1.46; respectively), as was the number of anticancer treatments (HR = 1.23, 95% CI 1.29, 1.26). Higher comorbidity burden and older age at baseline were significant prognostic factors for mortality, whereas sex was not significantly associated with survival.

Summary / Conclusions: In this real-world cohort of patients with CLL, hospital and ICU admissions due to infection were significantly associated with increased risk of death, however Ig use was not significantly associated with overall survival. Given the increasing use and costs of Ig, these results highlight the need to evaluate the real-world

relationship between Ig and infections further, and determine which patient subgroups (and at what point in their disease) may benefit more from Ig treatment.

P287 | Use of intravenous albumin—a guideline from the International Collaboration for Transfusion Medicine Guidelines

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Background: Albumin is commonly employed across a wide range of clinical settings to improve hemodynamics, facilitate fluid removal, and manage complications of cirrhosis. The International Collaboration for Transfusion Medicine Guidelines (ICTMG) developed guidelines for the use of albumin in patients requiring critical care, undergoing cardiovascular surgery, kidney replacement therapy, or experiencing complications of cirrhosis.

Aims: To develop clinical recommendations on the use of albumin in patients requiring critical care, undergoing cardiovascular surgery, kidney replacement therapy, or experiencing complications of cirrhosis.

Methods: A guideline development panel was appointed to facilitate this guideline development process. This included clinicians, methodologists, subject matter experts, and a patient representative. The evidence informing this guideline arises from systematic literature reviews and meta-analyses of randomized clinical trials. The search for evidence was conducted in multiple databases from inception to November 23, 2022. Analytical framework and PICO (Population, intervention, comparator, and outcome) questions were developed that focused on mortality and other clinically important outcomes for adults and pediatric patients requiring critical care, undergoing cardiovascular surgery, kidney replacement therapy, or with cirrhosis. The guideline panel reviewed the evidence and formulated guideline recommendations using the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) methodology. The guideline also underwent a revision upon public consultation.

Results: The panel formulated 14 recommendations on the use of albumin. In adult critical care (3 recommendations), neonatal critical care (2 recommendations), pediatric critical care (1 recommendation), cardiovascular surgery (2 recommendations), kidney replacement therapy (1 recommendation), and complications of cirrhosis (5 recommendations). Of the 14 recommendations, 2 had moderate certainty of evidence, 5 had low certainty of evidence, and 7 had very low certainty of evidence. Two of the 14 recommendations suggested conditional use of hyperoncotic albumin for patients with cirrhosis undergoing large-volume paracentesis or with spontaneous bacterial peritonitis. Twelve of 14 recommendations did not suggest transfusing albumin in a wide variety of clinical situations where it is commonly transfused.

Summary / Conclusions: There are currently few evidence-based indications that support the routine use of albumin in clinical practice to improve patient outcomes. This guideline provides clinicians with actionable recommendations on the use of albumin in patients requiring critical care, undergoing cardiovascular surgery, kidney replacement therapy, or experiencing complications of cirrhosis.

P288 | Octaplex usage in UCLH NHS Foundation Trust

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Background: Prothrombin complex concentrate (PCC) allows rapid reversal of anticoagulation with VitaminK antagonists and has a role

in reversing the action of direct factor Xa inhibitors in context of major/life-threatening bleeding and prior to emergency surgery with high bleeding risk. The use of PCC at University college London hospital, UCLH was audited against the National Institute for Health and Care Excellence, NICE guidance NG24 and local guidance.

Aims: Assessment of the use and outcomes of PCC (Octaplex®) at UCLH.

Methods: Retrospective case note review of 67 patients who received PCC at UCLH over 12 months, September 2022-2023.

Results: There was a male predominance (42/74, 63%), median age 77 years (range 46-97 years) and majority 66/67 on anticoagulant. In our anticoagulated cohort, there was an even split between patients on warfarin (33/67) and DOAC (33/67). Patient comorbidities: 85% atrial fibrillation (AF), 69% cardiac/renal failure, 75% hypertension (HTN), 13% previous thrombosis, 5% liver impairment. Main indication for anticoagulation for patients on a DOAC and on warfarin was AF, 91% (30/33) and 70% (23/33) respectively. Rest of 10% (3/33) patients on a DOAC were anticoagulated for previous thrombosis and cardiomyopathy with an ICD. In the warfarin group, other indications: 21% (7/33) Antiphospholipid Syndrome, 6% (2/33) metallic heart valve, 3% (1/33) previous thrombosis. Majority of patients required PCC for bleeding: 91% patients on DOAC, 82% patients on warfarin. Intracranial haemorrhage was the main bleeding complication: 76% (25/33) on DOAC and 52% (17/33) on warfarin. Gastrointestinal haemorrhage was primarily observed in patients on warfarin, 24% (8/33), only 3% (1/33) in patients on DOAC. Small number of patients had other bleedings: traumatic or post procedure haematomas - 9% (3/33) patients on DOAC and 6% (2/33) on warfarin had per vagina bleeding post biopsy and spontaneous muscle haematoma. 18% (6/33) patients on warfarin compared to 9% (3/33) patients on DOAC, required PCC prior to emergency procedures. These included bowel surgeries, spinal surgery, orthopaedic surgery, procedures under Interventional Radiology and Lumbar Puncture. The average dose of PCC was 2549 units (1000 to 3000 Units). None of the patients needed a repeat dose and it was well tolerated with no reactions. In around 30 days following administration of PCC, 12% (8/67) patients had a thrombotic event, which included 6/8 stroke, 1/8 aortic occlusion and 1/8 pulmonary embolism. None of these patients were on anticoagulation at the time of thrombosis, had any previous thromboembolic events, but had prothrombotic factors including cancer, complicated inpatient admission, antiphospholipid syndrome and AF with CHADS-VASc >5. 3/67 (4.4%) developed a stroke within 7 days of PCC. On a 30 day follow up 18/67 (18%) died. 8/18 (44%) primary cause of death was bleeding, which was mainly intracranial haemorrhage in 7/8 patients. In 3/18 (17%) patients, cause of death was an ischemic event. Higher mortality following a major bleeding event was observed in patients on warfarin (57%, 4/7) compared to DOAC (45% 4/11).

Summary / Conclusions: Our findings demonstrate 100% compliance with the NICE guidance. We did not identify any inappropriate use or waste of PCC. We demonstrate high rates of thrombosis following use of PCC and therefore clinicians need to consider a risk benefit approach in patient at risk of thrombosis. We also found that there

was a higher mortality rate following major bleeding for patients anticoagulated with warfarin compared to a DOAC.

P289 | Navigating the healthcare journey—immune globulin recipients' experiences and engagement in the health system

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Background: Human plasma is a vital source of over 25 therapeutic proteins used to treat a range of bleeding, hemostatic, immunological, and metabolic disorders. At the forefront of these plasma-derived products is immune globulin (Ig), which has seen a substantial increase in demand in recent years due to its expanding applications in various medical treatments. The growing reliance on Ig underscores the escalating need for plasma donations, highlighting its critical role in healthcare. In Canada, efforts to enhance plasma sufficiency have led to establishing centers specifically dedicated to plasma collection via voluntary non-remunerated. Research in the field of plasma donation indicates that plasma donors are motivated by a desire to contribute to the national plasma supply, and are interested in knowing more about the recipient. This study aims to draw a connection between plasma donors and recipients by investigating Ig recipient experiences in Canada's healthcare system.

Aims: This narrative study delved into the experiences of Ig recipients in Canada, focusing on their understanding, involvement, and engagement with the dissemination of immune globulin in Canada's health system.

Methods: Employing a qualitative narrative methodology, this study engaged Ig recipients across Canada through two rounds of interviews. The initial interviews focused on personal treatment experiences, shedding light on the recipients' knowledge about their condition, the impact of Ig therapy, and the challenges involved in managing chronic conditions. Subsequent interviews, which form the focus of this presentation, delved deeper into their interactions with the blood system. These interactions encompassed their knowledge about donation and the blood system, involvement in Ig approval and decision-making, experiences with healthcare providers, and participation in patient advocacy groups, exploring how recipients interface with a complex healthcare system.

Results: We interviewed 23 Ig recipients across Canada. Ig recipients tap into various information sources, including advocacy groups and healthcare providers. However, there is a need for information tailored to their unique health conditions and the daily life challenges they face. This gap underscores a profound quest for personalized knowledge and empowerment within an often complex health system. Central to this empowerment was the desire for a deeper understanding of Ig manufacturing, donation, and decision-making regarding availability and approval. Our findings indicate a spectrum of engagement among recipients, ranging from those who feel marginalized and experience a sense of limited control over decisions impacting their

health to those actively engaged with the blood system through advocacy activities that promote greater availability of Ig.

Summary / Conclusions: This study sheds light on the experiences of Ig recipients in Canada, emphasizing their need for tailored information and participation in healthcare decisions. It underscores the imperative for a more responsive, inclusive healthcare system that empowers patients with knowledge and agency, fostering a patient-centric model in treatment and policymaking.

P290 | Development of plasma fractionation infrastructure in low- and middle-income countries—success story of Armenia in meeting domestic demand for anti-D immunoglobulin

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Background: Plasma fractionation plays a vital role and often the only technological approach in producing life-saving medicines for treating various pathologies, such as immunological disorders, primary immunodeficiency, congenital deficiencies, and many bacterial and viral infections. While high-income countries have proper access to plasma-derived medicinal proteins (PDMPs), low- and middle-income countries (LMICs) often have limited or no access to these essential medications due to lack of local infrastructure and high costs, making them unaffordable for much of the population. Industrial plasma fractionation is a complex and highly regulated technology, and therefore, remains largely inaccessible to many LMICs. This situation is worsened by the lack of domestic fractionation capabilities, leading to the discard of surplus plasma from blood processing.

Aims: The main aim was to address the challenges of limited access to PDMPs in LMICs. This involves exploring the possibility of implementing small-scale production procedures in local blood establishments or national service centers, as recommended by the WHO and the Working Party for Global Blood Safety under the ISBT. Local production using available technologies, can improve access to vital plasma products and reduce plasma wastage with small investment, making it a practical option. Specifically, the model of plasma fractionation in Armenia was examined as a case study.

Methods: The method include exploring Armenia as one of successful examples of implementing this approach. The plasma fractionation facility in Armenia was initially established in 1971 at the Yolyan Hematology Center, where the production of plasma-derived fibrinogen, albumin, anti-staphylococcal plasma, and fibrin sheets was carried out. These products were distributed in the republic's hospitals and exported to other Soviet countries. In 1975, the production of Rho(D) immunoglobulin preparation began, a practice that continues to this day. Following the collapse of the Soviet Union, most of the fractionation operations stopped, except for the production of anti-D immunoglobulin. However, the production method used became globally outdated, failing to meet the purity levels

required by current international standards, particularly concerning protein composition. In 2018, a public-private collaboration led to the establishment of a small-scale plasma fractionation facility, adopting advanced technologies such as the Kohn 6th cold ethanol fractionation method with additional purification and viral removal steps.

Results: The updated technology in Armenia enabled the small-scale production of high-quality anti-D IgG, which is now supplied to maternity hospitals through state funding and ensuring whole demand of this essential medicine effectively used to prevent hemolytic disease of the fetus and newborn (HDFN). The pharmaceutical facility, production processes and quality/safety control measures for raw materials and final product have been aligned with EU GMP, WHO guidelines, and European Pharmacopoeia requirements. Currently, efforts are ongoing to expand the production to include other IgG and plasma-derived products using the existing infrastructure and technologies.

Summary / Conclusions: The Armenian model demonstrates a successful strategy for LMICs to develop their own PDMPs. It serves as an example model within the global movement towards self-sufficiency in plasma fractionation, particularly in LMICs where such resources are most needed.

P291 | Albania experience in plasma transfusion

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Background: Plasma is a product of whole blood prepared after centrifugation of donated blood, but also obtained through apheresis. Plasma is the main component of blood that contributes to 55% of total blood volumes. Transfusions of plasma are used to correct massive bleeding, coagulopathies disorders and immune deficiency. Plasma transfusions can also help increase blood volume and prevent shock.

Aims: To identify how many fresh frozen plasmas (ffp) and frozen plasmas (fp) have been fractionated and how they are distributed for transfusion according to the needs of clinics in the university hospital centers.

Methods: This is a retrospective study conducted in the blood collection and processing department at the National Blood Transfusion Center, Tirana. The data was collected through the EmoWeb electronic system over a one-year period, from January 2023 to January 2024. The two largest university hospital centers, such as "Mother Teresa" (QSUNT) and the Hospital of Trauma (SUT), were taken into consideration for the distribution of fresh frozen plasmas and frozen plasmas. Whole blood was taken in the amount of 350–450 mL in top and bottom bags (Macopharma) and/or Reveos blood bags (Terumo) in CPDA-1. After collecting whole blood, the components are separated after 5 to 8 h and kept at room temperature. The plasmas were obtained by centrifugation of whole blood by single-step heavy spin in the Roto Silenta 630 Hettich centrifuge and the Reveos automated blood processing system. After centrifugation, extraction was carried

out in an automatic macopress separator, followed by rapid freezing at -65°C for 1 h and storage at -20°C in the KW Freezer. All those complete blood units centrifuged within 24 h after the collection of donated blood were considered fresh frozen plasma, while frozen plasma was centrifuged after 24 h.

Results: From the national blood transfusion center, 17,589 units of ffp and 5306 units of fp were obtained, of which it turns out that 9834 ffp and 3883 fp units were transfused. In the university hospital center "Mother Teresa," the unit of critical care prevails in the transfusion of fresh frozen plasma with 1213 units and frozen plasma with 351 units due to clinical conditions, followed by gastro-hepatology with the use of 800 units of fresh plasma and 276 of frozen plasma. Meanwhile, at the University Trauma Hospital, surgery prevails with 361 units of fresh frozen plasma against frozen plasma with 105 units, followed by ortho-trauma with 347 units of fresh frozen plasma and 92 frozen plasmas due to the nature of the hospital.

Summary / Conclusions: When comparing fresh frozen plasmas to frozen plasmas, it is observed that during a one-year period, the number of units has increased, increasing the quality of transfusion products. Both university hospital centers, QSUNT and SUT, have managed to successfully cover all requests for ffp and fp according to the clinical needs of hospitals.

Blood product / components—pathogen inactivation

P292 | Amotosalen/UVA treatment of platelet and plasma components to inactivate WHO reference bacterial strains

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Background: The INTERCEPT® Blood Systems (IBS) for platelets and plasma utilize amotosalen and UVA light to inactivate a wide range of pathogens and leukocytes in platelet concentrates (PC) and plasma. IBS for Platelets is routinely used for the treatment of apheresis and whole blood (WB) derived platelets in Europe, and in the US for the treatment of apheresis platelets (TRIMA® in 100% plasma or AMICUS® for 65% PAS-C/35% plasma). IBS for Plasma is available both in Europe and the US. The World Health Organization (WHO) Expert Committee on Biological Standardization (ECBS) in association with the Paul-Ehrlich-Institut (PEI) approved an extended panel of bacterial strains to evaluate methods for improving the microbial safety of blood components (Spindler-Raffel *et al*, 2015).

Aims: The aim of this study was to evaluate the inactivation of WHO reference (PEI) bacterial strains in platelet and plasma components using the INTERCEPT® Blood Systems.

Methods: Apheresis PC collected in 100% plasma or 65% Platelet additive solution (PAS-C)/35% plasma were pooled into individual units of 420 mL with platelet doses of 4.0 to 5.0×10^{11} and 4.0 to 7.9×10^{11}

P292 - Table 1: Bacterial inactivation using amotosalen/UVA treatment for human plasma and platelet concentrates in 100% plasma (PLS) or in 65% PAS-C/35% plasma (PLS).

Bacteria (Strain)	Matrix	Log Reduction (Log cfu/mL)
<i>Pseudomonas fluorescens</i> PEI-B-P-77	PC (100% PLS)	6.6 ± 1.4
<i>Pseudomonas fluorescens</i> PEI-B-P-77	PC (65% PAS/35% PLS)	7.8 ± 0.1*
<i>Pseudomonas fluorescens</i> PEI-B-P-77	Plasma	7.2 ± 0.8
<i>Enterobacter cloacae</i> PEI-B-P-43	PC (100% PLS)	6.5 ± 0.1
<i>Enterobacter cloacae</i> PEI-B-P-43	PC (65% PAS/35% PLS)	6.7 ± 0.1*
<i>Bacillus thuringensis</i> PEI-B-P-07	PC (100% PLS)	5.7 ± 0.1
<i>Bacillus thuringensis</i> PEI-B-P-07	PC (65% PAS/35% PLS)	5.5 ± 0.1*

* No residual bacteria were detected post UVA treatment. The limit of detection is 1 cfu/50 mL.

respectively (IBS for Platelets – Dual Storage (DS) Processing Set). Human plasma donations were collected and pooled to yield individual units of ~650 mL (IBS for Plasma). Four replicates per platelet matrix were performed for 3 PEI strains of transfusion-relevant bacteria, as well as for *P. fluorescens* in plasma, with each replicate consisting of one component spiked with a single PEI strain. The contaminated PC and plasma units were then treated with the IBS for platelets and plasma, respectively. Samples were taken pre-UVA illumination (~5 mL) and post-UVA illumination prior to CAD treatment (~50 mL) and were analyzed for bacterial titer by plating on appropriate media (100µL–10mL/plate) and incubating for at least 24 h.

Results: INTERCEPT treatment of platelet and plasma units spiked with bacteria from the PEI/WHO strains (Table 1) led to robust bacterial inactivation (Table 1).

Summary / Conclusions: The INTERCEPT Blood System for Plasma consistently inactivated high titers of *P. fluorescens*. The INTERCEPT Blood System for Platelets efficiently inactivated *P. fluorescens*, *E. cloacae* and *B. thuringiensis*. The data demonstrate that IBS robustly inactivates the tested WHO standardized bacteria strains associated with transfusion-transmitted bacterial infections (TTBI).

P293 | Amotosalen and UVA treatment of *Bacillus mobilis*, *Acinetobacter seifertii*, *Staphylococcus saprophyticus*, and *Leclercia adecarboxylata* from a contaminated apheresis platelet unit

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Background: The INTERCEPT® Blood System for Platelets uses a combination of amotosalen and UVA light to inactivate pathogens and leukocytes in platelet concentrates (PC). The system is in routine use

in US and EU blood centers to treat either apheresis- or whole-blood derived platelets. In 2021, a septic transfusion reaction involving an INTERCEPT-treated apheresis PC was reported in Ohio, USA. Bacteria were isolated from the storage bag and identified as *Bacillus mobilis* (BM), *Acinetobacter seifertii* (AS) (previously identified as *Acinetobacter baumannii* complex), *Staphylococcus saprophyticus* (SS), and *Leclercia adecarboxylata* (LA). Only SS and AS were cultured from the patient. A subclinical leak near the platelet unit port suggested that environmental contamination of the unit occurred after pathogen reduction. AS, SS and LA are effectively inactivated by the INTERCEPT Blood System for Platelets alone and in combination (Fadeyi *et al.*, *Transfusion*, 2020). This was the first published report of a *Bacillus mobilis* contamination and no inactivation data are available.

Aims: The aim was to assess the inactivation of BM alone and in combination with AS, SS and LA in apheresis platelets using the INTERCEPT Blood System for Platelets.

Methods: The bacterial strains were submitted to Charles River Laboratories for sequence typing using AccuGENX-ST. For the pathogen inactivation assessments, 335 mL of PC in 35% plasma and 65% PAS was inoculated with 3.4 mL of a log culture of an individual bacterial species or a 1:1:1:1 (volume) mixture of BM, AS, SS and LA. The contaminated platelet component was pathogen-inactivated utilizing the INTERCEPT Dual Storage Platelet Processing Set including the compound adsorption device (CAD), transferred into storage bags, and stored at 22°C under standard conditions. Samples were taken pre- and post-treatment, post-CAD, day 5, and day 7. Bacterial titer was measured by plating on LB agar.

Results: AccuGENX-ST sequencing showed that the AS strain was clonally related to two previously reported *A. baumannii* complex bacteria implicated in TTI cases in California and North Carolina (Fadeyi *et al.*, *Transfusion*, 2020; Fridey *et al.*, *Transfusion*, 2020; Villa *et al.*, *Transfusion* 2023). Both the SS and LA were clonally related to isolates involved in prior TTI cases in either Virginia or North Carolina, respectively. For the individual pathogen reduction assessment of a vegetative culture of BM, 3.6 ± 0.1 log CFU/mL was inactivated with no detectable bacteria observed out to 7-days post-collection. For the combination assessment, inactivation of the combined mixture of BM (vegetative culture), AS, SS and LA were observed at >7.0 ± 0.0 log CFU/mL with no detectable bacteria post-treatment out to 7-days. The calculated inactivation for each bacteria in this combined inactivation was 3.0 ± 0.0 log cfu/mL, 7.5 ± 0.0 log cfu/mL, 7.4 ± 0.1 log cfu/mL and 6.5 ± 0.1 log cfu/mL for BM, AS, SS, and LA, respectively.

Summary / Conclusions: Isolates of AS, SS and LA from the Ohio case were clonally related to bacteria involved in TTIs across the United States suggesting a possible common origin, as previously described (Villa *et al.*, 2023).

We demonstrate that amotosalen/UVA treatment can effectively inactivate multiple bacterial species in a single unit, including BM, AS, SS, and LA alone and in combination. Further studies are needed to understand the mechanism of contamination of units after treatment during the storage period.

P294 | Elevated plasma testosterone concentrations with males on testosterone replacement therapy are mitigated with pathogen reduction technology.

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Background: Donors on testosterone replacement therapy (TRT) may require frequent whole blood donation due to erythrocytosis. However, FDA guidelines prevent the use of plasma-based products secondary to transfusing components with possible increased testosterone concentrations. The number of TRT donors with supraphysiologic testosterone levels that could pose a risk to patients has not been studied in a large cohort. Additionally, there are no current mitigation strategies to remove excess testosterone from transfusable plasma.

Aims: Our study aims were twofold: compare red blood cell (RBC) component supernatant and plasma testosterone concentrations from TRT donors and use the INTERCEPT pathogen reduction technology to evaluate if testosterone levels can be degraded by UVA light or adsorbed with the compound adsorption device (CAD).

Methods: Over six-months, all donors were questioned about TRT use at our blood center. Select male TRT donors and controls were collected as leukoreduced CPDA1 whole blood and processed into RBC and plasma components. Less than one-day old RBC component supernatants and plasma samples were dialyzed and analyzed by liquid chromatography tandem mass spectrometry to measure free and total (free and bound to carrier proteins) testosterone. Pathogen reduction (INTERCEPT® Blood System) on plasma components with supraphysiologic levels was completed 10 times and sampled: before treatment, after UVA illumination, and after the CAD adsorption step. Statistical significance was evaluated with a nonparametric, two-tailed t-test.

Results: TRT donors made up 14% of the male donor population over 18 years of age (range: 20-77 years) and gave 456 whole blood units. TRT donors ($n = 76$) and matched controls ($n = 48$) were selected for testosterone testing. Supraphysiologic testosterone was seen in 33 TRT donors (42%) compared with 2 control donors (8%). Free and total testosterone was significantly higher in all TRT donor components compared with controls (Table 1). Testosterone levels were similar between the RBC supernatant and plasma with a mean difference of: 4.8 pg/mL for free testosterone and 18.8 ng/mL for total testosterone. The 10 pathogen reduced units showed that testosterone was not degraded by UVA light ($p > 0.9999$), but the CAD incubation reduced the free and total testosterone by 88.4% (571.72 to 73.8 pg/mL) and 84% (1498.61 to 240.59 ng/mL), respectively ($p = 0.0065$).

Summary / Conclusions: Donors on TRT presented with significantly higher testosterone levels than controls, and testosterone was similarly distributed between RBC and plasma components after manufacturing. The risk of supraphysiologic testosterone levels being transfused may be abrogated by INTERCEPT pathogen reduction treatment process. The CAD dramatically decreased free and total testosterone concentrations to the lower reference range or below. Further studies to validate the removal of testosterone from plasma can support the transfusion of pathogen reduced plasma from TRT donors.

P294 - Table 1. Free and Total Testosterone in TRT and Control Donors*

		Control Mean \pm SD (range)	TRT Mean \pm SD (range)
Testosterone	Component	$n = 46$	$n = 78$
Free (pg/mL)	Plasma	66.9 ± 30.5 (32.6-195.9)	257.3 ± 313.3 (12.9-2357)
	RBC supernatant	66.9 ± 32.8 (34.7-197.1)	265.2 ± 332.8 (14.0-2482)
Total (ng/mL)	Plasma	359.5 ± 144.5 (149.6-680)	829.7 ± 722.6 (45.1-5685)
	RBC supernatant	354.9 ± 137.5 (149.6-673.4)	801.0 ± 755 (44.3-6140)

*All p value comparisons are <0.0001 .

P295 | Pathogen-reduced platelets for the prevention of bleeding—systematic review and meta-analysis (an update)

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Background: Despite improvements in donor screening and laboratory testing, a small risk of transfusion-transmitted infections (TTIs) remains for the recipient of a platelet transfusion. Pathogen-reduction may reduce TTI risk further, however, some published clinical studies have raised concerns about the clinical effectiveness of pathogen-reduced platelets (PRPs) compared to standard platelets (preventing bleeding, platelet count increment).

Aims: To assess the effectiveness of PRP for preventing bleeding in people of any age requiring platelet transfusions.

Methods: This is an update of a Cochrane review, previously published 2013 & 2017, incorporating new data, and is current to January 2024. We included randomised controlled trials (RCTs) comparing

transfusion of PRPs with standard platelets only. The review was conducted in accordance with PRISMA and Cochrane guidelines.

Results: We identified 4 new trials in this update, therefore 16 RCTs were included in the analysis. Data were split into two comparisons and included PRP methods using InterceptTM, MirasolTM, and TheraflexTM. There were no credible subgroups differences between these different methods, and so here we present the aggregate data.

Summary / Conclusions: In people with haematological or oncological disorders who are thrombocytopenic due to their disease or its treatment, we found that PRP transfusions resulted in a greater risk of developing platelet refractoriness, and probably resulted in a higher risk of "clinically significant" bleeding and an increase in the number of platelet transfusions required. There may also be an increase in "any" bleeding. There is probably no difference in serious side effects, there may be no difference in mortality, and we are uncertain about the impact on the risk of severe bleeding. In this population, none of our outcomes suggested any benefit from using PRP, and sometimes suggested a harm when compared to standard platelets. There were no credible differences between the different methods of pathogen-reduction assessed (InterceptTM, MirasolTM, or TheraflexTM).

P295 - Table 1: Results

Outcome	PRP vs standard platelets stored in plasma (or unknown solution)	PRP vs standard platelets stored in platelet additive solution (PAS)
Any bleeding (WHO 1+)	Increased risk with PRP Low certainty RR 1.11 95% CI 1.06 to 1.17, 7 RCTs, N = 1654	Increased risk with PRP Low certainty RR 1.10 95% CI 0.98 to 1.21, 4 RCTs, N = 938
Clinically significant bleeding (WHO 2+)	Increased risk with PRP Moderate certainty RR 1.10 95% CI 1.00 to 1.22, 6 RCTs, N = 1804	Increased risk with PRP Low certainty RR 1.16 95% CI 0.98 to 1.38, 6 RCTs, N = 1348
Severe bleeding (WHO 3+)	Uncertain Very low certainty	Uncertain Very low certainty
All-cause mortality	No difference Low certainty RR 0.85 95% CI 0.50 to 1.46, 7 RCTs, N = 1912	No difference Low certainty POR 0.77 95% CI 0.41 to 1.47, Peto, 8 RCTs, N = 1548
Serious adverse events	No difference Moderate certainty RR 0.96 95% CI 0.79 to 1.17, 8 RCTs, N = 1956	Uncertain Very low certainty
Platelet refractoriness [#]	Increased risk with PRP High certainty RR 2.04 95% CI 1.74 to 2.39, 8 RCTs, N = 1937	Increased risk with PRP Moderate certainty RR 1.47 95% CI 1.28 to 1.70, 4 RCTs, N = 1288
Platelet transfusions per participant	Increased with PRP Moderate certainty MD 1.29 95% CI 0.84 to 1.74, 6 RCTs, N = 1610	Increased with PRP Moderate certainty MD 0.94 95% CI 0.55 to 1.33, 4 RCTs, N = 1302

[#] Defined differently across studies most commonly: 2 successive 1-h CCI below 7.5×10^3 or 24-h CCI below 4.5×10^3 .

P296 | Comparison of pathogen-reduced platelets for 5 days of storage treated with two different commercially available pathogen-inactivation technologies

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Background: Pathogen inactivation treatment (PIT) reduces the risk of bacterial, viral and parasite transmission through platelet transfusion, as well as the incidence of leukocyte-mediated transfusion reactions. However, it also may have an impact on platelet quality and efficacy. Comparison studies of different commercially available PIT technologies with platelets in platelet additive solution (PAS) have been conducted reporting significant differences between technologies with respect to platelet quality during the course of storage, but such studies with platelets in 100% plasma are lacking to date.

Aims: Assessment of the impact of commercially available PIT technologies on the quality of apheresis platelets in 100% plasma until end of shelf life.

Methods: Apheresis platelets in 100% plasma from voluntary donors with comparable attributes were collected with a Trima (Terumo BCT, U.S.A.), or an MCS+ (Haemonetics, U.S.A.) device and treated with either amotosalen/UVA (AS) (INTERCEPT Blood System, Cerus, U.S.A.) or Riboflavin/UVB (RB) (Mirasol PRT System, Terumo BCT) technology, followed by 5-day storage. Platelet count and pH (local standard QC testing) were monitored during the course of storage using a D3 hematology analyzer (Drew Scientific, U.S.A.) and a pH-102 device (Portlab, Russia) respectively. The two-sample (unpaired) t-test was used to calculate statistical significance (a p-value of <0.05 was considered significant, a p-value of <0.01 highly significant).

Results: 30 platelet units per arm were collected with an average platelet count of $5.3 \times 10^{11} \pm 0.3$. There was no significant difference of donor characteristics with respect to sex (20% / 27% female), body weight in kg (83.3 ± 3.8 / 83.9 ± 4.9 , $P = 0.598$), total number of former apheresis donations (33.0 ± 14.2 / 32.6 ± 14.8 , $p = 0.915$) and donor platelet count (220.7 ± 12.2 / $223.2 \pm 11.8 \times 10^9/L$, $p = 0.423$) between the study arms. Red blood cell (RBC) and white blood cell (WBC) counts of all platelet units were within specifications for PIT ($<4 \times 10^6$ RBC/mL and $<250 \times 10^6$ WBC/unit). The platelet loss during processing was higher in AS units (5.3%) compared to RB units (2.7%). Until day 3 of storage, there was no significant difference in platelet quality parameters between the study arms. Between day 3 and day 5 of storage, the average platelet count in the AS arm dropped to $98.4\% \pm 7.7$ of the post-treatment value (day 5 platelet count 5.0 ± 0.3), and in the RB arm to $84.2\% \pm 15.7$ (day 5 platelet count 4.2 ± 0.2) ($p < 0.01$). Between day 3 and day 5 the average pH, as surrogate marker for metabolic activity, dropped in the AS arm from 7.2 ± 0.3 to 6.9 ± 0.5 ; and in the RB arm from 7.1 ± 0.1 to 6.5 ± 0.5 ($p < 0.01$).

Summary / Conclusions: The treatment of apheresis platelets with PIT technologies affected the in vitro quality differently, with potential impact

on the clinical outcome. We recognized a highly significant reduction of platelet count and pH of RB-treated platelets compared to AS-treated platelets after day 3 of storage, leading to a reduction of storage time to maximum 3 days for RB platelets in 100% plasma in our blood center.

P297 | Implementation of pathogen-reduced cryoprecipitated antihemophilic factor from restricted quarantine plasma to maintain adequate supply

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Background: Massive bleeding protocols require the transfusion of cryoprecipitated antihemophilic factor (CP) as the main source of fibrinogen in our institution. It was manufactured from released quarantine fresh frozen plasma (QFFP), to ensure product safety. The demand for CP increased 64%, from 2019 (1082 units) to 2020 (1779 units) and another 216%, from 2020 to 2021 (5615 units). The increased demand was mainly driven by the COVID-19 pandemic (ECMO treatment of severe COVID-19). From 2020, the available released QFFP quantities were not sufficient to meet the demand of CP in our institution. To overcome that shortage, we evaluated the production of CP from pathogen-reduced (PR) restricted (non-released) QFFP and introduced PR-CP in Q3/2021. In 2023, the demand for CP was still on a comparable high level, mainly due to the opening of a new emergency ICU, leading to permanent production of PR-CP. For long-term implementation, we developed a new production method for improved PR-CP quality.

Aims: Evaluation of the quality treatment-eligibility of PR-CP from restricted QFFP and optimization of the production process.

Methods: 650 mL of FFP was collected by apheresis with an Aurora device (Fresenius Kabi, Germany) or a PCS2 device (Haemonetics, U.S.A.). 430 mL were treated with amotosalen/UVA technology (INTERCEPT Blood System, Cerus, U.S.A.), treated- and untreated plasma units were subsequently frozen within 6 h post collection at -40°C . CP was produced by thawing at 4°C for 18 h followed by centrifugation at 4°C with an RC 3C+ centrifuge (Sorvall, U.S.A.). The final target volume of a single CP unit from approx. 210-220 mL plasma is 40 mL. The fibrinogen content and plasma factors quantity were determined with an ACL TOP 750 device (Werfen, Germany). CP thawing was conducted with either a water bath or a Sahara-III plasma thawer (Sarstedt, Germany). Statistical analysis was conducted with the student t-test.

Results: Patients in the infectious diseases ward received on average 10.3 CP transfusions/patient, while other patients in the emergency unit received in average 6.6 CP units/patient ($p < 0.001$) in 2021. The average fibrinogen content of a conventional CP unit from 220 mL QFFP was 0.23 g and of a PR-CP-Unit 0.19 g ($p < 0.05$). Increasing the QFFP volume pre-PR-treatment from 220 mL to 325 mL resulted in an average fibrinogen content of 0.23 g, comparable to a conventional CP unit. In a comparative analysis, the fibrinogen content was 57.1%

higher when we used a plasma thawer (average 0.22 g) compared to a water bath (0.14 g/L) ($n = 25$, $P < 0.001$) for thawing CP. In Q4/2021 11% of the issued CP was PR-CP, 40% from plasma outside our center. In Q1/2022 62% of the issued CP was PR-CP, only 1% from plasma outside our center. A modified production method (M2) including plasma pooling pre-PR-CP production was compared to our standard method of pooling PI-CP post-production (M1). Twelve 240 mL CP units were produced with each method from 24 PR-QFFP units. The fibrinogen content was 1.1 (0.8-1.6) g/dose in M1 PI-CP and 1.5 (0.9-2.1) in M2 PI-CP ($p < 0.05$). The FVIII content was 362 (249-524) IU/dose in M1 PI-CP and 415 (287-721) in M2 PI-CP ($p > 0.05$). The average FXIII content was 274 (201-358) IU/dose in M1 PI-CP and 359 (280-358) in M2 PI-CP ($p < 0.05$).

Summary / Conclusions: The introduction of PR-QFFP allowed us to fulfill the growing CP demand and to maintain self-sufficiency. Improved production methodology increased the average content of fibrinogen (36%) and FXIII (31%) significantly.

P298 | Amotosalen/UVA light pathogen reduced pooled plasma stored frozen for 3 years and liquid for 5 days

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Background: Two types of therapeutic plasmas are produced by the French Blood Service (EFS), plasma secured by quarantine (Q) and plasma pathogen reduced (PR) by amotosalen/UVA light treatment (A-UVA) (INTERCEPTTM Blood System, Cerus). A “previously frozen plasma” (PFP) process consisting after a frozen Q period in thawing

the plasma, A- UVA treating and refreezing for storage was validated. This PFP process is approved in France since November 2022.

Aims: To evaluate 3-years frozen ($\leq -25^{\circ}\text{C}$) stability of A-UVA PFP with subsequent 5-days liquid (4°C) storage.

Methods: 18 groups of 5 CPD whole blood derived plasma units were frozen and stored at $\leq -25^{\circ}\text{C}$ for 30 weeks. After thawing, pools of 5 units were constituted, split in 2×640 mL minimum. Each of the two sub-pools was A-UVA treated, split in 3 units and frozen within 6 h. All plasma units were non-O. Plasma parameters were measured in the thawed plasma pools after the Q period (baseline) and in the plasma units after A-UVA treatment and up to 14-days frozen storage (post treatment), and at 1, 2 and 3 years from collection. After 3 years of frozen storage the plasma units were thawed and stored at 4°C for 5 days. 30 plasma parameters were tested for the frozen storage study and 6 parameters for the liquid storage study.

Results: The results of the 6 parameters tested in the 18 replicates through frozen and liquid storage are reported in the Table below. The requirements from the French Official Journal (JO), at least 70 % of the units with FVIII ≥ 0.5 IU mL and Fibrinogen ≥ 2.0 g/L were met at all periods.

Summary / Conclusions: Previously frozen plasma treated post-thawing with A-UVA retained sufficient levels of plasma proteins, coagulation factors and inhibitors and normal thrombin generation capacity. A sufficient stability of these parameters was observed over a 3-year storage period at $\leq -25^{\circ}\text{C}$ followed by 5-day storage at 4°C .

P298 - Table 1: Results

N = 18	Baseline (after 30-week $\leq -25^{\circ}\text{C}$ storage)	After thawing / A-UVA PR / & frozen storage up to		
		1-year from collection*	3 years from collection*	5 days post-thaw (4°C storage)**
Total proteins (g/L)	58.2 \pm 2.0	60.1 \pm 2.7s	58.99 \pm 2.19	59.28 \pm 2.48
Fibrinogen (g/L)	2.65 \pm 0.17	2.37 \pm 0.14s	2.50 \pm 0.15s	2.48 \pm 0.17
FVIII (IU/mL)	1.10 \pm 0.11	0.78 \pm 0.09s	0.74 \pm 0.06s	0.54 \pm 0.05 s
FVII (IU/mL)	1.03 \pm 0.11	0.79 \pm 0.08s	0.8 \pm 0.10s	0.72 \pm 0.24
Prothrombin Fragment 1+2 (pM/L)	152.8 \pm 28.4	147.8 \pm 29.9	306.63 \pm 107.32s	360.60 \pm 255.07
C3a (ng/ml)	245.8 \pm 77.5	117.9 \pm 44.0	169.23 \pm 84.75s	372 \pm 146.42s
Thrombin generation ETP – (nM thrombin \times min) – Tissue factor 5 pM	1673.7 \pm 159.3	1477.9 \pm 202.1s	1395.7 \pm 117.8s	1364.62 \pm 127.48
				Thrombin generation ETP—(nM thrombin \times min)—Tissue factor 1 pM

* Two-tailed (alpha 0.05) t-test for paired values comparing each frozen storage period to the “pre” data, “s” if significant difference $p < 0.05$.

** two-tailed (alpha 0.05) t-test for paired values comparing 5-day post thaw to 3-year frozen storage data, “s” if significant difference $p < 0.05$.

P299 | The impact of plasma pooling, pathogen-reduction and the freeze/thaw process on plasma quality and standardization

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Background: In Poland, plasma must be quarantined or pathogen-reduced to increase blood safety, especially with respect to potential window-period infections. The majority of plasma is derived from whole blood donations, with a high variability in the total protein profile and volume, due to individual donor factors and production methodology. Plasma pooling to obtain a more standardized product was not common due to potential pathogen transmission risk increase. We implemented a 5 single-donor plasma (SDP) units plasma pooling concept in combination with pathogen-inactivation to generate 6 pathogen-reduced, pooled plasma (PPP) therapeutic units. Besides product standardization and blood safety this has an economic impact, allowing for the production of 20% more plasma units from whole blood donations.

Aims: Assessment of the impact of plasma pooling, PI-treatment and the plasma freeze/thaw process on plasma product quality and standardization.

Methods: Plasma was produced from whole blood (WB) donations with the buffy coat method using a Compomat G5 automated press (Fresenius-Kabi, Germany). Five ABO-identical SDPs were pooled using an Optipool DONOpac plasma pooling set (LMB Technologies, Germany) (1250-1300 mL). After mixing, the pool was separated into 2 minipools (625-650 mL), which subsequently were pathogen-reduced with the amotosalen/UVA technology (INTERCEPT Blood System for Plasma, Cerus). In the integrated disposable set, the minipools were separated into 3 storage bags, resulting in 6 PPPs. The plasma units were frozen within 8 hours after preparation using a plasma freezer (MABAG, Germany) and stored at -30°C. Analysis for FVIII (chromometric assay), fibrinogen (Clauss method) and total protein content (colorimetric assay) was conducted using a bk6100 analyzer (bio-ksel, Poland) in a standardized procedure.

Results: Five experiments pooling 5 SDPs were conducted. The average volume of SDPs ($n = 25$) was 267.4 ± 4.7 mL, (range 21.4 ± 10.1 mL). The average volume of PPPs ($n = 30$) was 219.4 ± 2.4 mL range (0.6 ± 0.5 mL, a variability reduction of 97.2%). The average total volume loss during processing was $1.7 \pm 1.0\%$. The average fibrinogen content of SDPs was 324.6 ± 30.0 mg/dl, (range 114.0 ± 88.0 mg/dl). The average fibrinogen content of PPPs was 264.1 ± 24.0 mg/dl, with a recovery of 81.3%. Post freezing for 1 month the recovery was 232.2 ± 24.3 (71.5%). The average range was 18.6 ± 1.8 mg/dl (a variability reduction of 81.1%). The average FVIII content of SDPs was 131.0 ± 16.4 IU/dl, (79.1 ± 43.9 IU/dl). The average FVIII content of PPPs was 96.4 ± 17.1 IU/dl, a recovery of 73.4%. Post freezing for 1 month the recovery was 85.5 ± 16.9 , (65%). The average range was 26.6 ± 7.4 IU/dl (a variability reduction of 66.4%). The average total protein content of SDPs was 60.7 ± 2.7 g/L, the range 8.3 ± 2.4 g/L. The average total protein content of PPPs was 57.5 ± 2.3 g/L, a recovery of 94.7%. After freezing for 1 month the recovery was 56.5 ± 1.4 , a recovery of 92.9%. The average range was 1.0 ± 0.6 g/L (a variability reduction of 87.9%).

Summary / Conclusions: The product quality post-treatment was within EDQM (FVIII ≥ 50 IU/dl, $\geq 60\%$ fibrinogen recovery) and Polish guidelines (FVIII ≥ 50 IU/dl, FVIII recovery $\geq 70\%$, total protein ≥ 50 g/L, fibrinogen recovery $\geq 60\%$). The volume variability between plasma units (range between highest and lowest values) was reduced in average for 97.2% comparing PPPs to SDPs, the plasma factor/proteins variability in average for 78.5%, increasing product standardization.

P301 | Abstract withdrawn

P302 | The platelet-derived microvesicles in pathogen inactivated apheresis bags stored in PAS

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Background: The impact of pathogen reduction technology (PRT), such as Mirasol, and the effect of platelet additive solutions (PAS) on platelet-derived microvesicles (P-MVs) of transfused apheresis platelets remain unknown.

Aims: Aim of this study was to assess the P-MVs of Mirasol treated platelets in PAS during a 7-day storage period.

Methods: Ten bags containing apheresis platelets stored in PAS were split into two groups: control platelets and PRT-treated platelets. The bags were fitted with a sterile sampling-site coupler and aliquots were withdrawn at regular storage time (1st, 3rd, 5th, and 7th day). The platelet poor plasma (PPP) was isolated from the platelet concentrates by double centrifugation ($2000 \times g$ for 10 mins). The PPP was stored at -80°C until P-MVs evaluation. Nanoparticle tracking analysis was performed by the same operator using the NanoSight NS300 instrument, which is equipped with a 532 nm laser, a high sensitivity sCMOS camera and a syringe pump. The PPP were diluted in particle-free PBS (0.02 μ m filtered) to obtain a concentration within the recommended measurement, corresponding to 1:200 dilution of the initial sample concentration. Each sample was loaded on 1ml syringe that was then placed to the pump. Autofocus was adjusted so that indistinct particles were avoided. For each measurement, five 30 s videos were captured under the following conditions: cell temperature: 25°C; Syringe speed: 100 ml/s. Experiment videos were analyzed using NanoSight NTA 3.4 build 3.4.4 software after capture in script control mode. A total of 1500 frames were examined per sample.

Results: The PRT triggered the P-MVs release on the first day of storage (Control P-MVs = $1.42 \times 10^{10}/10^5$ platelets, PRT P-MVs = $2.47 \times 10^{10}/10^5$ platelets, $p < 0.001$). Regarding the size of P-MVs, the treated platelets on first day had significantly higher numbers of small

20-300nm P-MVs (Control P-MVs = $1.39 \times 10^{10}/10^5$ platelets, PRT P-MVs = $2.41 \times 10^{10}/10^5$ platelets, $p < 0.001$) but no statistically significant difference was observed in the number of large (300-1000nm) P-MVs (Control P-MVs = $2.41 \times 10^8/10^5$ platelets, PRT P-MVs = $6.26 \times 10^8/10^5$ platelets). The microvesiculation rate had a statistically significant increase during the storage period in the controls (Day 1: $1.42 \times 10^{10}/10^5$ platelets, Day 3: $2.65 \times 10^{10}/10^5$ platelets, Day 5: $2.37 \times 10^{10}/10^5$ platelets, Day 7: $2.74 \times 10^{10}/10^5$ platelets) while the rate was observed constant in the inactivated platelets (Day 1: $2.47 \times 10^{10}/10^5$ platelets, Day 3: $2.42 \times 10^{10}/10^5$ platelets, Day 5: $2.18 \times 10^{10}/10^5$ platelets, Day 7: $2.76 \times 10^{10}/10^5$ platelets). Finally, the mean size of the vesicles did not statistically differ in the controls throughout the storage period (Day 1: 153.9 nm, Day 3: 161.8 nm, Day 5: 161.3 nm, Day 7: 164.6 nm), while in the inactivated platelets it increased on the 5th and 7th day (Day 1: 159.3 nm, Day 3: 165.2 nm, Day 5: 173.9 nm ($p < 0.05$), Day 7: 173.1 nm ($p < 0.05$)).

Summary / Conclusions: Microvesicle formation displayed increasing levels for PRT on the first day of storage, but no statistically significant differences were observed throughout the storage period. This observation suggests that PRT treatment has moderate effects on platelet activation during storage. However, the clinical impact of these results needs further investigation. Last, the development of novel biomarkers for evaluation of stored platelets' *in vitro* viability and activity would be valuable in order to determine the optimal time for transfusion.

P303 | Successful Inactivation of pathogenic bacterial strains in double-dose pooled buffy coat platelet using Amotosalen/UVA pathogen reduction treatment

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Background: Platelets components provide ideal conditions for bacterial proliferation which can lead to septic transfusion reactions and significant morbidity and even mortality for vulnerable recipients. Blood establishments worldwide mitigate this risk using different strategies including bacterial screening of platelets and the use of pathogen-reduction technology. Here we describe successful bioburden

reduction of pathogenic strains in whole blood derived 7 buffy coat (BC) double-dose pools (prepared using TerumoBCT TACSI[®] System) through use of the Ceros INTERCEPT[™] Blood System (IBS).

Aims: To demonstrate the ability of the IBS to successfully reduce pathogenic bacteria level to 'Sterile at out-date' under conditions of normal operation. Eight bacterial strains (WHO and IBTS) were used (Table 1).

Methods: A paired case-control study, comparing bio-burden reduction of pathogenic bacteria in pathogen reduced and untreated (control) platelets, was performed. Successful pathogen reduction was indicated by 'Sterility at out-date'. Estimating initial inoculating dose is difficult due to mitigation and production steps taken to reduce bacterial contamination; a 'high' likely initial dose of 500 CFU per platelet dose was used. BCs were separated from whole blood by hard-spin centrifugation on Day 1. Seven ABO-identical BCs were then pooled with 280 ml of Platelet Additive Solution (Macopharma SSP+) using TerumoBCT TACSI system, producing a double-dose (DD) pool. Two DD pools were then combined and mixed; sample was taken to confirm Sterility (8ml in each of BPA and BPN bottles (Biomérieux BACT/ALERT (BTA) 3D)). Combined DD pool (14-BCs) was inoculated with 2,000 CFU (500 CFU/dose). After 2-h rest, the combined DD pool was split in two identical DD pools. Day 1 Pre-PR samples were taken from each DD pool for CFU and BTA 3D. One DD underwent pathogen-reduction using IBS with Dual Storage (DS) sets; the second DD remained untreated as control. On Day 2 after removal of amotosalen (CAD 14-15 h), the PR DD was split into two identical single-dose pools; control DD pool was split into two equal untreated single-dose pools (using DS bags). Samples were taken from each SD pool for CFU and BTA 3D. All platelet pools were agitated at 22°C until expiry. Samples were taken on Day 8 (out-date) for CFU and BTA 3D. All BPA/BPN bottles were incubated on BTA 3D to positivity or 7-days. Strain confirmation was performed by MALDI-TOF (Vitex MS, Biomérieux). This was repeated for all eight strains.

Results: All strains of bacteria proliferated in control pools, increasing in concentration over shelf-life. 7 of 8 selected strains did not grow in pathogen-reduced pools, and were 'sterile at outdate'. *Bacillus cereus* was specifically selected as a spore-producing organism; PR had no effect on the ability of *B. cereus* to grow in pools. This is a known limitation of PR technology. Identification of all bacterial strains isolated from positive bottles was as expected.

Summary / Conclusions: IBS Pathogen reduction technology performed as expected for all eight bacterial strains used, for what we believe to be realistic inoculating doses.

P304 | Development of a next generation illuminator for photochemical inactivation of a broad spectrum of pathogens in platelet concentrates and plasma

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Background: The INTERCEPT[®] Blood System for Platelets and Plasma uses amotosalen and ultraviolet A (UVA) light to inactivate a broad

P303 - Table 1. Bacterial Strains

Organism	Strain	Source	Sterile at Outdate
<i>Staphylococcus epidermidis</i>	IBTS-567/18	IBTS	Y
<i>Klebsiella pneumoniae</i>	PEI-B-P-08	WHO	Y
<i>Staphylococcus aureus</i>	IBTS-073/21	IBTS	Y
<i>Serratia marcescens</i>	IBTS-374/20	IBTS	Y
<i>Enterobacter cloacae</i>	PEI-B-P-43	WHO	Y
<i>Proteus mirabilis</i>	PEI-B-P-55	WHO	Y
<i>Streptococcus dysgalactiae</i>	PEI-B-P-71	WHO	Y
<i>Bacillus cereus</i>	PEI-B-P-57	WHO	N

P304 - Table 1: Pathogen Inactivation in platelets using the INT100 and INT200 Illuminators.

Pathogen	LRF (cfu/mL or pfu/mL) PC in 35% plasma/65% PAS-3		LRF (cfu/mL or pfu/mL) PC in 100% plasma	
	INT100 Illuminator	INT200 Illuminator	INT100 Illuminator	INT200 Illuminator
BTV	≥4.4	≥4.4	5.2	5.0
Ad5	>6.5	>6.5	≥5.7	≥5.4
DENV	>6.3	>6.3	>5.8	>5.8
BVDV	>4.7	>4.7	>4.6	>4.6
<i>Pseudomonas fluorescens</i> *	≥7.8	≥7.6	6.6	7.5
<i>Klebsiella pneumoniae</i> *	≥5.6	≥5.7	3.4	3.6
<i>Clostridium perfringens</i>	>6.6	>6.7	>6.7	>6.7
<i>Staphylococcus aureus</i> *	>7.6	>7.6	≥7.7	≥7.7

* WHO approved bacterial reference strains provided by PEI were used.

P304 - Table 2: Pathogen Inactivation in plasma using the INT100 and INT200 Illuminators.

Pathogen	LRF (cfu/mL or pfu/mL)	
	INT100 Illuminator	INT200 Illuminator
BTV	≥4.3	≥4.3
Ad5	≥4.9	≥5.4
DENV	>6.0	>6.0
BVDV	>5.0	>5.0
<i>Pseudomonas fluorescens</i> *	7.2	≥7.5
<i>Klebsiella pneumoniae</i> *	4.5	5.1
<i>Clostridium perfringens</i>	>6.7	>6.7
<i>Staphylococcus aureus</i> *	>6.5	>6.5

* WHO approved bacterial reference strains provided by PEI were used.

spectrum of pathogens and leukocytes in donor platelet concentrates (PC) and plasma, respectively. The current commercial illuminator (INT100) uses fluorescent bulbs to deliver a controlled dose of UVA light. In recent years, Cerus has developed a new LED-based illuminator (INT200) as a planned replacement for the INT100. There is no change to the intended use of the Illuminator.

Aims: The objective of these studies was to compare the performance of the INT200 Illuminator to the INT100 Illuminator through the evaluation of pathogen inactivation levels achieved in PC and plasma.

Methods: Two-arm pool and split studies were performed with apheresis PC (35% plasma/65% PAS-3 and 100% plasma) and with plasma. Pooled PC or plasma were spiked with the pathogen of interest and split into two identical units. The contaminated PC and plasma components were treated with the INTERCEPT processing sets for platelets or

plasma, respectively. One unit was illuminated using the INT100 Illuminator and the second unit was illuminated using the INT200 Illuminator.

Results: Tables 1 and 2 show the inactivation levels achieved with amotosalen using the INT100 and INT200 Illuminators, as indicated by the log reduction factors (LRFs). The efficacy of inactivation was tested and compared for a wide spectrum of pathogens in PC and plasma, a subset of which is shown in Tables 1 and 2.

Summary / Conclusions: Equivalent levels of inactivation (LRF difference ± 0.5 log) could be achieved for all pathogens in PC and plasma at the UVA light doses tested for INT200, except for *Klebsiella pneumoniae* in plasma and *Pseudomonas fluorescens* in platelets in 100% plasma, where a higher level of inactivation (difference in LRF >0.5 log) was obtained using the INT200 Illuminator. Overall, these results demonstrate that the INT200 illuminator can provide similar performance compared to the INT100 for inactivating pathogens in donor PC and plasma.

P305 | Therapeutic comparative study of whole blood derived platelet concentrates treated with amotosalen-UVA pathogen reduction technique versus irradiated in patients with hemotological pathology

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Background: Different types of platelet concentrates (PC) are currently available for transfusion, such as those that have undergone irradiation (IRPC) or pathogen reduction techniques (PRPC). Knowing its variability both from an analytical point of view and its clinical effectiveness is necessary.

Aims: To know the clinical-analytical differences between PRPC and IRPC prepared from whole blood in adult patients with different onco-haematological conditions.

Methods: After high-speed whole blood centrifugation, Buffy-coats (BC) were obtained. In the case of IRPC, 4 BCs were pooled together with 300 mL of PAS-E and centrifuged with the TACSI system (Terumo®), 12 h later PCs were irradiated. The PRPC were prepared by mixing 8 BCs and 280 mL of PAS-E. This mixture was centrifuged, and a “mega” PC was obtained, which was inactivated with a pathogen reduction technology based on amotosalen and UVA (INTERCEPT™ Blood System) after which it was split into two adult doses. The selected patients with onco-haematological conditions belonged to a single hospital. In the first period, IRPC were transfused exclusively and in the second period, PRPC were transfused. The following clinical-analytical data were recognized: sex, date of birth, weight, height, blood group, underlying pathology, type of treatment, transplantation and type, haemorrhagic risk factors, indication for transplantation, type of platelet transfusion indication (prophylactic vs therapeutic), platelet count before and after

transfusion, PC platelet content, the existence of previous refractoriness, adverse transfusion reactions, the existence of bleeding, bleeding severity, and location, transfusion of packed red blood cells (RBCC), exitus, and its cause. The patient's platelet counts were taken before and after transfusion at 1 and 24 h, and the CCI was calculated. For the analysis, the chi-square statistic was used for categorical variables and Student's t-test for continuous variables with the SPSS software.

Results: We share the preliminary results of this study. We have studied 64 transfusion episodes with 31 IRPCs and 33 PRPCs, with a similar distribution of sex, age, and body surface area between both groups. Also, there are no significant differences in the type of indication for platelet transfusion, previous bleeding risk factors, subsequent adverse reactions, post-transfusion bleeding, or number of transfused RBCC. On the other hand, a significant difference is observed between the groups (IRPC vs PRCP) in: initial diagnosis (more CART and AML in IRPC, more aplastic anaemia and MDS in PRCP), treatment phase (more transplantations in PRPC; more CART and relapse in IRPC), the source of progenitors (more bone marrow in PRPC), death (more in IRPC), and higher CCI, posttransfusion platelet count and platelet content in the product in IRPC (9697 vs 4420; 31225/ μ L vs 19787/ μ L and 2.86×10^{11} vs 2.42×10^{11}).

Summary / Conclusions: No definitive conclusions can be drawn from the preliminary results shown here. It is important to highlight the absence of differences in adverse reactions, post-transfusion bleeding or the number of red blood cell concentrates transfused, which may lead to considering a similar clinical effectiveness for the intended use of PC.

P306 | Impact of a 100% pathogen-reduced platelet inventory and extended shelf-life policy on platelet transfusion requirements in haematological patients. A retrospective study in France

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Background: Pathogen reduction (PR) technology (Intercept, Cerus corporation) was introduced for all platelet concentrates (PC) issued in France in Nov 2017. Shelf life of PC was extended from 5 to 7 days (d) in July 2018. These changes resulted in an increased PC issuing, in part driven by a decrease in platelet content per unit, and a facilitated PC inventory management.

Aims: To assess the impact of this policy on the average age of PR PC transfused and on repeat transfusion rates within 3 days in haematology patients.

Methods: PC characteristics and % of repeat PC transfusion(s) within 3d were analysed between 2021 and 2023. Patients included were

adults hospitalized in haematology departments nation-wide who received no more than 1 PC per day and who underwent at least one additional PC transfusion before d8.

Results: Over the study period, 91,322 PC were transfused to haematological patients, and 34 243 PC met the study criteria. Mean platelet content/PC was 3.9×10^{11} . Shelf-life was 60.5% \leq d5 (0.0% d1, 2.3% d2, 11.8% d3, 21.6% d4, 24.8% d5, 23.5% d6, 15.6% d7). Time to next PC transfusion was 2.62 days for PC \leq d5 vs 2.48 days for PC>d5 ($p < 0.0001$). There was 71.1% repeat transfusions within 3 days: 69.5% for PC \leq d5 vs 73.6% for PC>d5 ($p < 0.0001$). In multivariate analysis, PC>d5 and PC with $<0.5^{10}$ platelets/ recipient kg) were significantly associated with a higher risk of repeat transfusions within 3 days ($p < 0.0001$ for both variables). An incremental increase in the frequency of repeat transfusion within 3 d was observed with increasing storage duration (d1 PC to d7PC) ($p \leq 0.0001$).

Summary / Conclusions: Shifting to 100% PR PC production in France and extending PC shelf life resulted in an increased mean age of PC issued as well as an increased repeat transfusion rates within 3 days in haematology patients. This observational study is based on PC issuing data, irrespective of precise clinical settings. It allows to highlight trends in repeat transfusion rates associated to PC shelf-life in a context associating both PR technology implementation and extended shelf life to 7 days. These results suggest that, when possible, transfusion of PR PC stored \leq 5d storage should be preferred in haematology patients.

P307 | Evaluation of plasma treated for pathogen reduction with amotosalen and a prototype LED illuminator

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Background: The INT100 illuminator used for INTERCEPTTM Blood System (Cerus) processing of platelets and plasma provides a controlled amount of ultraviolet A (UVA) light emitted by fluorescent bulbs. Cerus has developed a new illuminator with a light-emitting diode (LED) light source that is currently being validated. The ergonomics, the software and the user interface have been updated.

Aims: The purpose of this study is to evaluate the quality of fresh frozen whole blood-derived pooled plasma pathogen reduced by amotosalen and UVA LED light (FPP A-UVA), using a pre-production LED illuminator.

Methods: 19 pools of 5 units of plasma derived from leukocyte depleted whole blood were prepared, then divided into 2 parts of close to 650 mL, each connected to an INT31 plasma processing set and treated with amotosalen and UVA (LED light), before excess compound adsorption (through a CAD) and freezing to $\leq -25^{\circ}\text{C}$ within 9 ± 1 h. The results of a selection of parameters out of the 30 tested before treatment (T2),

P307 - Table 1: Results of a selection of parameters tested at different time periods in FFP A-UVA treated with a LED Illuminator

N = 19	T2 Pre-treatment pool	T3 FFP A-UVA after CAD	T5* FFP A-UVA 1 year at $\leq -25^{\circ}\text{C}$	T5/T2
Total Proteins (g/L)	60.08 \pm 1.45	57.79 \pm 1.45	59.21 \pm 1.18s	98.5%
PT (%)	107 \pm 7.0	98 \pm 6.8	91 \pm 4.4s	85.0%
APTT (ratio)	0.85 \pm 0.05	0.88 \pm 0.04	0.99 \pm 0.05s	117.0%
Endogenous thrombin potential (ETP) (%)	112.9 \pm 8.5	112.1 \pm 8.6	102.7 \pm 12.1s	90.9%
Fibrinogen (g/L)	2.68 \pm 0.16	2.43 \pm 0.17	2.49 \pm 0.16	93.1%
Factor II (IU/mL)	1.06 \pm 0.07	0.92 \pm 0.07	0.80 \pm 0.06s	76.1%
Factor V (IU/mL)	0.88 \pm 0.07	0.86 \pm 0.07	0.82 \pm 0.06s	92.9%
Factor VII (IU/mL)	0.99 \pm 0.13	0.86 \pm 0.09	0.81 \pm 0.09	81.2%
Factor VIII (IU/mL)	1.15 \pm 0.25	0.83 \pm 0.18	0.79 \pm 0.17	69.2%
N = 19	T2 Pre-treatment pool	T3 FFP A-UVA after CAD	T5* FFP A-UVA 1 year at $\leq -25^{\circ}\text{C}$	T5/T2
Factor IX (IU/mL)	0.98 \pm 0.09	0.80 \pm 0.07	0.77 \pm 0.06	78.2%
Willebrand activity (Rco %)	85 \pm 16.5	82 \pm 16.5	90 \pm 17.8	105.8%
Protein S (% activity)	100 \pm 8.4	91 \pm 7.0	85 \pm 8.3s	85.9%
Antithrombin (% activity)	102 \pm 5.1	96 \pm 4.2	97 \pm 4.4	94.8%
alpha 2-antiplasmin (% activity)	102 \pm 3.4	87 \pm 4.0	79 \pm 3.6s	77.2%
Plasminogen (% activity)	95 \pm 5.2	87 \pm 5.1	85 \pm 5.3	89.5%
TAT complexes ($\mu\text{g/L}$)	43 \pm 24.7	39 \pm 22.2	27 \pm 15.3	62.3%
ADAMTS 13 (% activity)	101 \pm 16.8	94 \pm 11.2	84 \pm 6.5s	82.8%
C3a ($\mu\text{g/L}$)	384.6 \pm 105.5	10.4 \pm 5.2	62.3 \pm 18.7s	16.2%

* Paired t-test T5 versus T3 – s if significant difference $p < 0.05$.

then after treatment & CAD before freezing (T3), and after 3 months (T4: not shown), and 1 year (T5) at $\leq -25^{\circ}\text{C}$, are presented.

Results: The results of the analyses are shown in Table 1. FFP A-UVA treated with the LED illuminator complies with the French regulatory requirements (Official Journal of 4 June 2020) of at least 70 % of the units with Factor VIII:C ≥ 0.50 IU/mL and Fibrinogen concentration ≥ 2.0 g/L, with 100% compliance after 1-year storage, and are equivalent to those obtained in routine quality control with INT100. All tested parameters at T5 are within $\pm 25\%$ of T2 baseline values, except Factor VIII (69.2%), TAT (62.3%) and C3a (16.2%, effect of the CAD).

Summary / Conclusions: The process of preparing pathogen reduced plasma with amotosalen and UVA generated by a LED illuminator delivers products that meet the expectations with regard to the quality of therapeutic plasma after frozen storage of at least one year. The LED illuminator brings advances in ergonomics and user interface.

P308 | Introduction of 7-day amotosalen/UVA pathogen reduced platelets in Honduras—impact on platelet availability in a lower middle income country

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Background: The Honduran Red Cross (HRC) produces and distributes $\sim 70\%$ of the platelet concentrates (PCs) transfused in

Honduras, a lower middle income country (LMIC) of approximately 10 million people in Latin America. Honduras became the first LMIC to adopt amotosalen/UVA pathogen reduced (PR) PCs as the standard of care in 2018. HRC produces platelet rich plasma (PRP) PCs derived from whole blood (WB). Prior to 2018 all PCs were screened for bacterial contamination (Bact/Alert) and issued as single, non-pooled, donor PRPs (SD-PRP) with a 5-day shelf-life. The dosing regimen was based on patient weight. The transition to PR PCs was accompanied by a shift to pools of 6-PRPs with a standardized dose of $\geq 3.0 \times 10^{11}$, the elimination of bacterial culture screening, and an extension of PC shelf-life to 7-days. **Aims:** To describe PC production and distribution trends in Honduras over 8 years before and after introduction of PR, standardized pooling, and 7-day shelf-life.

Methods: Annual PC production and distribution data, and WB collection data were retrospectively extracted from HRC records for 2015-2023 and analyzed in 2 periods. Period 1 (P1) included 3 years of SD-PRP doses (2015-17) and the transition year (2018). Period 2 (P2) included 5 years of pooled PR PC doses (2019-23). PC doses were standardized to an equivalent adult dose for both periods: SD-PRP doses in P1 used a 1 SD-PRP per 10 kg dosing formula and a mean adult (male and female) weight estimate of 63 kg; pooled PR PC doses in P2 included 6 SD-PRPs. Descriptive statistics were calculated for 2-period comparisons and multi-year trends. Population data were obtained from the National Institute of Statistics.

Results: HRC produced 10% more PC doses per year on average in P1 compared to P2. Standardization with 7-day shelf-life enabled the

P308 - Table 1: Results

Pooled or 6-pool equivalent PC doses	Year	Produced	Distributed	Est. Waste (%)
P1: SD-PRP + Transition	2015	2.854	1.844	35.4%
	2016	3.283	2.223	32.3%
	2017	2.91	2.166	25.6%
	2018	2.818	2.752	2.4%
P2: Pooled PR PC	2019	3.316	3.278	1.1%
	2020	2.826	2.791	1.2%
	2021	3.914	3.884	0.8%
	2022	3.364	3.33	1.0%
	2023	3.164	3.126	1.2%

distribution of 58% more PC doses per year on average in P2. The mean annual distribution of produced PC doses increased from 64.9% in P1 to 98.9% in P2 (Table). A total of 27,635 adult PC doses were produced by HRC between 2015 and 2023. Of these, 24,579 (88.9%) were distributed to 16 of 18 regions in Honduras (2 regions did not report any PC use in either period). Two regions accounting for ~40% of the population consumed 96% of PC doses in P1 and 88.3% in P2. PC distributions increased in 15/16 regions and declined in 1 region. Six of 16 regions (37.5%) received no or <1 PC dose per year on average in P1; all 16 regions received at least 1 PC dose per year in P2. WB collections increased by ~40%, driven in part by ~13.7% population growth.

Summary / Conclusions: Standardized pooling, PR and shelf-life extension to 7-days allowed HRC to increase distribution of PC doses without requiring additional WB donations. PC distribution is concentrated in urban areas, but increased production permitted additional distribution in some rural areas. Access to PC transfusion remains limited in Honduras (<1 PC dose per 1000 population); however, the conversion to pooled PR PCs illustrates the potential to sustainably expand PC availability in an LMIC through optimization and standardization. Additional research is needed to describe the cost and clinical use of PCs in Honduras.

P309 | Optimization of pathogen-reduced therapeutic plasma and platelet units for pediatric use—a 5-year experience

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Background: Pediatric patients have different requirements for plasma and platelet units with respect to component volume and platelet dose. In a clinical ward, splitting of adult doses to meet pediatric needs is not possible. Furthermore, pediatric patients are vulnerable patients requiring high blood safety standards. To facilitate

pediatric transfusion and increase safety, pathogen-reduced pediatric platelet and plasma units (pooled plasma to increase standardization) were produced and optimized for pediatrician need since 2019 in the Warsaw Regional Blood Transfusion Center, mainly for the Monument Child Health Center, the Mother and Child Institute and the University Clinical Center in Warsaw. Platelet and plasma quantities and dose sizes for children, infants and neonates were adapted according to transfusing pediatricians (TP) needs, with latest adaptations in 2023.

Aims: Production of pathogen-reduced pediatric therapeutic plasma (PTPLS) and platelet (PTPC) units and adaptation to TP needs.

Methods: Plasma was produced from whole-blood donations with the buffy coat method using automated Compomat G5 presses (Fresenius-Kabi, Germany). Five ABO-compatible whole-blood derived single donor plasma units (≥ 270 ml) were pooled using the DONOpack plasma pooling set (LMB Technologies, Germany). Subsequently the pool (≥ 1260 mL) was split into 2 equal minipools (≥ 630 mL) which were treated with amotosalen/UVA pathogen inactivation (INTERCEPT Blood System, Cerus, U.S.A.). Each minipool was split into 6 PTPLS units (between 50 and 280 mL) into plasma storage bags (from INTERCEPT plasma processing sets), the final product was frozen within 8 h of collection (FFP) with a plasma freezer (HOF, Germany). Platelets in 65% PAS (SSP+, Macopharma, France) and 35% plasma were collected by apheresis ($4.3 \pm 0.4 \times 10^{11}$ platelets, 266.4 ± 4.4 mL volume) with an Amicus device (Fresenius-Kabi). After 2 h resting time, platelets were treated with amotosalen/UVA (INTERCEPT Blood System, Cerus). The units were split into PTPC units according to clinician orders (15-150 mL) in pediatric 150 mL platelet storage bags (Ravimed, Poland) and incubated at 20-22°C under continuous agitation.

Results: Initially we started in 2019 (demand 1049 PTPLS units) with a split ratio of 2×50 mL, 1×80 mL, 2×100 mL, and 1×250 mL of plasma. In 2020, the demand increased to 1362 PTPLS units. The split ratio was adapted based on TP feedback to 2×50 mL, 3×100 mL and 1×230 mL. The demand dropped in 2021 back to 1049 units, likely due to a COVID-19 pandemic related reduction of routine procedures. The third TP-feedback based adaption led to a split ratio of 3×50 mL, 2×100 mL and 1×280 mL. In 2022 the demand increased to 2697 released PTPLS units, likely due to restart of routine procedures and additional pediatric wards using our pediatric products. The last adaption was conducted in 2023 to a split ratio of 5×100 mL and 1×130 mL for PTPLS. The demand for PTPLS units dropped to 2007 units 2023. PTPC units were split on demand, 5 mL/kg of body weight (in average 8.1×10^9 platelets per kg). The demand for pediatric PTPC units increased from 3480 (2019) to 3779 (2020). In 2021-2023 it was stable at 4346 ± 20 units annually.

Summary / Conclusions: The demand for pathogen-reduced pediatric plasma and platelet units increased over a 5-year period by increasing adoption of regional hospitals. The ideal unit size of PTPLS to meet clinical needs is, after the 4th adaption by pediatricians, 100 mL.

P310 | Optimization and in-vitro qualification of pathogen reduced double dose pooled platelets of seven buffy coats throughout 8 days of storage

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Background: Platelets carry a greater risk of bacterial contamination than other blood components. Blood establishments worldwide have implemented pathogen reduction technologies, which provide inactivation of a broad range of potentially harmful agents in the platelet concentrates. We describe, for the first time, the *in-vitro* platelet quality assessment of whole blood derived double dose pools, prepared using 7 buffy coats (BCs) pooled on the Terumo TACSI® System, and pathogen reduced (PR) with the INTERCEPT™ Blood System (IBS), throughout 8 days of storage.

Aims: The primary aim was to optimize and validate the preparation and storage of double dose pools of 7 BCs, PR with the IBS and verify that they comply with the EDQM Guidelines 21st Edition. Secondly, to investigate the impact of the IBS photochemical process on the quality and function of pooled platelets compared to conventional untreated platelets.

Methods: A paired case-control study comparing the biochemical and functional differences detected between PR and untreated BCs was carried out. BCs (n = 70), were separated from whole blood by hard-spin centrifugation. Seven ABO-identical BCs were then pooled with 280 ml of additive solution (SSP+), and double dose (DD) pools (n = 10) were produced using the TACSI® System. Then, two DD pools were combined and split to generate 10 paired DD platelet units. Platelet units were treated with either the IBS (n = 5), or were untreated controls (n = 5). All platelet units were then split into two equal single-dose units and stored agitated in the INTERCEPT storage bags for 8 days at 22°C. *In-vitro* quality and function of platelets were assessed throughout storage with routine quality control tests and flow-cytometry at baseline, day 2, day 6 and at expiry on day 8.

Results: All quality parameters tested throughout the optimization and process validation met the IBS processing requirements. The final units complied with the EDQM specifications. Platelet concentration significantly dropped in PR from day 2 storage onwards (p<0.05), while remained constant in the controls. All platelet units met the required specification for platelet concentration on all days tested. No aggregates and good swirling were observed for all units throughout 8 days of storage, with higher swirling scores on day 2 for PR platelets. MPV values were stable and comparable and minimal difference was observed for pCO₂ and pO₂. All units had satisfactory pH values and sufficient glucose reserves at the end of storage. Higher glucose values were detected in IBS treated platelets. CD62P expression and Phosphatidylserine

(PS) exposure significantly increased over storage in both groups, indicating comparable levels of storage lesion and apoptosis. Platelet stimulation by the thrombin agonist TRAP and PS agonist calcium resulted in high levels of responsiveness in both groups at each time point, indicating that PR platelets and standard PCs are capable of haemostatic response and pro-coagulant activity beyond 7 days of storage.

Summary / Conclusions: This study validated an optimal process for the production of 7 buffy coat-double-dose pooled platelets suitable for IBS treatment and provides evidence that *in vitro* platelet quality and function is adequately maintained following IBS treatment and storage in SSP+ for at least 7 days.

P311 | Impact of a 46 hours CAD duration on pathogen reduced buffy coat platelet concentrate double storage quality

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Background: The photochemical process with amotosalen + UVA (IA) (INTERCEPT™, Cerus) has a Compound Adsorption Device (CAD) that allows the reduction of residual amotosalen contained in platelets concentrates (PC) after pathogen inactivation. The maximum duration of this step is 16 hours as determined in the initial validation.

Aims: To evaluate the quality of buffy coat PC-IA treated prepared in double storage (BC-PC-IA DS) and treated with a CAD time of 16hours (h) or 46h.

Methods: Two BC-PC were pooled and split in order to obtain two identical BC-PC-IA DS. BC-PC 1 was treated with a CAD duration of 16 hours (Control = C); BC-PC 2 was treated with a CAD duration of 46 h (CAD46) (n = 6 each condition). The biological parameters (swirling index = SI, platelet content, pH, pO₂, pCO₂, glucose) were measured before pathogen inactivation, at the end of CAD (day (D) 1, 6 for BC-PC C; D2, 8 for BC-PC CAD46) and at D3, D4, D5, D7 et D8 of storage. Soluble p-selectin was measured during the storage (D2-D8) while residual amotosalen was analyzed only at the end of CAD.

Results: The results show no difference between the groups with a CAD duration of 16h vs 46h for all biological parameters. In each group, the residual amotosalen is less than 7µM, even with a lower concentration in the CAD46 group vs C. All units have an optimal SI (+++) during the entire storage duration. The pH of group C is higher on D3 (7.128 ± 0.055) vs CAD46 (6.875 ± 0.088) corresponding to the day of CAD output. On D4, the values of the two groups are again similar until D7 (Group C: 7.047 ± 0.07 vs group CAD46: 6.996 ± 0.058). The evolution of pO₂ and pCO₂ are similar between the Control group and the CAD46 group from the end of the CAD, reflecting comparable gas exchanges during storage. Glucose level is similar in the two groups indicating that 46 h of CAD does not affect glucose consumption. Soluble p-selectin evolves in an equivalent manner between the two groups leading to moderate and equivalent activation throughout the storage period.

Summary / Conclusions: The evaluated parameters of BC-PC-IA DS do not show any difference between a CAD duration of 16h or 46h. The residual amotosalen complies with the French blood products characteristics, no release is observed after 46h of CAD. The platelets do not show distinct signs of activation. These results constitute a decision aid when the duration of CAD exceeds the specified maximum duration of 16h and complements the previous study relating to CP-IA processed in Large Volume kits.

P312 | Development of a preparation method for double dose platelet concentrates obtained from individual platelet units and ready for amotosalen / UVA treatment

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Background: Pathogen reduction (PR) of platelet concentrates allows reducing the risk of transmission of infectious agents due to pooling. This benefit has triggered the development of platelet concentrate (PC) preparation methods with up to 8 buffy coats using amotosalen/UVA (A-UVA) PR treatment (INTERCEPT™ Blood System). Alternatively, an automated whole blood separation technique (Reveos®) delivers Intermediate Platelet Units (IPU) ready for pooling. Increasing the pool size to 7-8 IPU instead of the usual 4-5-IPU can also be considered for the preparation of PR treated double dose PC.

Aims: The objective of the study was to determine the feasibility of pooling 7 or 8 IPU to prepare double dose PC, meeting after PR treatment with A-UVA the specifications outlined in the European (EDQM) and Spanish (CAT) guidelines.

Methods: IPU are obtained from 450 mL $\pm 10\%$ donations on the day of collection and rested for 1 h before overnight storage under controlled agitation. The target volume is calculated to be 28 mL for 7 BC and 25 mL for 8 BC pools to obtain after addition of 250 mL PAS (T-PAS+) a volume lower than 420 mL and a plasma ratio below 47%. Pools of either

7 or 8 IPU are constituted and filtered for leukocyte depletion without delay in a Reveos platelet pooling set. Platelet counts, volumes and plasma ratio are measured to compare with the A-UVA process entry and final PC specifications. In this development phase, IPU not selected for their platelet content are used to determine platelet recovery and extrapolate what would be the minimum Platelet Yield Index (PYI, selected on the Reveos TOME IPU triage tool) to use for meeting the EDQM and CAT specifications particularly for platelet content, respectively $\geq 2.0 \times 10^{11}$ and $\geq 2.4 \times 10^{11}$ in 90% of the PC. The 28 ml IPU's not used for pooling of 7 were used to make simple pools of 5 units.

Results: Table 1 shows key performance indicators of the A-UVA PC preparation process without PYI selection of the IPU. These preliminary results indicate that the EDQM requirement for platelet content could be met in at least 90% of the units (100%) but only in 57% of the units using the CAT requirements with both 7 and 8 IPU processes, not selecting the IPU. The simulation indicates that selecting IPU with a minimum average PYI in the pool of ≥ 60 would allow producing enough double dose PC meeting the requirements from both guidelines and the supply needs. Table 1 also shows that 100 % pools of 7 IPU, 28 ml, PYI > 60, selected met EDMQ requirements and 80% met CAT requirements. All pools in this study met the requirement of $< 1 \times 10^6$ residual leukocytes.

Summary / Conclusions: Both methods of pooling 7 or 8 IPU to obtain platelets compatible with the A-UVA (INTERCEPT) process entry requirements are doable. A selection of IPU with a PYI ≥ 60 would allow meeting the platelet content specifications in the EDQM and CAT guidelines. Pooling 7 BC is the preferred method if confirmed to be suitable in routine because using IPU of a standard volume of 28 mL. In addition, the leukoreduction filter used for the pools is valid for up to 8 units.

P312 - Table 1

	IPU Pool				IBS PLT			Standars	
	IPU Vol PYI	IPU	Potential	PLS	Vol	Yield	PLT Recovery (%)	EDMQ	CAT
POOL 8 IPU N = 14	25.6	74.8	6.0	45.0	194.9	2.4	91%	100 %	57%
POOL 7 IPU N = 9	28.1	75.5	5.3	44	197.4	2.2	94%	100%	12%
POOL 7 IPU > 60 PYI N = 20	27.9	79	5.5	43.9	197.4	2.6	93%	100%	80%

P313 | Abstract withdrawn**P314 | Analysis of pooled platelets preparation after implementation of pathogen reduction technology at a Brazilian service.**

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Background: HEMOMINAS Foundation (HF) is part of the Brazilian Blood System, operates with 21 collection centers and 11 processing units and is responsible for more than 90% of the blood supply in Minas Gerais state. In 2023 HF collected 283,148 blood bags and produced 740,910 components. Pathogen reducing technology (PRT) is not routinely used in Brazil. In 2020 HF assessed the technical and financial viability of PRT. The biggest HF's unit is the Service of Belo Horizonte (HBH), which processes around 90,000 whole blood bags per year and used to prepare pooled platelets (PBC) from 4 or 5 donors by buffy-coat pool. In 2021 HBH implemented PRT but changed procedures in an effort to decrease input costs without compromising platelet quality: using dry buffy-coat instead of wet BC. Dry BC is used to prepare

single pool (5 BC +1 plasma; PBC5) or double pool (8 BC +1 plasma) which produces two units of therapeutic dose corresponding to 4 BC each (PBC4).

Aims: Compare the quality of PBC platelets pre-PRT and post-PRT implementation; assess improvement of the production process.

Methods: Five months pre and post-PRT implementation, PBC were assessed for platelets/unit (pt/un), leucocytes/unit (lc/un) and pH, whereas plasma for volume. In the pre period, 94 PBC4 and 208 PBC5 were assessed, and post-PRT (PR-PBC), 295 PBC4 originated from double pool and 85 PBC5. It was also studied the yield of whole blood (WB) considering the amount of produced PBC.

Results: PBC4 showed significantly different pt/un, lc/un and pH (Table 1), but with greater conformity of pt/un and lc/un in the PR-PBC.

Platelet counts post-PRT increased in PBC4 and were similar in PBC5. Leukocyte counts reduced and pH remained within the reference value in both PBC. Higher platelet yield in PR-PBC may be explained by using dry BC. Plasma volume increased ($p < 0.001$, Mann-Whitney at 5%) between pre-PRT (237.8 ± 17.49 mL, $n = 447$) and post-PRT (256.7 ± 18.47 ; $n = 335$), because plasma extraction was increased to prepare dry BC. Regarding the BC amount needed to produce a therapeutic platelet dose, it was evidenced that the changes in the process increased WB yield. In average, 5.28 BC were used for each produced PBC pre-PRT. It was reduced to 4.64 BC post-PRT, due to the greater production of double pools (91% of produced PBC). Thus, PBC production increased 13.8% post-PRT.

Summary / Conclusions: PRT increases transfusion safety, replaces some processes like bacterial control and irradiation, and allows to extend PBC shelf life to 7 days. The changes in method also resulted in improved WB and platelet yields, reduced leukocyte contamination and greater availability of plasma. Considering this, HF has gradually increased PRT usage in platelets. In 2022, 44% of the prepared platelets were submitted to PRT, increasing to 55% in 2023 and aims to reach 65% of platelets in 2024.

P314 - Table 1: Comparison between PBC4 and PR-PBC4

Variable	Group	Mean \pm SD	P	% standard
Platelets	PBC4	25.10 \pm 4.29	<0.001**	76
($\times 10^{10}$ pt/un)	PR-PBC4	27.67 \pm 3.73		95.3
Leukocytes	PBC4	1.11 \pm 3.30	0.001*	95
($\times 10^6$ lc/un)	PR-PBC4	0.17 \pm 0.48		99.3
pH	PBC4	7.38 \pm 0.14	<0.001*	100
	PR-PBC4	7.24 \pm 0.13		100

*Mann Whitney test at 5%. **t Student's test 5%.

PBC5 showed significantly different lc/un and pH (Table 2), with better leukoreduction after PRT. Platelet counts were similar.

P314 - Table 2: Comparison between PBC5 and PR-PBC5

Variable	Group	Mean \pm SD	p	% standard
Platelets	PBC5	33.68 \pm 5.64	0.712	88
($\times 10^{10}$ pt/un)	PR-PBC5	33.30 \pm 5.09		87
Leukocytes	PBC5	0.20 \pm 0.31	0.005*	98.7
($\times 10^6$ lc/un)	PR-PBC5	0.13 \pm 0.70		99
pH	PBC5	7.32 \pm 0.10	0.002*	100
	PR-PBC5	7.27 \pm 0.12		100

* Mann Whitney Test at 5%.

P315 | Comparison of the quality parameters of irradiated vs pathogen-reduced platelet products

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Background: Currently, different platelet products are produced in Blood Establishments. Starting from whole blood (WB), after preparing the platelet pool, obtained from Buffy-coat (BC), and before releasing it to hospitals, it can be irradiated (PCIR) or treated with pathogen reduction techniques (PCPR). Due to this diversity, it is necessary to know the characteristics of these products because those parameters may influence on the clinical response.

Aims: To know the different attributes of PCIR and PCPR, such as platelet count/microL, total platelet count/unit, platelet

P315 - Table 1. Descriptive study PCIR and PCPR

	Mean \pm Standard Deviation	Median	Min-Max	p
Volume (mL) PCIR (523)	364.60 \pm 12.49	364.00	300.00-421.00	<0.001
Volume (mL) PCPR (311)	184.38 \pm 10.11	184.00	155.00-286.00	<0.001
N° platelets $\times 10^{11}$ /unit PCIR (523)	2.93 \pm 0.41	2.91	2.00-4.07	<0.001
N° platelets $\times 10^{11}$ /unit PCPR (311)	2.56 \pm 0.29	2.54	1.87-3.58	<0.001
Platelets $\times 10^9$ /mL PCIR (523)	0.8 \pm 0.1	0.7	0.5-1.1	<0.001
Platelets $\times 10^9$ /mL PCPR (311)	1.3 \pm 0.15	1.3	1.0-1.7	<0.001
Platelet storage (days) PCIR (523)	3.14 \pm 0.98	3	2-5	<0.001
Platelet storage (days) PCPR (311)	2.84 \pm 1.02	2	2-6	<0.001

P315 - Table 2. Study of categorical variables, swirling.

	PCIR (%)	PCPR (%)	p*
≤ 2	3 (0.6)	0 (0)	0.298
3	520 (99.4)	311 (100)	
Total	523	311	834

* Fisher test.

concentration, volume, swirling, and days of storage at the time of their distribution.

Methods: A total of 523 PCIR and 311 PCPR have been analysed. In the first period, between May and July 2023, the PCIR were produced, and in the second period, between October 2023 and January 2024, the PCPR. BCs were obtained after a fast centrifugation of the WB. PCIRs were obtained by mixing 4 BC with 300 mL of PAS-E (Grifols®) and TACSI automated system (Terumo®) was used. The day after their preparation platelets were irradiated and analysed. PCPR were created by mixing 8 BC and 280 mL of PAS-E (Macopharma®), this prepool was centrifugated at 1650 RPM (G: 850), acceleration: 3, brake: 3, and time: 7 min. Then, platelets were desplasmatized using Macopress Smarter fractionators (Macopharma®), after that, platelets were transferred to the inactivation kits (Cerus®) and treated with amotosalen and UVA. Amotosalen was removed in the compound adsorption device for 14 h, and platelets were split in two products with the same volume. To obtain weight an electronic balance was used, platelet count was determined using Coulter BC-3600 (Mindray®), and the presence of swirling was classified as 0: absent, 1 slight, 2: moderate, 3: strong. A statistical study was carried out using the SPSS program. This study consisted of a descriptive study of the two types of platelet concentrates (mean, median, range, percentiles, and standard deviation), as well as a comparison of the continuous and categorical variables with t Student and Fisher test respectively, a p lower than 0.05 was considered as significant.

Results: They are shown in the attached tables.

Summary / Conclusions: Statistically significant differences were shown in volume, platelet count and concentration and, in the number of days of storage. Despite that, both PCIR and PCPR met all the requirements of the Council of Europe. These differences may not have an influence on clinical response, because of that, a study about

their therapeutic effect will be conducted. Differences in the day of the distribution are significant. PCPR can be delivered sooner and, also, released until day 7.

P316 | Dose-dependent inactivation of *Plasmodium falciparum* in red blood cell concentrates by treatment with short wavelength ultraviolet light

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Background: *Plasmodium* species are blood-borne pathogens that are naturally transmitted by female *Anopheles* mosquitos. The parasite infects red blood cells (RBCs) and can therefore be transfused with blood products. The risk of infection in non-endemic areas is limited to travellers coming back from malaria endemic regions. However, transfusion-transmitted malaria (TTM) constitutes a significant risk of infection in endemic countries. TTM may cause life-threatening complications in patients dependent on blood donations.

Aims: This study investigates the effectivity of *Plasmodium falciparum* inactivation in RBC by treatment with short wavelength ultraviolet (UVC) light in the absence of photochemical additives.

Methods: RBC suspended in the new developed additive solution PAGGS-C were diluted to a haematocrit of approximately 30% before UVC treatment. RBC (n = 3) were spiked with *Plasmodium falciparum* to a final parasitemia of 0.1-1% and were irradiated with up to 4.5 J/cm² UVC (254 nm) under agitation (300 rpm). Samples were taken at different time points and the parasitemia was determined by serial dilutions followed by either by GIEMSA staining or flow cytometry over three weeks post irradiation treatment.

Results: The lowest dose of 1.5 J/cm² UVC led to a ≥ 3 log unit reduction in parasite load compared to the untreated control. The inactivation capacity was shown to be dose-dependent. Strikingly, 4.5 J/cm² led to ≥ 5 log unit reduction, which was equivalent to a complete inactivation in two out of three experiments.

Summary / Conclusions: Additive-free pathogen reduction with UVC light was previously shown to be effective for different bacteria and viruses, but the inactivation of parasites was not addressed until now. The present study provides evidence for significant inactivation of *P. falciparum*-infected RBCs by UVC light.

P317 | The impact of pathogen reduction technology for platelets on the incidence of transfusion reactions — a single center study

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Introduction: The incidence of platelet-related non-hemolytic transfusion reactions (TRs) is considered 10%-30% per transfusion (allergic 1%-2%), ranging from mild to severe, the incidence of septic transfusion reactions (STRs) is lower (approx. 1:10,000 per transfusion), but often severe to lethal. Prevention of STRs and a significant reduction of (allergic) TRs (between 26% and 66% of general TRs in Switzerland and 52% of platelet-related TRs in Strasbourg) was reported as consequence of treating the platelets with pathogen reduction technology (PRT). The reduction of TRs by PRT was mainly due to efficient inactivation of contaminating white blood cells in the platelet unit (while the additional implementation of PAS contributed to the reduction of TRs as well, mainly due to antibody and plasma factors depletion). We implemented PRT for platelets in 2022 to improve the clinical outcome and well-being of patients.

Aims: Assessment of the impact of the introduction of PRT for platelets on the incidence of transfusion reactions.

Material & Methods: Platelets in 100% plasma were collected at the King Fahd Medical City (KFMC) Blood Center (Riyadh, KSA) either by apheresis (Trima, Terumo-BCT, USA, and MCS+, Haemonetics, USA) or from whole blood donations (processed with an automated Reveos device, Terumo-BCT). 5 Reveos IPU's were pooled for one adult transfusion dose ($\geq 3 \times 10^{11}$ platelets/unit). Apheresis platelets were treated with amotosalen/UVA PRT (INTERCEPT Blood System, Cerus Corp, U.S.A) from 02/2022, whole-blood derived platelets from 09/2022. In 2023, approx. 50% of platelets issued were pathogen-reduced. The incidence of transfusion reactions was collected by an in-house passive reporting system. Data was analyzed in 3 periods, P1 (2021, conventional platelets); P2 (2022, transition to 50% PR platelets), P3 (2023, 50% PR platelets).

Results: In P1 33,907 (27.2% platelets), in P2 33,921 (23.6% platelets) and in P3 37,193 (22.1% platelets) blood components were transfused at the KFMC in total. The number of RBCs transfused per platelet transfusion was in P1 1.93, in P2 2.25, and in P3 2.33. The incidence of general transfusion reactions (all components per 1000 transfusions) was steadily declining during the study period (P1 1.18, P2 1.03, P3 0.81). The rate of platelet-related transfusion reactions per 1000 platelet transfusions (absolute numbers between 11 and 15 per year) was 1.19 in P1, 1.87 in P2 and 1.46 in P3. The rate of total allergic transfusion reactions (absolute number between 13 and 30) per 1000 transfusions decreased

between P2 and P3 of 47% (0.71 in P1, 0.6 in P2 and 0.32 in P3). The rate of non-hemolytic transfusion reactions did not change significantly (0.38 in P1, 0.44 in P2, 0.32 in P3), the incidence of other transfusion reactions was too low for a meaningful analysis.

Summary / Conclusions: Our preliminary data points towards a reduction of allergic transfusion reactions (47%) after partial implementation of PRT for platelets as described previously. The number of RBC transfusions per platelet transfusion, as a surrogate marker for bleeding, was not increased.

P318 | Pathogen inactivation in plasma and platelet concentrate in Polish Blood Establishments (BEs) in 2020–2022

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Background: In 2009, pursuant to the Health Policy Program of the Minister of Health, the Mirasol pathogen inactivation system, for plasma and then also for platelet concentrates (PCs) was implemented in 9 of 23 Blood Establishments (BEs). In the years that followed most Polish BEs had implemented some system of pathogen inactivation, and by the outbreak of the pandemic in 2020, all BEs had at least 1 inactivation system installed. Currently in Poland there are 2 pathogen inactivation systems for plasma and PC: Mirasol PRT and Intercept, and Theraflex MB Plasma system for pathogen inactivation in plasma. One of the tasks of the Institute of Hematology and Transfusion Medicine (IHTM) as the BE supervising unit is to monitor the status of implementation of pathogen inactivation systems in BEs.

Aims: The study aim was to estimate the extent of use of pathogen inactivation systems in Polish BEs in the period 2020–2022.

Methods: The analysis of the percentage of inactivated plasma and PC units was based on data from annual BE reports.

Results: in 2020, 16 Mirasol systems, 4 Intercept systems and 10 Theraflex MB Plasma systems were installed at BEs. In 2022, the number of Mirasol systems increased to 18, and as of 2021, the number of Theraflex MB Plasma systems in BEs increased to 12. In 2022, only 2 Intercept systems were used. In the following years, 0.02% to 12.5% (2020), 0.02% to 15.91% (2021) and 0.11% to 17.51% (2022) of plasma was subjected to inactivation. On the national level, 3.01% (in 2020), 3.71% (in 2021) and 4.69% (in 2022) of all plasma was inactivated. Plasma collected from COVID-19 convalescents was also subjected to inactivation: 27.93%–100% in 2020 (nationwide 63.36%), 24.64%–100% in 2021 (nationwide 66.9%) and 58.96%–100% in 2022 (nationwide 43.07%). The following were issued for clinical use: 14.21% of inactivated FFP and 6.89% of inactivated cryoprecipitate in 2020, 15.64% of inactivated FFP and 10.47% of inactivated cryoprecipitate in 2021, 24.73% of inactivated FFP and 9.48% of inactivated cryoprecipitate in 2022. In 2020, 0.04%–100.00% of pooled PC (nationwide 5.18%) and 0.05%–98.13% of apheresis PC (nationwide 26.19%) were inactivated. A total of 11.01% of inactivated PC units were issued for clinical use in 2020. In 2021, 0.1%–

99.21% of pooled PC (nationwide 5.38%) and 0.61%-98.53% of apheresis PC units (nationwide 27.27%) were inactivated. For clinical purposes, a total of 12.53% of inactivated PC units were issued in 2021. In 2022, 0.31%-99.59% of pooled PC (nationwide, 5.38%) and 0.68%-96.40% of apheresis PCs (nationwide 24.34%) were inactivated. In 2022, altogether 12.70% of inactivated PC units were issued for clinical purposes.

Summary / Conclusions: In the 2020-2022 period, there was a slight nationwide increase in the percentage of inactivated FFP. Significant disparities were however recorded as regards the use of inactivation systems in individual BEs. The percentage of annually inactivated PCs did not change.

P319 | Abstract withdrawn

Blood products / components— novel blood products/ components

P320 | The quality of red blood cells isolated from cord blood during storage

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Background: Anemia is an often occurring problem in premature neonates. Standardized treatment is a leukocyte-depleted red cell concentrates (RCC) from adult whole blood, which contains >95% HbA, as opposed to cord blood-derived red cells, which contains >90% HbF. HbA has a lower oxygen affinity than HbF, therefore transfusion with HbA potentially results in excessive oxygen delivery, which may contribute to the onset of retinopathy of prematurity.

Aims: The aim of this study is to optimize the storage conditions such that CB-RCC can be stored while maintaining adequate in vitro quality.

Methods: Cord blood was processed within 24 hours after collection and was leukocyte and platelet depleted by a whole blood filter and

centrifuged to replace the plasma with additive solutions SAGM (n = 9), PAGGSM (n = 6) or SOL-X (n = 8). A volume of 15 ± 1 ml of concentrated red cells was diluted in additive solution in a 100 ml DINCH-PVC bag to obtain a hematocrit of 50%-65% and an end-volume of 20 ± 2 ml. The units were stored for 21 days at 4°C. Hematological and metabolic quality parameters were determined on days 1, 7, 14 and 21.

Results: Storing CB-RCC in SOL-X or PAGGSM resulted in significantly lower levels of hemolysis than storage in SAGM (0.4 ± 0.1 % and 0.3 ± 0.1 % vs. 0.6 ± 0.2 %; on day 21 of storage). CB-RCCs stored in SOL-X or PAGGSM also swell significantly less during storage than in SAGM as shown in MCV (122 ± 7 fl and 121 ± 5 fl vs. 132 ± 6 fl). When stored in SOL-X the pH and internal pH is significantly higher on day one and during the rest of storage. The percentage of undeformable cells on day 21 is also significantly higher when stored in SOL-X (6.1 ± 3.0% in SAGM, 10.8 ± 2.6% in PAGGSM vs. 17.6 ± 2.3% in SOL-X). The glucose/ lactate ratio on day 21 is significantly better in PAGGSM when compared with SAGM (1.79 ± 0.75 vs. 0.44 ± 0.10).

Summary / Conclusions: CB can successfully be processed and stored in DINCH-PVC bag in SOL-X or PAGGSM with low levels of hemolysis up until day 21. However storage of CB-RCC in PAGGSM has the preference above storage in SOL-X because of a lower percentage of undeformable cells and a better glucose/lactate ratio. Furthermore PAGGSM is already commonly used in Europe and CE-marked, these data provide a basis for future clinical evaluation on the efficaciousness of the use of CB-RCC in premature neonatal care and the onset of retinopathy of prematurity.

P321 | Quality comparison of red blood cell concentrates between DEHT-PAGGSM bags and DEHT-SAGM bags 49 days storage study

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Background: The use of di(2-ethylhexyl)-phthalate (DEHP) has long been the plasticizer in polyvinyl chloride (PVC) whole blood collection sets. However, studies data suggested its potential toxicity as endocrine disruptors and human carcinogens that have led to regulatory changes being introduced in the European Union banning the use of this plasticizer in medical devices. The DEHP is also known for its protector effect

P321 - Table 1

Parameter	Additive solution	D1	D35	D49
ATP (μmol/g Hb)	PAGGSM	4.37 ± 1.06	3.56 ± 0.71	2.50 ± 0.52
	SAGM	4.06 ± 1.04	2.86 ± 0.67*	1.85 ± 0.54*
Haemolysis (%)	PAGGSM	0.073 ± 0.013	0.179 ± 0.062	0.320 ± 0.115
	SAGM	0.065 ± 0.007	0.304 ± 0.085*	0.590 ± 0.206*

Results are expressed as mean ± standard deviation.

* Significant difference between the 2 storage solutions (p < 0.05).

and stabilizer of the red blood cell (RBC) membrane during their storage and its ban leads to test new combinations of DEHP-free plasticizers/storage solutions to maintain the same current quality of RBC.

Aims: Assess the RBC quality stored in di(2-ethylhexyl)-terephthalate DEHT (Transfufol DEHT 3126 from RENOLIT Nederland B.V.)/PAGGSM (Phosphate Adenine Glucose Guanosine Saline Mannitol) compared to RBC in DEHT (Transfufol DEHT 3126 from RENOLIT Nederland B.V.)/SAGM (Saline Adenine Glucose Mannitol) until 49 storage days.

Methods: 16 RBC were prepared from non-therapeutic whole blood (WB) collected with DEHT/PAGGSM bags: with 'Top and Bottom' (TB) kit + filter RBC (LCRD2). 16 another RBC were prepared with DEHT/SAGM bags with the same configuration.

Results: There is a significant difference on haemolysis from day 21 between SAGM and PAGGSM and a significant difference for ATP from day 35. The T&B/PAGGSM kit is the combination giving the lowest haemolysis rate ($p < 0.001$). The two storage solutions have the same behaviour for the glucose, lactate and potassium.

Summary / Conclusions: The storage solution PAGGSM allows to counterbalance the RBC stability loss due to the plasticizer change. This new association DEHT/PAGGSM able to keep haemolysis conformity until 49 days.

P322 | Neuroprotective effects of extracellular vesicles from platelet concentrates in traumatic brain injury and Parkinson's disease models

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Background: Platelets contain a complex array of growth factors, anti-inflammatory agents, cytokines, neurotransmitters, and antioxidants. Recent preclinical research has highlighted the therapeutic potential of human platelet lysate (HPL) in regenerative medicine and, in particular, in treating a spectrum of central nervous system (CNS) neurodegenerative disorders and trauma. Platelet-derived extracellular vesicles (PEVs) found abundantly in HPLs and could play a specific role in neuroprotective effects that still need to be determined.

Aims: Our study aims to reveal the composition of PEVs isolated from PC supernatants and assess their potential in aiding the recovery of damaged neurons and providing neuroprotection in cellular and animal models of traumatic brain injury (TBI) and Parkinson's disease (PD).

Methods: We obtained outdated apheresis clinical-grade PCs from the Taipei Blood Center (Guandu, Taiwan). The PCs were centrifuged ($3,000 \times g$, 30 min) to pelletize the platelets. The plasma supernatant was recovered and centrifuged ($6,000 \times g$, 10 min) to eliminate cellular debris and PEVs were isolated by using high-speed centrifugation ($25,000 \times g$, 90 min) at 18°C and resuspended in PBS. PEVs were characterized using biophysical, biochemical, microscopic, and LC-MS/MS proteomics techniques to elucidate their biological properties. Their functionality was assessed *in vitro* using SH-SY5Y neuronal cells, LUHMES dopaminergic neurons, and BV2 microglial cells. *In vivo*, PEVs were administered intranasally (i.n) a CCI (controlled cortical impact)-TBI mouse model, for their capacity to reduce neuroinflammation. RT-qPCR was used to evaluate the levels of inflammatory markers (*Gfap*, *Cd68*, *Trem2*, *Ccl4*, *Tlr2*, *Tnf- α*). Their neuroprotective activity was also evaluated in an MPTP mouse model of PD through an actimetry behaviour test and immunohistochemical analysis of tyrosine hydroxylase (TH)-positive cell expression in substantia nigra (SN).

Results: PEVs had a predominant size of approximately 200 nm, ranging from 50 to 350 nm, and were obtained at concentrations between 10^{10} and 10^{11} /mL. They expressed platelet and EVs markers, and displayed a lipid bilayer structure, as observed using cryo-electron microscopy. Notably, there was a reduced expression of pro-coagulant phosphatidylserine. LC-MS/MS revealed a diverse composition of trophic factors within PEVs, encompassing neurotrophins, anti-inflammatory agents, neurotransmitters, and antioxidants, suggesting multifaceted biological functions. PEVs facilitated neurorestoration and maturation *in vitro* in SH-SY5Y cells, exhibited neuroprotective activity against erastin-induced ferroptosis in LUHMES cells, and promoted anti-inflammatory responses, particularly evident under inflammatory conditions, in microglial cells. *In vivo* experiments showed the therapeutic potential of i.n delivered PEVs, evidenced by robust anti-inflammatory effects in a TBI mouse model, with significant reductions in *Gfap* and *Tnf- α* levels. Additionally, they improved motor function, as demonstrated in rearing behaviour tests, and preserved TH expression by dopaminergic neurons in the SN of MPTP-treated mice.

Summary / Conclusions: Our data underscore the neuroprotective potential of PEVs isolated from PC supernatant as a biotherapy of the CNS. These findings open new possibilities for treating brain trauma and neurodegenerative disorders, supporting further translational research.

P323 | Restoration of actin network dynamics as a possible key to efficient *in vitro* production of erythrocytes

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Background: An erythroid progenitor cell line capable of efficient *in vitro* production of mature red blood cells (RBCs) represents a promising alternative for traditional red blood cell concentrates in transfusion medicine. Since an erythroblast cell line (imBMEP), previously established in our laboratory, showed limitations in efficient maturation, a knockout (K.O.) of the enucleation inhibitor miR-30a-5p was performed. Although this modification led to a significant promotion of terminal erythropoiesis, insufficient enucleation remains a major challenge.

Aims: Through a comparative transcriptome analysis of imBMEPs and hematopoietic stem cells (HSCs), we aimed to systematically investigate possible causes for the inhibited final maturation and to uncover key candidates to improve enucleation efficiency.

Methods: Samples were obtained from imBMEP cells and HSCs, acting as control, at different time points during erythroid differentiation in biological replicates. Extracted RNA was used for transcriptome analysis using next-generation sequencing. After read alignment to a human reference genome and quality control, differential gene expression (DGE) analysis was carried out. Gene expression data were further examined via Ingenuity Pathway Analysis (IPA) in order to identify altered pathways and biological functions between imBMEP cells and HSCs. To further investigate the functionality of the cytoskeleton, actin, α -tubulin and vimentin were analyzed at protein level via Western Blot. The distribution of the three proteins within the cells was visualized using immunofluorescence.

Results: DGE analysis revealed high variance between imBMEP cells and HSC controls. Using IPA, these differences in gene expression were assigned to specific pathways and functions. A notable number of significantly altered pathways were associated with the cytoskeleton and the actin network. In particular, actin polymerization appeared to be inhibited at several levels within the regulatory networks. While Western Blot data of actin, α -tubulin and vimentin are showing no quantitative differences in protein expression between imBMEPs and HSCs, the immunofluorescence analysis of the cells showed distinct structural changes. The actin network in particular appears to be affected, with actin forming unstructured accumulations within the imBMEP cells instead of a homogeneous network as it was seen in HSCs.

Summary / Conclusions: RNA Sequencing is a powerful tool for analysing differential gene expression. Analysis and comparison of RNA sequencing data between imBMEP cells and HSCs gave first insights in dysregulated processes and possible candidates causing the differences in differentiation behaviour. Using IPA, regulation of the

cytoskeleton, in particular the actin network, was identified as altered. Analysis of cytoskeletal elements at the protein level confirmed this finding. The observed structural changes indicate that primarily actin polymerization and reorganization seem to be affected. With regard to terminal erythropoiesis, this could be a reason for the limited enucleation of the cell line, as the actin network plays a decisive role in this process. It is now necessary to identify key candidates that are responsible for the observed differences and whose modification leads to a restoration of the actin network dynamics.

P324 | A prospective single-center study of the efficacy and safety of using cord blood lysate-derived platelet concentrate eye drops for the treatment of severe ocular surface diseases

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Background: Ocular surface diseases, like chronic Graft Versus Host Disease (cGVHD) and Sjögren Syndrome, cause considerable patient distress and may have an adverse impact on long-term quality of life. There are reasonable evidences for efficacy of autologous and allogenic eye drops of human blood origin in the treatment of ocular surface disease refractory to conventional treatment exists.

New clinical trials conducted in recent years are aiming to investigate the potential therapeutic effect of topical blood-derived products like Umbilical Cord Blood (UCB). This product provides the ocular surface with a high concentration of growth factors and anti-inflammatory molecules, so it is suitable for managing chronic dry eye inflammation. Collection from an allogeneic source and preparation according to an optimised procedure resulted in eye drops derived from platelet pools standardised by platelet and cellular number, safety for infectious disease markers (HIV, HBV, HCV, and syphilis), sterility testing, (presence of aerobic and anaerobic bacteria and fungi) and characterised by blood group. All patients received the same batch of product for the entire treatment period (120 days).

Aims: We conducted a single center prospective study to assess the efficacy and safety of a lysed platelet concentrate (CP_{lys}) derived from UCB in patients suffering from ocular cGVHD after allogeneic transplant and Sjögren Syndrome. Primary objectives were 1) the evaluation of change in corneal damage in patients with moderate/severe keratoconjunctivitis (via Oxford score) and 2) the evaluation of improvement in the extent of damage to the corneal epithelium and post-treatment lacrimation (through Schirmer test and BUT test). Secondary objective is the evaluation of change in dry-eye symptoms, pain and photophobia affecting ADLs (via OSDI score and VAS).

Methods: Twenty-five cGVHD and nine Sjögren Syndrome patients with moderate/severe Keratoconjunctivitis were enrolled in a registered clinical trial (Clinical Trial Registry: CPCB19 v.1.0 - 08.04.2019). The treatment lasted 4 months with a dosage of one drop per eye 3-6

times a day. Upon enrollment and at 1-2-4 months of treatment, we assessed both parameters of ocular surface and symptoms.

Results: Thirty-one of 34 patients completed the study. After 120 days of treatment, comparing the VAS score, we registered substantial improvements, especially in patients' quality of life: at T0 the mean value was 6.53 (range 2-10), while at T4 it was 2.4 (range 0-8). The OSDI score is consistent with these results, going from a mean value of 68.9 at T0 to 25.63 at T4 (scale of 0 to 100). Slighter objective differences were measured with T. Schirmer and S-BUT. However, Oxford scheme witnessed an objective improvement in the corneal damage: all patients enrolled had at least a grade 2 at T0, while, at T4, most of them reached a grade 0 and only 5 had a grade 1.

Focusing on the 25 patients with cGVHD, at T0, according to NIH score, 4 had a score 1 ocular cGVHD, 20 had a score 2, and 1 had a score 3; at T4, 2 had a grade 0, 19 had a score 1, and 2 had a score 2 (78.26% are improved 8.7% reached a score 0).

Summary / Conclusions: According to the results of our study, CP_{lys} derived from UCB eye drops represent a promising therapeutic approach in the healing of Severe Dry-eye in Keratoconjunctivitis Sicca and patients' subjective symptoms relief.

P325 | Cold-storage of amotosalen-UVA pathogen-reduced buffy-coat platelet concentrates for up to 21 days - biochemical and functional characterization, and identification of platelet subpopulations

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Background: Current platelet transfusion requirements are evolving rapidly, with a significant increase in the need for therapeutic platelet transfusions compared to prophylactic transfusions. In this context, cold-stored platelets have gained interest owing to their specific potentially advantageous characteristics, which however require thorough *in vitro* characterization during storage.

Aims: To assess the effects of cold storage on the quality of buffy-coat (BC) platelet concentrates (PCs) treated with amotosalen-UVA

pathogen reduction (PR) and stored in PAS-E (SSP+) additive solution for up to 21 days.

Methods: A pool-and-split strategy was used to obtain double-dose BC-PCs collected into PAS-E/plasma (55/45) treated with amotosalen-UVA and stored at (i) 22°C with constant agitation or (ii) 4°C without agitation.

Results: Platelet counts declined similarly in both groups of PCs during storage without appearance of macroscopic aggregates, while platelet swirling was lost in PCs stored at 4°C, with platelets having a spherical shape as visualized by scanning electron microscopy. Storage at 4°C resulted in a significant reduction in glucose consumption and lactate generation as compared to storage at 22°C as of day 7, and a progressive decrease in pH, which however remained above 6.5 at day 21. Notably, sufficient glucose was still available on day 14 in PCs stored at 4°C, unlike in PCs stored at 22°C. Multicolor flow cytometry analysis for markers of platelet activation, apoptosis and mitochondrial transmembrane potential (MTP) revealed emergence of several platelet subpopulations. Among these, the resting, activated, aggregatory, procoagulant, apoptotic and senescent platelets are of prime interest for a better understanding of the functional properties of platelets. Spontaneous exposure of P-selectin, a marker of α -granule secretion, and of phosphatidylserine, a marker for platelet activation and apoptosis evaluated by annexin V binding, were significantly increased in PCs stored at 4°C as compared to 22°C during storage. MTP, evaluated using the TMRM fluorescent dye retained in functional mitochondria, decreased more rapidly in PCs stored at 4°C as compared to 22°C. The subpopulation of resting platelets remained predominant on day 7 at 22°C, while at 4°C this subpopulation was replaced by an equal proportion of procoagulant and apoptotic platelets, the latter becoming dominant on day 21 (Table). Finally, the ability of platelets to form thrombi on collagen in a microfluidic chamber was conserved until day 14 at 4°C but only until day 7 at 22°C.

Summary / Conclusions: Amotosalen-UVA PR BC-PCs stored at 4°C displayed preserved metabolism, increased spontaneous activation and apoptosis, and preserved *in vitro* platelet adhesive properties for at least 14 days. Further experiments are aimed at better understanding the characteristics and functions of the various platelet subpopulations, which may lead to the development of new or improved platelet products to enhance inventories and access to platelet hemostatic support for bleeding patients.

P325 - Table 1

	Day 1		Day 7		Day 14		Day 21	
Temperature (°C)	22	4	22	4	22	4	22	4
Resting %	84 ± 1	-	64 ± 1	13 ± 5	27 ± 1	3 ± 1	3 ± 1	5 ± 5
Activated %	9 ± 0	-	29 ± 2	31 ± 14	18 ± 5	8 ± 7	3 ± 2	8 ± 9
Aggregatory %	0	-	0	2 ± 2	2 ± 1	2 ± 0	2 ± 1	1 ± 0
Procoagulant %	0	-	1 ± 0	16 ± 6	4 ± 1	23 ± 2	1 ± 0	9 ± 2
Apoptotic %	5 ± 2	-	3 ± 1	20 ± 17	33 ± 2	36 ± 1	50 ± 5	48 ± 19
Senescent %	0	-	2 ± 0	2 ± 1	5 ± 3	8 ± 3	12 ± 1	18 ± 4
Undefined %	1 ± 0	-	2 ± 0	15 ± 3	11 ± 3	21 ± 1	29 ± 5	12 ± 3

P326 | Platelet extracellular vesicle membranes-coated mesoporous silica nanoparticles as a drug delivery carrier for the treatment of brain disorders

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Background: The use of platelet extracellular vesicle membranes (P-EVMs), obtained from human platelet concentrates (PCs), in medical applications is paving the way for targeted drug delivery and regenerative medicine. Sharing the same membrane antigens as their parent platelets, such as CD62P and CD41, P-EVMs play a key role in intracellular signaling pathways and can be recognized by various target cells in different diseases such as brain disorders. Among them, brain cancers are known for their poor prognosis with glioblastoma (GBM) being the most aggressive type. Conventional therapy, with temozolomide (TMZ) remains the standard therapy for GBM. However, the presence of the blood-brain barrier (BBB) limits its access to the brain. Nanotechnology enables combination therapies using theragnostic nanoparticles (NPs) to address these challenges. Thus, we developed TMZ-loaded P-EVMs-coated mesoporous silica nanoparticles (MSNs), a compatible drug carrier based on MSN core structure with a high porous surface area enabling high drug loading and controlled release. To control TMZ release, enhance biocompatibility, and avoid quick clearance, TMZ-loaded MSNs surface functionalized with P-EVMs isolated from platelets may become a potent delivery system. P-EVMs coating may confer the nanocarriers with platelet biomimetic properties providing tailored drug delivery, controlled release and enhanced circulation.

Aims: Enhancing the GBM-targeting efficacy and drug release of TMZ-loaded MSNs by coating with p-EVMs isolated from human (PCs).

Methods: Clinical-grade apheresis PCs were obtained from healthy donors at the Taipei Blood Center, Taiwan. Serum-converted platelet lysate (SCPL) was prepared from PCs through platelet activation by calcium chloride and glass beads treatment followed by differential centrifugation. SCPL-derived P-EVMs were then isolated using qEV Gen2(35nm) size exclusion chromatography columns. MSNs were synthesized by a sol-gel process, conjugated with Rhodamine B dye and functionalized with amine trimethylammonium (TA) group and polyethylene glycol (PEG), to enable subsequent surface coating with P-EVMs, with TMZ loading carried out through adsorption. Finally, the NPs were coated with P-EVMs by extrusion. These drug-loaded nanocarriers were characterized by dynamic light scatter (DLS), nanoparticle tracking analysis (NTA), UV-Vis spectrophotometer, TEM, and cryo-EM. Membrane surface markers were determined by western blot.

Results: The synthesized MSNs were 25 nm by DLS. Functionalization with TA and PEG increased the size of the Rhodamine B dye-conjugated MSNs (RMSNPEGTA) to 39 nm with a positive charge of 17 mV, confirming the successful synthesis of RMSNPEGTA. The P-EVs suspension contained 1.48×10^{12} P-EVMs/ml, and was used at

a total protein concentration of 1mg/ml for MSN coating. The P-EVMs-coated MSNs showed a spherical core-shell configuration by TEM. Western blot analysis confirmed the presence of CD41 and CD62P typical platelet membrane surface markers on the P-EVMs-coated MSNs, consistent with a successful coating. MSNs, prior to TMZ loading, exhibited no cytotoxicity on GBM cell lines, as expected.

Summary / Conclusions: The successful coating of MSNs by P-EVMs evidenced by TEM and by the expression of specific surface markers CD41 and CD62P supports the feasibility of this coating methodology. Further pre-clinical work is needed to demonstrate the safety and efficacy of such a P-EVMs coating platform of MSN for delivery of drugs to the central nervous system to treat GBM.

P327 | Intranasal Heat-treated Platelet Pellet Lysate (HPPL) - impact on hippocampal protein expression in aging

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Background: There is an increasing interest in using preparations derived from outdated platelets for multiple applications in regenerative medicine, including for modulating the impact of brain disorders and trauma. Our laboratory has shown that a purified heat-treated platelet pellet lysate (HPPL), when administered intranasally to the brain, a mode of delivery that by-passes the blood brain barrier, could diffuse and provide neuroprotective effects in various preclinical models of neurodegeneration. Understanding better how intranasal HPPL diffuses into the brain and particularly to the hippocampus, the center of memory and learning, can be extremely important for mitigating memory loss and cognitive decline associated with aging. Proteomics, which allows a comprehensive analysis of protein expression in specific parts of the brain, could be a valuable tool for such objectives.

Aims: To investigate the diffusion and impact of intranasally administered HPPL on hippocampal protein expression in 18-month-old mice.

Methods: Outdated apheresis platelet concentrates (PCs) were obtained from the Taipei Blood Center (Guandu, Taiwan). HPPL was produced by centrifuging PCs at 3,000g for 30 min to remove the plasma. After three freeze-thaw cycles to lyse the platelets, the lysate was heat-treated for 30 minutes at 56°C and then centrifuged at 10,000g for 15 min to obtain HPPL. Mice (18-month old) received HPPL (or PBS control) intranasally for 7 days. At the end of the treatment, the hippocampal proteins were extracted, digested and desalted before being submitted to an Orbitrap Fusion™ Tribrid™ mass

spectrometer (Thermo Fisher Scientific, San Jose, CA). Differently expressed proteins were defined as proteins with a 1.2-fold change, $p < 0.05$ between HPPL-treated mice and controls. Bioinformatic tools were used to construct diagrams, heat maps and functional annotation.

Results: There were slight differences in the proteome of HPPL-treated mice. Particularly, 49 hippocampal proteins were expressed differently in 2693 quantified proteins in HPPL-treated mice compared to 2659 quantified proteins in PBS controls. HPPL treatment increased proteins associated with nucleosome assembly and downregulated some involved in the cell cycle. One might hypothesize that when the cell cycle duration is shortened, progenitor cells cease proliferation and transition to a differentiation phase, ultimately becoming mature neurons. With enhanced genomic stability through nucleosome assembly, this could result in a more organized and synchronized process of neuronal differentiation and maturation. Consequently, these processes might regulate neurogenesis and produce more mature and functionally integrated neurons, which could be beneficial for the repair of damaged tissue and the preservation of hippocampus function in aging.

Summary / Conclusions: One-week intranasal administration of HPPL modulates protein expression in the hippocampus of old mice. Changes in nucleosome assembly and cell cycle processes, suggest mechanisms that could lead to enhanced neuronal differentiation and integration, potentially supporting cognitive functions in aging. While these preliminary findings are suggesting a therapeutic benefit of HPPL for neurodegenerative conditions associated with aging, future studies with longer-term administration involving behavioral assessments and detailed molecular analyses by transcriptomics in established models of aging are needed to delineate HPPL's role in neuroregeneration and cognitive preservation.

P328 | Clinical evaluation of red cell concentrates collected and stored in non-DEHP plasticized bags compared to usual DEHP plasticized bags - a quasi-randomized controlled multicenter surveillance trial

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P328 - Table 1

	PAGGSM/BTHC TTR (95% CI)	SAGM/DEHP TTR (95% CI)	OR (95% CI)
Study 1 (n = 7312)	0.24 (0.00–0.48)	0.44 (0.27–0.61)	0.55 (0.24–1.65)
Study 2 (n = 32878)	0.35 (0.22–0.49)	0.20 (0.15–0.26)	1.76 (1.10–2.81)
Study 1 + 2 (n = 40190)	0.33 (0.22–0.45)	0.24 (0.19–0.30)	1.36 (0.90–2.08)

Abbreviations: n = Total number of products issued; TRR (95% CI) = Transfusion Reaction Rate per 100 transfusions (95% Confidence Interval); OR (95% CI) = Odds Ratio (95% Credibility Interval).

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Background: Di-ethyl-hexyl-phthalate (DEHP) is currently the main plasticizer used for whole blood collection systems. By order of the European Commission the use of DHEP will end July 1st 2030. In view of the continued supply of blood components, it is instrumental to assess DEHP-alternatives in blood bag systems.

In a recently published hemovigilance pilot study in two Dutch hospitals, it was found that transfusions of red blood cell concentrates (RCC) collected and stored in non-DEHP blood bags with storage solution PAGGSM did not result in increased transfusion reaction frequency. In the current follow-up study we included a larger number of transfusions with RCC in PAGGSM/non-DEHP and transfusions using traditional DEHP bags, to compare the frequency of transfusion reactions.

Aims: The follow-up haemovigilance surveillance aimed to clinically evaluate the effect of non-DEHP storage of RCC on transfusion reaction frequency.

Methods: In this quasi-randomized controlled multicenter surveillance, recipients of non-irradiated RCC of at least 18 years old were included, in eight hospitals in the Netherlands. Recipients received RCC stored in PAGGSM/BTHC or RCC stored in SAGM/DEHP as standard treatment. The primary outcome was transfusion reaction frequency. The odds ratio was expressed by a 95% credibility interval obtained from a Bayesian analysis. In a secondary analysis, the data from this study was pooled with data of the earlier study and the odds ratio of transfusion reactions, along with a 95% credibility interval, was re-calculated.

Transfusion reactions were further classified by category, seriousness and imputability. User issues were registered and evaluated.

Results: During the study period, a total of 32878 RCC were issued to 10220 patients. Of all patients, 78 developed a transfusion reaction. Twenty-six reactions occurred after a total of 7507 PAGGSM/BTHC transfusions and 51 after 25371 SAGM/DEHP RCC transfusions, yielding a transfusion reaction frequency of 0.35% and 0.20% respectively (odds ratio 1.76, 95% CI (1.10-2.81)). One reaction occurred after receiving both types of RCC within 24 hours. When pooling the data of both studies, the odds ratio was found to be 1.36 (95% CI 0.90-2.08). Most (97%) reactions were mild to moderate (grade 1 or 2), with low imputability (82%) in both groups. Mild non-haemolytic transfusion reactions and fever were the most common (65%). No user issues, both in clinic as well as during production of the blood components, were reported.

Summary / Conclusions: In study 1 a decrease in transfusion reaction frequency in the non-DEHP group was found whereas in study 2 an increase was established, with the aggregate data being consistent with a range of -10% to 108%. Furthermore, as transfusion reactions were often associated with low imputability, it is uncertain whether this increase is of clinical importance. Continued monitoring in subsequent studies and/or hemovigilance is strongly advised.

P329 | Standardizing manufacturing and assessing quality of leucoreduced umbilical cord blood red blood cells for clinical use

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Background: Prematurity poses a significant health concern due to its prevalence and associated complications, with anemia being a common issue in extremely preterm infants (EPI) often requiring blood transfusions. Concerns over oxygen-related complications in EPI linked to red blood cells (RBC) sourced from adult blood (AB) donors have prompted exploration into umbilical cord blood (CB) as an alternative, containing foetal haemoglobin, more physiological for EPI. However, before embarking on clinical studies, it is imperative to standardize the manufacturing process for CB-RBC and compare its characteristics with AB-RBC.

Aims: This study assesses leucoreduced, gamma-irradiated CB-RBC prepared using a commercial closed system (Solutran Hemo Bionest ABC and EF kits from Meditalia srl, Lovero, Italy), from CB bank donations not meeting hematopoietic transplantation criteria.

Methods: Comparative analyses of CB-RBC with AB-RBC units stored in saline-adenine-glucose-mannitol (SAGM) were conducted. Parameters such as haematological and biochemical characteristics, pH, 2,3-diphosphoglycerate (2,3-DPG) levels, blood gases, and potential toxicants were scrutinized throughout the storage period.

Results: Post-fractionated CB-RBC units exhibited satisfactory initial quality parameters, with a comparable haematocrit (55±2%) to AB-

RBC. Notably, during storage, a faster rise in haemolysis levels was observed in CB-RBC compared to AB-RBC. Potassium (K⁺) levels significantly increased in both sources, while glucose levels decreased, and lactate levels rose, indicative of similar patterns of anaerobic glycolysis during storage. pH levels decreased, influencing the oxygen dissociation curve due to diminished 2,3-DPG levels.

The study evaluated the tolerance to irradiation at 14 days of storage, revealing that CB-RBC exhibited less stability with increased haemolysis and K⁺ compared to AB-RBC at 24 hours. Phthalate concentrations, indicative of plasticizers, increased during storage, albeit significantly less in CB compared to AB-RBC. Most measured metals remained within acceptable ranges.

Based on quality control parameters, we proposed a 14-day expiration date for CB-RBC stored with SAGM, with irradiation performed < 24 hours before transfusion. To accelerate quality release, new methods for determining slow-growth bacteria were evaluated. These additional assays, including molecular biology, enable product availability from day 4 to day 14.

Summary / Conclusions: In conclusion, leucoreduced CB-RBC quality during storage is influenced by haemolysis levels and extracellular K⁺ content. Our data suggest that 14-days of storage render RBC meeting basic safety standards, allowing their use in clinical settings. Our study group is now actively recruiting for first clinical trial (NCT05612919) using this new blood component.

P330 | Abstract withdrawn

P331 | Paediatric patient with plasminogen deficiency treated with artificial tears from serum and from platelet-rich plasma

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Background: Congenital plasminogen deficiency is a rare haematological disorder often accompanied by Ligneous conjunctivitis, whereby the patient suffers from recurrent membrane formation on the eyes. Membrane formation is caused by fibrin accumulation and consequent inflammation. The condition is most often diagnosed in infants and children and the incidence rate has not as yet been determined precisely. In Poland only symptomatic treatment of this condition is available.

Aims: The study aim is to analyse the impact of using artificial tears from serum and from platelet-rich plasma (PRP) in a paediatric patient with plasminogen deficiency.

Methods: A 6-year-old male patient with congenital plasminogen deficiency and consequent Ligneous Conjunctivitis was administered allogeneic artificial tears for a period of 38 months (November 2020 to January 2024). The first four donations came from the patient's mother, but because there was no significant improvement due to insufficient plasminogen levels a decision was made to turn to related

and unrelated donors. Initially, the artificial tears were of 100% serum, then from PRP for better treatment effect.

Each donor was tested for plasminogen levels and blood-borne infectious agents (HIV, HCV, HBV, *Treponema pallidum*). 150 ml of whole blood for serum drops and 300-450 ml of blood with CPD for PRP were collected. Drops were made according to standard procedure by centrifugation of clotted whole blood or blood with CPD respectively. The serum/PRP was then dispensed into sterile tubing and divided into single-use capsules. The patient's mother was instructed to apply the drops every 2 hours. A haematologist and an ophthalmologist assessed the patient's condition after each donation.

Results: The patient was administered drops prepared from donations of eight donors. A total of 15 donations were collected (11 for serum, 4 for PRP). Altogether the patient received 7832 single-use serum capsules and 5864 single-use PRP capsules.

Good treatment results were reported following the use of serum-based eye drops. Even better results were observed after PRP drops because plasminogen level in PRP is higher than in serum.

Summary / Conclusions: Artificial tears prepared from serum or PRP collected from donors with high plasminogen levels proved very effective in the management of Ligneous conjunctivitis. The drops reduced inflammation, accelerated healing of the ocular surface and provided good lubrication of the eyeball.

P332 | COVID-19 Vaccinated Convalescent Plasma (VaxCCP): A consolidated program in the Blood Bank of Catalonia (Spain)

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Background: Plasma from COVID-19 convalescent donors was initially suggested as a potential therapy for critically-ill patients. Three years later, high antibody titer convalescent plasma is recommended by the FDA for immunosuppressed individuals, with acute or persistent COVID-19 infection. Vaccine boosts the production of antibodies in COVID-19 exposed individuals, and the polyclonal antibody response to exposure plus vaccination might enhance effectiveness against a wider range of COVID-19 variants (Original, Omicron). Since February 2021, when COVID-19 vaccine was first administered to the general population in Spain, the blood bank in Catalonia has been supplying high-titer anti-Spike SARS-CoV-2 plasma units from convalescent and vaccinated donors (VaxCCP).

Aims: We present a three-year experience of characterization, production, and supply of VaxCCP in Catalonia (Spain).

Methods: VaxCCP was obtained from plasmapheresis of male donors that reported COVID-19 infection and vaccination against COVID-19.

Information on donors' self-reported COVID-19 infection, reinfection, and vaccination was recorded. The anti-Spike SARS-CoV IgG concentration to qualify VaxCCP was 20,000 AU/mL or higher (Architect, Abbott), which equals to 2840 binding antibody units (BAU)/mL or higher. The VaxCCP therapeutical unit consisted of two fractions (2 × 250 mL, methylene blue inactivated and filtered) obtained from the same plasmapheresis.

Results: From February 2021 to January 2024, 726 VaxCCP units have been issued for transfusion (2021: 125 units; 2022: 279 units; 2023: 294 units; January 2024: 28 units). VaxCCP units were obtained from 418 different donors (51 ± 11 years, range 19-69), and mean anti-Spike titer was 58625 ± 18177 AU/mL. The first vaccine shot in VaxCCP donations was 60% Pfizer, 24% Moderna, and 17% AstraZeneca, conversely, the first shot in the general population was either Pfizer or AstraZeneca. VaxCCP units were obtained from donors with at least one self-reported COVID-19 infection and two Original vaccine shots (N = 184 units, 49998 ± 18327 AU/mL), and from donors with additional boosters: 3rd Original vaccine shot (N = 259, 59173 ± 17057), 3rd Original vaccine shot and 4th Bivalent Original/Omicron shot (N = 126, 59234 ± 16322), COVID reinfection and 3rd Original vaccine shot with or without 4th Bivalent shot (N = 157, 68512 ± 15436). The median time from the last COVID booster to VaxCCP collection was 87 days, the median time from VaxCCP collection to supply for transfusion was 92 days, and the median time from COVID last booster to supply 254 days. Waning of anti-Spike antibodies over time was observed in the 142 donors with multiple VaxCCP donations. Four high-complexity hospitals, located in Barcelona Metropolitan area, ordered 654 of the 726 VaxCCP units (90%), while the remaining 72 VaxCCP units were ordered by 11 high and medium complexity hospitals. The majority VaxCCP units were ordered for patients with oncohematological conditions and severe immunosuppression.

Summary / Conclusions: Although morbidity and mortality of COVID-19 have decreased dramatically, there is an increasing demand for VaxCCP, namely for immunosuppressed patients. In Catalonia, the use of VaxCCP is concentrated in four tertiary hospitals, while anecdotic demand by other tertiary hospitals evidence that VaxCCP therapy is not a common practice. Continuous renewal of donors and an update of eligibility criteria are required in order to maintain a high titer VOC-specific VaxCCP stock.

P333 | Abstract withdrawn

P334 | Impact of different process preparations on the red cell concentrate quality with a combination DEHT-PAGGSM · multicentric study

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Background: The use of di(2-ethylhexyl)-phthalate (DEHP) has long been the plasticizer in polyvinyl chloride (PVC) whole blood collection

P334 - Table 1

Parameter	Kit 'Top and Bottom' / PAGGSM–RCC		Significative difference (Student test, $p < 0.05$)
	SFS preparation	EFS preparation	
Delay between collection & preparation	8h19	20h08	Yes ($p < 0.001$)
Haematocrit D1 (%)	58.0 ± 1.7	59.9 ± 3.3	No
Haemoglobin D1 (g/unit)	47.4 ± 3.9	53.0 ± 7.3	No
Residual white cells D1 (10^6 /unit)	0.06 ± 0.05	0.05 ± 0.06	No
Haemolysis D49 (%)	0.30 ± 0.15	0.35 ± 0.06	No
Glucose D49 (mMol)	14.9 ± 1.8	13.4 ± 2.3	No
Lactate D49 (mMol)	29.9 ± 3.3	30.6 ± 2.6	No
K ⁺ D49	44.2 ± 3.3	49.7 ± 6.1	Yes ($p = 0.05$)
ATP D49 (μ M/gHb)	2.22 ± 0.41	2.69 ± 0.57	No
pH D49	6.41 ± 0.04	6.43 ± 0.05	No

sets. However, studies data suggested its potential toxicity as endocrine disruptors and human carcinogens that have led to regulatory changes being introduced in the European Union banning the use of this plasticizer in medical devices. The DEHP is also known for its protector effect and stabilizer of the red blood cell (RBC) membrane during their storage and its ban leads to test new combinations of DEHP-free plasticizers/storage solutions to maintain the same current quality of RBC.

Aims: Assess the impact of different process preparations on the RBC quality stored in di(2-ethylhexyl)-terephthalate DEHT (Transfulol DEHT 3126 from RENOLIT Nederland B.V.)/PAGGSM (Phosphate Adenine Glucose Guanosine Saline Mannitol).

Methods: 16 RBC were prepared from non-therapeutic whole blood (WB) collected with DEHT/PAGGSM bags with 'Top and Bottom' (TB) kit + filter RBC (LCRD2). 8 Red Cell Concentrates (RCC) have been prepared by Belgian SFS during the night and 8 other RCC have been prepared by French EFS overnight according to their own work instructions.

Results: The potassium rate is more important when the process is performed with a delay above 12 hours. The significant difference on the potassium level does not translate in a significant increase of haemolysis. There is no statistical difference for the other parameters.

Summary / Conclusions: There is no significant impact on the different process preparations on the RCC quality with a combination of DEHT-PAGGSM, outside of the potassium that is more pronounced when the process is performed above 12 h after WB collection.

P335 | A comparative study on the properties of pig and human blood—implications for xenotransfusion practice

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Background: A worldwide shortage of human blood has led to the exploration of alternative blood sources for medical purposes. As one

of these fields, xenotransfusion has gained attention as a promising practice, with the use of pig red blood cells (RBCs) considered a viable option due to their marked resemblance to human RBCs. The processing method and storage condition for pig RBCs post-collection have not been as rigorously investigated and standardized as those applied to human RBCs.

Aims: The objectives of this research are: (1) to investigate the similarities and differences between pig and human blood properties; (2) to track changes in hematological and biochemical markers over time in both blood products; and (3) to assess the effectiveness of the buffy coat (BC) method compared to the platelet-rich plasma (PRP) method in the production of packed RBCs from whole blood (WB).

Methods: Pig WB ($n = 14$) from genetically modified pigs was provided by Optipharm Inc. (Cheongju, Republic of Korea), including TKO, TKO/hCD55.hCD39KI, and QKO types. Human WB ($n = 29$) was obtained from fresh blood products discarded after phlebotomy treatment of patients with hematologic-oncological conditions. Methods for producing packed RBCs from WB were compared: the hard spin BC method (centrifugation at 3,000g for 8 minutes) versus the soft spin PRP method (centrifugation at 400g for 8 minutes). Hematological and biochemical parameters of WB and packed RBCs were analyzed according to the storage period on days 0, 1, 7, 14, 21, and 28. To compare the viability of white blood cells (WBCs) on days 1 and 19, apoptosis were assessed by flow cytometry.

Results: The study found the BC method increased free hemoglobin ($P = .0048$) and lactate dehydrogenase (LDH) ($P = .000138$) compared to the PRP method. In pigs, the BC method also raised potassium levels ($P = .043$). Significant baseline differences between human and pig blood included RBC count, total protein, albumin, LDH, potassium, and calcium ($P < .05$). In human and pig blood, the hematological and biochemical parameters significantly differed ($P < .05$) according to storage period, including WBC count, RBC count, total protein, albumin, and free hemoglobin. Levels of free hemoglobin ($P = .03125$) and LDH ($P = .000488$) significantly increased by day 7 in pig packed RBCs. Conversely, these indicators did not show significant changes in pig WB products. The neutrophil and mononuclear cell populations

were gated on days 1 and 19 by flow cytometry. As time progressed, there was an increase in the percentage of apoptosis: in neutrophils, it rose from 33.15% to 98.17%, and in mononuclear cells, it increased from 54.11% to 96.55%.

Summary / Conclusions: In the BC method, hemolysis is more prevalent than in the PRP method. As hemolysis may adversely affect red blood cell preservation and transfusion outcomes, the PRP method is considered more effective. The degree of hemolysis differed depending on whether it was WB or packed RBCs. This suggests that processing of packaged RBCs may affect the stability of blood cells differently compared to WB. The apoptosis assay reveals an escalation in the apoptotic fraction of pig WBCs commensurate with storage duration. Interestingly, the WBC count remained relatively unchanged. This suggests that the observed patterns are caused by changes in cellular function rather than quantitative changes in cell populations (grant No. 22-CM-EC-18).

P336 | Understanding the potential benefits of universal platelets and universal plasma for the English blood service

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Background: Currently platelet and plasma transfusions require the donor and recipient to be blood group compatible. In England, clinical guidelines are to transfuse ABO and RhD compatible components, but this is not always possible. This leads to a high demand for group A RhD negative platelets (17%) and group AB plasma (8%) which are seen as the most suitable alternatives due to their low titre status or lack of anti-A/B. However, only 8% of the UK population are group A RhD negative, and only 4% are group AB. Meeting these demands from hospitals necessitates complex blood supply planning, and when stocks are low, different blood group substitutions may be made to fulfil hospital orders. The development of a device to reduce anti-A/B antibodies (to produce Universal Plasma (UP)) and additionally removing residual red blood cells from blood donations to produce Universal Platelets (UPLts), is predicted to bring many benefits.

Aims: Our aim was to understand the benefits to the English blood service (NHS Blood and Transplant, NHSBT) from the potential introduction of UPLts and UP.

Methods: Following initial scoping discussions, senior representatives from across the NHSBT supply chain were nominated to attend a one day in-person event in November 2023. The 11 participants were from: blood supply and stock management (1), forecasting and demand planning (2), donor experience (1), hospital

services and operations (3), manufacturing and testing (3) and component development (1). Utilising the “six thinking hats” approach (de Bono, 1985), the attendees were asked to consider the benefits (yellow hat), costs/barriers (black hat), instincts (red hat) and creative solutions (green hat) of introducing UPLts and UP. In a further session, participants discussed preferences for ABO/RhD Universal Components.

Results: Benefits: Of the 58 statements for the blood service, the most frequent areas reported were: simplified blood supply planning (28%); easier management of blood stock holding (14%); manufacturing benefits (12%); fewer complexities issuing to hospitals (12%); and reduced wastage (12%). The stakeholders felt introducing UPLts and UP would improve the resilience of the platelet/ plasma supply chains. It was agreed that the current high wastage of A negative red cells from whole blood collected to meet platelet/plasma demands would significantly reduce.

Costs/barriers: Of the 47 statements for the blood service, the most frequent areas mentioned were: manufacturing (23%); testing (19%); blood service computer systems (11%); and overall costs of implementation (11%). Many of the costs and barriers reported related to set up and roll out considerations. Concerns were raised about additional processing time and quality assurance.

Universal preferences: Of the nine participants who stated a preference for ABO or RhD universal components, six favoured ABO and three favoured RhD universality. If a non-100% universal supply model was adopted there may be multiple product streams, which would add complexity to the blood supply chain.

Summary / Conclusions: The stakeholders confirmed multiple benefits for the blood service with the introduction of UPLts and UP. Further planned work includes a health economic analysis across the whole blood supply chain from donor to patient and developing a device to manufacture UPLts and UP. We will also seek international views on the impacts of these components which may differ between countries.

P337 | Analysis and comparison of in-vitro properties of Cryopreserved Platelet Concentrate (CPPC) versus Liquid Stored Platelet Concentrate (LSPC)—a single-center study from India

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Background: In India, there is an increasing use of single donor platelets (SDP), especially in tertiary health care centers. It is also true that in low-resource settings like India, these are directed donations. Substantial number of these directed SDPs are expired, if they are not required in the intended recipient(s). Conventional storage of platelet concentrates limits their shelf-life to 5-days. Newer method of storage which involves cryopreservation could potentially prolong their shelf-life to months or even years. There is a paucity of data on cryopreserved platelets in India. The purpose of this study was to evaluate, if

P337 - Table 1

In-vitro Parameters	LSPC	CPPC	p-value	Inference
Platelet count ($\times 10^3/\mu\text{L}$)	1381.3 \pm 310.5	846.5 \pm 230.2	<0.001	Decrease
Platelet distribution width (fL)	10.9 \pm 2.7	12.6 \pm 1.7	<0.001	Increase
Mean platelet volume (fL)	8.8 \pm 1	9.4 \pm 0.7	<0.001	Increase
pH (37°C)	7.1 \pm 0.2	7 \pm 1.1	0.648	No change
Lactate (mmol/L)	3.3 \pm 0.7	12.7 \pm 5.2	<0.001	Increase
Glucose (mg/dl)	379 \pm 48.5	221.8 \pm 29.8	<0.001	Decrease

cryopreserved platelet concentrate (CPPC) is non-inferior to room temperature liquid stored platelet concentrate (LSPC). Further, it could be followed up with an in-vivo study and eventually help us realize conservation of an important resource, that gets wasted, otherwise.

Aims: The aim of this study was to assess the in-vitro properties encompassing the morphologic, metabolic, and functional aspects of platelets stored at room temperature on Day-5 (LSPC) in comparison to those of CPPC.

Methods: Leucodepleted SDP collected in autologous plasma were assessed on Day-5 of room temperature storage (LSPC) for morphologic (platelet count, platelet distribution width, mean platelet volume, grade of swirling), metabolic (pH, level of glucose and lactate), and functional (flow cytometry and platelet aggregometry) parameters. Following sampling, these platelet products were cryopreserved using Modified Valeri's method and then stored at -80°C for one month prior to thawing. Thawed platelets (CPPC) were then reconstituted in freshly thawed plasma and a platelet sample was removed, after two hours for in-vitro testing. The in-vitro survival and viability were assessed by repeating the same tests post-thaw.

Results: A total of 50 Day-05 SDP concentrates (LSPC) were analyzed. The recovery percentage post-thaw was 62.24%. The physical and biochemical parameters analysed have been summarised in Table 1. LSPC maintained 3+ swirl whereas CPPC did not swirl immediately after thawing, but regained 2+ swirl by 6-hours, post-thawing. On flow cytometry, the proportion of platelets expressing GPIIb/IIIa (CD42b) was significantly reduced in CPPC; while those expressing the activation marker P-selectin (CD62P) was equivalent in CPPC as compared to LSPC. The percentage of annexin-V-positive platelets in LSPC was 7.2 ± 4 , which increased to 90 ± 11.4 in CPPC. On Light Transmission Aggregometry (LTA), LSPC responded strongly to agonists (ADP, Arachidonic acid, Collagen, Epinephrine and Ristocetin). CPPC, however, displayed a significant impairment to the maximum agonist-induced response.

Summary / Conclusions: A platelet recovery percentage of 62.24% and having other in-vitro parameters within permissible limits opens the possibility of a follow-up in-vivo study to analyse the platelet increment and effect on hemostasis. We hope and expect that this in-vitro and subsequent in-vivo data, would help us realize our long-term objective of salvaging near-expiry single donor platelets (SDP) for possible clinical use, in India.

P339 | Apheresis granulocytes versus pooled buffy coat derived granulocyte concentrates—a comparative analysis of component quality parameters

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Background: Methods used to collect, process and store granulocyte concentrates are likely to impact on their recovery, survival and function after transfusion. The FDA authorities in India have approved Granulocyte concentrates including apheresis derived and pooled buffy coat derived granulocytes as licensed products. Except for the total granulocyte content per bag, no other quality parameter has been currently specified for this component.

Aims: To evaluate and compare quality parameters like volume, WBC content, granulocyte content, RBC content and Platelet content of Granulocyte concentrates prepared by apheresis (AGC) and Pooled buffy coat derived granulocyte method (PBCGC).

Methods: Granulocyte donations were performed by volunteer donors after clearing medical and serological screening and undergoing mobilization using G-CSF and Dexamethasone. All apheresis procedures were performed using COM.TEC (Fresenius Kabi, Germany) automated continuous flow cell separator and P1YA kit after 8-10 hours of mobilization. PBCGC's were prepared by pooling six buffy coat units after initial standardization of the preparation procedure. All granulocyte components were gamma irradiated prior to issue. Variables impacting and including the quality parameters of product (PBCGC vs. AGC) were analysed using Mann-Whitneys test and Fishers exact statistical tests. A p value <0.05 was considered statistically significant. Analyses were performed using SPSS Statistics 21.0 software.

Results: Granulocyte concentrates were prepared over a period of 3 years for 149 patients, 225/510 were AGC and 285/510 were PBCGC. Pre-procedural WBC count of donors was mean $29.76 \times 10^3/\mu\text{L}$ and total blood volume of seven litres was processed per harvest session. On comparative analysis, the mean values for product volume was 266 mL and 285 mL, mean WBC content was 2.39×10^{10} and 1.85×10^{10} , Granulocyte content was 2.03×10^{10} and 1.11×10^{10} , RBC volume was 38.17 mL and 126 mL, Platelet content per bag was 3.42×10^{11} and 4.35×10^{11} respectively in the AGC and PBCGC, respectively. These differences were found to be statistically significant

($p < 0.05$). One donor experienced an anaphylactic reaction to G-CSF and two transfusion reactions were noted: one case of TRALI in a recipient of AGC and one allergic reaction in a recipient of PBCGC.

Summary / Conclusions: AGC prepared using COM.TEC cell separator had an acceptable volume, greater granulocyte content and less contamination with red cells and platelets when compared with the cellular content of PBCGCs. Further in vitro analysis of neutrophil functions and follow up of recipients for RBC and HLA alloimmunization is required for better understanding of safety and efficacy of these components.

P340 | Understanding the potential benefits of universal platelets for hospitals in England

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Background: Transfusing a patient with ABO incompatible platelets can cause haemolytic transfusion reactions and poor platelet increments due to ABO minor and major incompatibility respectively. Transfusing RhD incompatible platelets can lead to alloimmunisation. In England, clinical guidelines are to transfuse ABO and RhD compatible platelets, however due to their short 7-day shelf life this is not always possible. Within England, there is high demand (17%) from hospitals for group A RhD negative platelets (8% of English donor population), which are seen as the most universally compatible platelet option. In the last year there have been frequent blood service platelet stock shortages, especially for A RhD negative platelets. The development of a device to reduce anti-A/B antibodies and remove residual red blood cells from blood donations to produce Universal Platelets (UPIs), which would be suitable for transfusion to all patients regardless of their ABO/RhD blood group, is predicted to bring many benefits to hospitals and patients.

Aims: Our aim was to understand the benefits to hospitals and patients from the potential introduction of Universal Platelets (UPIs).

Methods: Two online surveys were distributed to 240 hospitals who are supplied by the English blood service (NHS Blood and Transplant). Version 1 (15 questions) was for Transfusion Laboratory Managers (TLMs) and received 62 responses from 55 hospitals. Version 2 (14 questions) was for Haematology Consultants (HCs) and Transfusion Practitioners (TPs) and received 49 responses from 38 hospitals. Questions relating to perceived benefits, barriers and costs were included.

Results: Benefits: From the TLM survey, the greatest benefit identified was a reduction in platelet wastage (27%), followed by easier organisational aspects of platelet inventory management (24%). In fact, 75% of TLMs predicted their platelet wastage would decrease on the introduction of UPIs, with 13% anticipating wastage would reduce by more than 25%. From the HC/TP survey, the greatest perceived benefit was faster availability of platelets for patients (63%) followed by reduced staff time managing platelet stock alerts (47%). Haematology/oncology and the emergency department were the clinical areas deemed to see the most benefits. 29% HCs/TPs reported that blood service platelet shortages had resulted in delays in transfusions in their hospital in the past year.

Costs/barriers: The greatest barriers anticipated were a potential increase in the cost of platelets (53% TLMs, 67% HCs/TPs) and computer systems within the hospital restricting or preventing use (47% TLMs, 35% HCs/TPs). Many TLMs (34%) felt unable to comment on a maximum acceptable price increase for UPIs, however 37% were willing to pay more. 23% TLMs and 29% HCs/TPs foresaw no barriers to the introduction of UPIs in their hospital.

Predicted use: If available, 98% TLMs said they would order UPIs, however 60% anticipated they would order UPIs and some conventional platelets. When questioned about stockholding of platelets, 74% of TLMs indicated that introducing UPIs would have no impact on the number of platelets they would stock in their hospital.

Summary / Conclusions: Both surveys confirm a number of benefits for hospitals and patients if UPIs were available. Further planned steps include a health economic analysis across the whole of the platelet supply chain from donor to patient and development of a device to produce UPIs.

P341 | Use of low titre group O whole blood in the Czech Republic

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Background: Massive bleeding is a leading cause of death in poly-trauma, especially in young people. Traumatic hemorrhagic shock in adults has a mortality approaching 20% at 24 h post-injury. High morbidity and mortality is caused by the „lethal triad“ – hypothermia, acidosis and trauma induced coagulopathy. Using early hemostatic resuscitation procedures can in many cases improve survival. In the last two decades the evidence for the lifesaving benefits of Low Titre

P341 - Table 1: Production of LTOWB in the Czech Republic by facility

Blood Transfusion Establishment	University Hospital Hradec Králové	Military University Hospital Prague	University Hospital Ostrava	University Hospital Olomouc
Start of production LTWB	Jun-20	Jun-20	Jun-20	Jan-23
Produced units of LTWB (until Dec.2023)	1762	449	467	63

P341 - Table 2: The use of LTOWB in Czech Republic

Initiating the use of LTWB	Bleeding cases	Transfused units of LTWB (until Dec.2023)	Transfused patients with LTWB (until Dec.2023)
Jun-20	polytrauma - emergency admission	1593	698
	other severe bleeding cases	665	309
	polytrauma - prehospital	246	141
	total	2504	1148

Group O Whole Blood (LTOWB) in bleeding trauma patients has increased in both the prehospital settings as well as in hospital.

Aims: Review of LTOWB use in the Czech Republic

Methods: LTOWB began to be gradually produced and used in the Czech Republic in June 2020. The LTOWB is leucoreduced, RhD-negative with titers of anti-A and -B of <256, TRALI risk mitigated and with a shelf life of either 14 or 21 days depending on the manufacturer. Unused LTOWB units are 1-3 days before expiry reprocessed into RBCs (usable for another 21 days) and plasma (discarded).

Results: Between June 2020 and December 2023, a total of 2741 units of LTOWB have been produced in the Czech Republic, of which a total of 2504 units have been transfused to 1148 patients. A detailed overview of production is given in Table 1 and an overview of use in Table 2.

Summary / Conclusions: LTWB is viable product to use in the treatment of massive bleeding, especially in polytrauma with the development of hemorrhagic shock. The rationale for the use of LTWB early in the resuscitation of massively bleeding patients is multifactorial: provides a balanced resuscitation that simultaneously addresses oxygen debt and coagulopathy, is a more concentrated product that contains a smaller quantity of anticoagulant and preservative solutions compared with an equivalent amount of reconstituted WB from blood components, the cold stored platelets in WB improve hemostasis more effectively compared to platelet units stored at room temperature.

P342 | Abstract withdrawn

P343 | Comparative in-vitro study of room temperature and cold stored double dose platelets pathogen-reduced with amotosalen and UVA light

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Background: Platelet concentrates (PC) are normally stored at 22°C for up to 5 days and extended storage up to 7 days can be applied only if bacterial detection or a pathogen reduction treatment is performed after production of PC. The current needs for platelet transfusions are changing and nowadays more platelets are given to patients who are actively bleeding and since cold-stored platelets are more hemostatically effective and potentially longer shelf life. There is an enormous interest in cold-stored platelets but before this product is used in routine or for specific patients, investigation of in vitro storage characteristics and in vivo transfusion effects is required.

Aims: This in vitro study compares room temperature (RT) and cold-stored double-dose platelets (DD PLTs) treated with Amotosalen and UVA light over 21 days. The objective is to assess biochemical and functional properties of RT and cold-stored DD PLTs. Parameters like volume, PLT content, pH, swirling, pO₂, pCO₂, HCO₃⁻, LDH, and glucose are evaluated. DD-PCs are prepared from 8 ABO-matched whole blood-derived buffy-coat units, pooled with platelet additive solution. Four storage conditions are studied: C-PC at 22°C or 4°C, and PI-PC at 22°C or 4°C. Results from 3 replicates show stable pH, moderate decreases in values, slightly increased LDH concentrations, and maintained glucose reserves until Day 7, especially at 4°C. Clinical studies of cold-stored platelets in bleeding patients are essential to assess their effectiveness in minimizing time to hemostasis and rebleeding events. Further tests, including CD62P measurement and ROTEM analysis, are ongoing, and additional replicates are needed for a comprehensive understanding.

Methods: Double dose platelet concentrates (DD-PC) are prepared from 8 ABO-matched whole blood-derived (WB) buffy-coat units (BC). The buffy-coats employed in this study are buffy-coats not used for the routine PC production because of the donor low platelet count. The 8 BC are pooled with 280 mL of platelet additive solution (PAS, Intersol) using manual pooling method. For each replicate, two ABO-identical double-dose platelet concentrates free of aggregates, are mixed and divided into two DDPC. Each replicate will include therefore 4 arms: Study arms 1,2: CONTROL PC (C- PC) stored at 22°C in a platelet agitator or at 4°C, without agitation. Study arm 3, 4: pathogen inactivated PC (PI - PC) stored either at 22°C in a platelet agitator or at 4°C, without agitation. All 4 types of PC will be stored in platelet containers from INTERCEPT DS kit up to 21 days. Samples during storage are taken on Days 2, 5, 7, 14 and 21 for evaluation of in vitro platelet function and metabolism.

Results: Preliminary results from 3 replicates have been analyzed for metabolic parameters. From this initial data set, we have observed that pH is maintained well during the storage period of 21 days even if values are moderately decreased over storage. LDH concentrations are slightly increased, and glucose reserves are maintained until Day 7, especially in PC stored at 4°C.

Summary / Conclusions: Even if the data set is limited and more replicates and further tests are required (CD62P measurement by flow cytometry and ROTEM analysis), our study provides additional information regarding in vitro parameters of PC stored at 4°C. It's clear that clinical studies of cold-stored platelets in acutely bleeding patients are needed to determine whether refrigerated platelets will minimize time to hemostasis and rebleeding events.

P344 | The power to heal ourselves—promising results for users of serum eye drops

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Background: Blood-derived autologous serum eye drops are an important treatment for severe dry eye disease. Patients who suffer from severe dry eyes due to their illness, for example: GVHD or Sjögren's syndrome, can improve their condition and prevent damage to the cornea. Bio iDrops at the Rambam blood bank is a joint service with the ophthalmology department whose purpose is to produce these special eye drops while utilizing the knowledge and experience of the blood bank laboratory in the donation, production preparation and documentation of various blood components and in accordance with the accepted protocols.

Aims: to assess the treatment satisfaction among our ocular surface disease patients treated with autologous serum eye drops

Methods: during 2023, 683 preparations were made for 418 patients. Patients were asked to fill out a dedicated questionnaire: OSDI - Ocular Surface Disease Index in which they rated the severity of their condition before and after a round of 3 months of serum treatment.

Results: Out of 88 patients who filled out questionnaires as required, 88% rated their pre-treatment condition as suffering from severe dry eye, 8% as suffering from moderate dry eye and 5% as suffering from mild dry eye. After 3 months of treatment only 40% defined their condition as suffering from severe dry eye (a decrease of 45% $p < 0.001$), 20% as suffering from moderate dry eye and 20% as mild dry eye with 19% defined as normal (resolution of dry eye disease).

Summary / Conclusions: Autologous serum preparation service at the hospital's blood bank increases the availability of the unique and successful treatment as well as enables multidisciplinary treatment in collaboration with hematologists, specialist ophthalmologists and blood bank laboratory workers.

P345 | Serum eye drops - our experience about tolerance and monitoring

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Background: Serum eye drops (SED) is a useful treatment for patients with severe ocular surface disease that do not respond to conventional treatment and commercially available artificial tears. The rationale is that serum contains many of the substances found in normal tears but also epitheliotropic factors and growth factors. Autologous SED are made from blood donated by the patients themselves but if autologous SED is contraindicated or not appropriate there is allogeneic SED made from blood of healthy volunteer donor. In our service we started to prepare SED from 2008: in the first year we prepared 15 autologous SED. In our experience the demand for SED preparations in continuous increase for the expanded indication of treatment so that in the last year (2023) we made 199 autologous preparations and 78 allogeneic preparations from AB group blood-donors. Our standard preparation of SED, in accordance with the National regulations, requires the centrifugation of 200 ml whole blood that is diluted with 30% saline to obtain 50 serum eye drop vials. Some of our patients were intolerant to standard SED and reported a worsening of ocular symptoms during the treatment. These patients well tolerate a SED diluted with 80% saline as it is suggested for the preparations of SED from cord blood. The efficacy of a SED therapy can be detected by improvement of patients ocular signs and symptoms.

Aims: We want to identify what kind of ocular surface disease most frequently need an hyperdiluted SED. Also we want to verify if a questionnaire can help early monitoring the impact of a SED treatment on ocular symptoms.

Methods: We analyzed the clinical data of our patients using hyperdiluted serum, in particular we were interested in the ocular disease. We gave to 30 new patients the Dry Eye Questionnaire (DEQ-5) for

dry eye symptoms and related quality of life before starting a therapy with SED and after almost 7 days of use of SED.

Results: In our case study 9 patients intolerant to standard SED preparation use hyperdiluted SED; of these patients, 7 have a story of bone marrow transplantations for hematological malignancies and secondary ocular graft versus host disease (oGVHD) (11 autologous and 6 allogeneic SED) and 2 have congenital aniridia (autologous SED). Among all our patients 12 have oGVHD and 5 (42%) use standard SED (3 autologous and 2 allogeneic SED): really one of these patients has initially used hyperdiluted SED for intolerance to standard SED but after a year for no more effectiveness, started the use of standard dilution with satisfaction. No other patients have aniridia in addition of the 2 patients using hyperdiluted SED. In all 30 patients analyzed the DEQ helped to monitor early treatment tolerance and satisfaction.

Summary / Conclusions: The question of SED dilution is still open. For most of our patients with oGVHD (58%) and all our patients with congenital aniridia overdiluted SED presents advantageous in terms of efficacy and tolerance probably related to the ocular severity disease and hypersensitivity. In our experience a questionnaire such as DEQ-5 can be useful for early identification of patients who requires dedicated preparations and also for periodical monitoring of treatment efficacy.

P346 | Participation of the blood bank of La Rioja during the pandemic -contributing our grain of sand

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Background: On March 11, 2020, the infection caused by the virus known as SARS-CoV-2 was declared a pandemic. Due to the scarcity of specific treatments for SARS-CoV-2 infection, clinical trials are beginning to be carried out, among others to assess the efficacy of transfusion of plasma from blood donors who have overcome the disease, called COVID-19, and who have generated specific antibodies against the infection, this plasma is called hyperimmune plasma (HP). The Blood Bank of La Rioja (BBR) participates in the study led by the Puerta de Hierro Hospital to obtain HP. Likewise, and following

P346 - Table 1: Results

	2020	2021	2022	2023
Units HP obtained by apheresis	196	1	0	0
Units HP obtained from whole blood	87	192	0	0
Units HP from transfused apheresis	64	99	0	0
Units HP from transfused whole blood	10	105	0	0

instructions from the Ministry of Health, since August 2020, the obtaining of HP is encouraged through plasmapheresis or whole blood donation, for which blood donors carrying IgG antiSARS CoV-2 antibodies are called for. In our community, the demand for these units

comes from the San Pedro Hospital from Logroño, where the BBR is located; the HP units are delivered by the BBR staff to the clinical unit that requests them.

Aims: To see evolution of the "supply and demand" of HP during the pandemic period of 2020, 2021, 2022 and 2023.

Methods: A retrospective study was carried out from 2020 to 2023, where the number of HP donations obtained and demanded was collected. This hemocomponent is obtained by an apheresis donation of 600 ml, aliquoted in 2 therapeutic units and obtained by whole blood. We also studied the number of patients transfused each year with this plasma, considering that the therapeutic guideline was 1 unit per patient, except in immunosuppressed patients, who were administered up to 3 units.

Results: The data collected between May 2020 to December 2023 are:

Summary / Conclusions: (1)- In 2020, 283 PH units are obtained, while that figure decreases to 183 in 2021, being 0 in 2022 and 2023. This data gives an idea of the great altruism and quick response capacity, facing a necessity, by blood donors, who once again, save lives with their generosity. (2)- The vaccine, with the immunization of the population that overcomes the COVID-19 disease, makes the current incidence of patients admitted for this reason decreases, with the consequent decrease in the demand for PH, going from 74 units of PH transfused in 2020 to 204 units in 2021 and null in 2022 and 2023. (3)- Emphasizing the capacity of BSR staff to adapt their work in order to respond to the needs of the moment, increasing the collection of PH by apheresis and whole blood, inactivating it with methylene blue and delivering it to units such as infectious diseases and intensive care medicine at such critical times, while continuing to meet the rest of La Rioja Health System demands.

Transfusion transmitted infections—Screening strategies for TTI

P347 | Improving NAT screening strategies in real life—simultaneous ID-NAT detection of HCV, HIV-1, HIV-2, HBV and HEV in blood donations

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Background: Commercial multiplex nucleic acid tests (NATs) for HIV-1/HIV-2/HCV/HEV are widely used in developed countries to screen blood donations. HEV NAT screening has also been implemented in some blood banks, as several cases of post-transfusion hepatitis E infections have been documented. Sensitive detection of viral load is enhanced by individual donation testing (ID-NAT) over minipool testing (MP).

Aims: We describe the two-year experience of simultaneous ID-NAT detection of HCV RNA, HIV-1 RNA, HIV-2 RNA, HBV DNA and HEV RNA using a new multiplex assay.

Methods: On March 10, 2022, in the Blood Bank of Catalonia (Spain) a decision was made to replace the HCV/HIV-1/HIV-2/HBV ID-NAT (Ultrio Elite, Panther, Grifols) and the HEV minipool of 16 donations (MP-16) NAT (Procleix HEV, Panther, Grifols) by ID-NAT testing with a single reagent that simultaneously detects HCV/HIV-1/HIV-2/HBV and HEV (Procleix® UltrioPlex E (UPxE), Panther, Grifols). UPxE assay returns two results (reactive or non reactive), for HCV/HIV-1/HIV-2/HBV and for HEV separately. Initially reactive donations for HCV/HIV-1/HIV-2/ HBV are compared with the serological markers, and re-tested five times with UPxE and the specific discriminatory assays for HCV, HIV-1 and HIV-2, and HBV. Initially reactive samples for the HEV signal are re-tested five times with UPxE. Samples are confirmed as positive when NAT result can be reproduced and/or anti-HBc serology is positive, otherwise the sample is considered a false positive result.

Results: From March 2022 to December 2023, a total of 500.066 blood donations were ID-NAT screened with UPxE. In all, 483 donations were initially reactive (205 in the HCV/HIV1-2/HBV signal and 278 in the HEV signal). After completing the confirmatory algorithms, 122 samples were deemed false positive for HCV/HIV1-2/HBV and 105 were deemed false positive for HEV, resulting in an identical specificity rate of 99.98%. In 12 donations initially reactive for HCV/HIV1-2/HBV, all NAT repeats and discriminatory assays were negative, but anti-HBc was positive, therefore the donors were considered low viral load occult hepatitis B infections (OBI). Confirmed NAT positivity was obtained for 71 samples initially reactive for the HCV/HIV1-2/HBV signal, of them 10 donors were HCV, 16 HIV-1, 15 OBI and 30 HBV. All samples had a concordant serological positivity, except for one HBV window phase. A confirmed NAT positivity was obtained for 173 samples initially reactive for the HEV signal, resulting in a HEV ID-NAT yield of 1 in 2891 donations screened (CI95%: 1 in 2491 to 1 in 3375). As expected, HEV ID-NAT yield was significantly higher than our previous experience with HEV NAT MP-16 donations (1 in 4341; CI 95%: 1 in 3703 to 1 in 5004). In the period of this study, there has been no reported transfusion-transmitted infections to the Hemovigilance System. Simultaneous detection of HCV, HIV-1, HIV-2, HBV and HEV improved the logistics of the screening laboratory by: (1) eliminating the step of pooling, (2) the equipment devoted to HEV testing could be used for HCV, HIV-1, HIV-2, HBV and HEV NAT thus increasing results throughput, (3) reducing wastes, (4) optimizing the management of stocks and (5) simplifying the capacitation of the laboratory staff.

Summary / Conclusions: Simultaneous detection of HIV-1/2, HCV, HBV and HEV is a coherent solution to increase blood safety with minimal technical complexity.

P348 | Using sequential immunoassays for the screening of syphilis in blood donors

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Background: Australian Red Cross Lifeblood performs *Treponema pallidum* serological testing for the screening of syphilis in blood donors. For over two decades, Lifeblood used an automated micro-haemagglutination assay (TPHA) for screening and in December 2018, added a treponemal chemiluminescent particle immunoassay (CMIA) as a sequential assay which is used to clarify the antibody status of donors with reactive screening results. Samples reactive on the screening assay but non-reactive on the sequential assay are considered to be screen false-positive. In December 2020, the Alinity s Syphilis assay replaced the TPHA as the screening assay and the use of the CMIA as the sequential assay ceased as it was expected using two assays with the same antibody detection system, would result in highly concordant results with overlap in non-specific reactivity. Following a small evaluation of an alternate sequential assay, Lifeblood implemented the Elecsys Syphilis as the sequential assay on 12 July 2021.

Aims: Report two-year data using the Roche Elecsys Syphilis assay as the sequential assay to the Abbott Alinity s Syphilis assay in the screening of Australian blood donors.

Methods: Syphilis testing data from donations collected between 7 July 2021 to 6 July 2023 was analysed. Donations were screened on the Alinity s Syphilis assay with repeatedly reactive samples then tested on the Roche Elecsys Syphilis assay. Samples that were Elecsys repeatedly reactive were further tested using three confirmatory assays (*T. pallidum* particle agglutination test [TPPA], a rapid plasma reagin [RPR] and a fluorescent treponemal antibody absorption test [FTA-ABS]). Following confirmatory testing which included donor follow-up where indicated, a syphilis outcome was assigned. Samples which tested Alinity repeatedly reactive/Elecsys non-reactive were considered to be Alinity false-positives.

Results: 1,767,782 samples were screened with the Alinity assay. 1,850 samples (0.11%) were found to be repeatedly reactive and 1,456 of these were non-reactive on the Elecsys assay. The Alinity false-positive rate was determined to be 0.08% (1,456/1,767,782). The common false-positive overlap between the two assays was calculated as 3.83% (58/1514). The 1,850 samples were collected from 1,376 donors. 1,061 (77.1%) donors were assigned a syphilis outcome of Alinity false-positive, 53 (3.8%) were assigned as Alinity/Elecsys concordant false-positive, 152 (11.0%) were determined to be past treated for syphilis, 84 (6.1%) had unknown infection and were referred for medical assessment and 26 (1.9%) had a current infection.

Summary / Conclusions: In our study using blood donors, we found both the false-positive rate for the Alinity screening assay and the common false-positive overlap between the Alinity and Elecsys assays, to be low (0.08% and 3.83% respectively). This suggests this combination of sequential assays is highly suitable for screening a low pre-test probability population such as blood donors.

P349 | False positive test results among blood donorsS D Olsen¹, D K Holm¹, J Georgsen¹, M T Bruun¹¹Department of Clinical Immunology, Odense University Hospital, Odense, Denmark

Background: High sensitivity of assays for screening of blood donations is necessary to assure that no infectious agent is transmitted. High sensitivity occasionally results in false positive test results (FPTR). The cause of FPTR is not always clear, but it is interpreted as a technical problem. In case of a first-time FPTR, the blood component is discarded, but the donor is not informed. In case of a subsequent second FPTR the donor is informed and deferred for two years. As plasma donations for fractionation are increasing, the frequency of donations is increasing, which may potentially result in more FPTR.

Aims: The aim of the present study is to examine the incidence of FPTR among whole blood (WB) and plasma donations, and determine the proportion of donors with two consecutive FPTR (first and second). Furthermore, we will determine whether time between first and second donation affects the risk for a second FPTR.

Methods: From January 2021 to November 2023 the incidence of first and second time FPTR among all blood donors screened in the Region of Southern Denmark was examined by extracting test results from the blood bank IT system. Abbott Alinity i HBsAg, HCV Ab, and HIV Ab/Ag assays and Roche Cobas MPX assay were used for screening of donors. To conclude whether a test result was true positive, a positive screen test had to be confirmed by a more specific assay. Comparisons between incidences were analyzed by Fisher's exact test. Using logistic regression, it was examined whether the risk of a second FPTR was affected by the time between first FPTR and the subsequent donation.

Results: WB and plasma were donated by 38,993 donors resulting in 223,399 donations. WB and plasma donations accounted for approximately 50 % each. The total number of FPTR (first and second) was 217. It was possible to pair samples from 83 donors with a first-time FPTR, of which 56 (67.5 %) had a second FPTR. In total 0.2 % of donors were deferred because of FPTR during the observation period. There was no significant difference when comparing the second FPTR incidence among WB (65.5 %) and plasma donations (72.4 %). The average time between the initial FPTR and the subsequent donation was 0.4 years. A decreased risk of a second-time FPTR was found when the time between the initial FPTR and subsequent donation increased (OR = 0.62, 95 % CI [0.18, 2.06], $p = 0.431$). When further paring 24 first-time FPTR registered before January 2021 to a second-time FPTR in the observation period, 19 of the 107 paired second-time donations was collected more than 1 year after the initial FPTR. Of these 19, 18 had a second FPTR. Twelve out of the 19 donations were collected more than 3 years from the initial FPTR and all resulted in a second FPTR.

Summary / Conclusions: Approximately 68 % of donors with a first-time FPTR, who came in for a subsequent donation during the observation period, had a second consecutive FPTR and was deferred. This may make a second screening redundant. A decreased risk of a second FPTR with increasing time between the donations was found. Despite

of this, 12 donors who came in for a subsequent donation more than 3 years after the initial FPTR still got deferred due to a second-time FPTR. This may be due to different mechanisms leading to a FPTR: a problem related to the batch number of the screening assay or a more permanent batch independent problem.

P350 | Preliminary clinical performance and analytical sensitivity of a new multiplex blood screening assay for detection of mosquito-borne viruses on a fully automated systemM Bes¹, M Piron¹, J Groves², K Livezey³, E Ong³, T Shin³, J Cooper³, C Rubies⁴, S L Stramer⁵, S Saulede¹, J M Linnen³¹Transfusion Safety Laboratory, Banc de Sang i Teixits, Barcelona, Spain,²American Red Cross, Gaithersburg, ³Grifols Diagnostic Solutions, Inc,San Diego, United States, ⁴Grifols Diagnostic Solutions, Inc, Barcelona,Spain, ⁵Infectious Disease Consultant, North Potomac, United States

Background: The Procleix ArboPlex assay is a new multiplex assay under development for use on the Procleix Panther[®] system. The assay is a qualitative Transcription-Mediated Amplification (TMA) nucleic acid test (NAT) designed to detect multiple arboviruses (arthropod-borne viruses) including chikungunya (CHIKV), dengue (DENV), West Nile (WNV), and Zika (ZIKV) viruses. In addition, the assay cross reacts with Usutu virus (USUV), a flavivirus that shares nucleic acid sequence similarity with WNV in the target capture, amplification and detection regions.

Aims: The aim of these studies was to determine the clinical sensitivity for detection of CHIKV, DENV, WNV and ZIKV in positive specimens and specificity in individual and pooled samples of 16 blood donations. Cross-reactivity with USUV was examined in clinical samples infected with USUV. Additionally, analytical sensitivity of the ArboPlex assay was determined using WHO standards or RNA *in-vitro* transcripts (IVTs).

Methods: The sensitivity for detection of CHIKV, DENV, WNV and ZIKV was examined by testing 181 positive specimens, comprising 57 CHIKV, 52 DENV, 51 WNV, and 21 ZIKV specimens obtained from the American Red Cross (Gaithersburg, Maryland) or from clinical specimen suppliers. Specimens were tested undiluted and diluted 1:16 in singlicate across 2 reagent lots. Cross-reactivity with USUV was determined in 4 clinical samples infected with USUV. Specificity was evaluated by screening 10,000 fresh individual donations from Spain, 1,501 frozen individual samples from US donors and 1,839 16-donation pools collected in the US. The limit of detection (LoD) was determined with 3 reagent lots using WHO standards for WNV Lineage 1 (18/206), WNV Lineage 2 (18/208), ZIKV (11468/16), CHIKV (11785/16), and DENV Types 1-4 RNA IVTs. Results were analyzed by probit analyses (SAS 9.4) and compared to commercially available monoplex Procleix assays.

Results: The ArboPlex assay detected all CHIKV, DENV, WNV and ZIKV specimens tested undiluted and diluted 1:16 and cross-reactivity was detected with clinical USUV specimens. Specificity for the ArboPlex assay was 100% (95% CI: 99.97-100%) for WNV (flasher channel)

and 99.99% (95% CI: 99.95-100%) for CHIKV, DENV, ZIKV (glower channel) in individual specimens. The assay demonstrated 100% (95% CI: 99.80-100%) specificity for WNV (flasher channel) and 100% (95% CI: 99.8-100%) specificity for CHIKV, DENV, ZIKV (glower channel) in fresh 16-sample pools. The 95% LoD values for the ArboPlex assay were 3.8 International Units/mL (IU/mL) for WNV Lineage 1(18/206), 2.2 IU/mL for WNV Lineage 2 (18/208), 2.5 IU/mL for ZIKV (11468/16), and 7.7 IU/mL for CHIKV (11785/16). The 95% LoD was 17.5 copies/mL (c/mL), 11.7 c/mL, 13.2 c/mL, 13.2 c/mL, for DENV Types 1-4 IVTs, respectively. The sensitivity of the ArboPlex assay was similar or improved compared to monoplex Procleix assays.

Summary / Conclusions: The ArboPlex assay on the Panther system demonstrated high specificity and sensitive detection of CHIKV, DENV, WNV, and ZIKV with overall comparable performance to individual Procleix assays. Cross-reactivity with USUV, a flavivirus closely related to WNV, was also detected with USUV clinical specimens. The assay may provide a solution for routine blood screening in geographic regions where arbovirus outbreaks may occur unpredictably. Additionally, screening blood donors with a multiplex assay may be useful for surveillance of regional arboviral activity.

P351 | The design and implementation of laboratory automation systems into blood screening laboratories in Australia

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Background: Blood donor testing in Australia is automated with state-of-the-art testing instruments in routine use. To further enhance automation and improve staff safety, the Australian Red Cross Lifeblood (ARCL) implemented the a3600 Accelerator Laboratory Automation System into four processing centres, with connection to the Abbott Alinity s and Griffiths Panther ART.

Aims: To describe the approach to designing and implementing a Laboratory Automation System into the ARCL testing laboratories to meet the following business objectives: Eliminate unnecessary manual handling of sample tubes. Enhance laboratory efficiency and capacity to support future business growth. Provide an optimal Health, Work and Safety laboratory environment. Improve system control and sample tube tracking and traceability.

Methods: Prior to tender, ARCL formed high-level requirements for a desired future state, with a laboratory automation system (LAS) as the centrepiece. A roadmap for implementation was established that included multiple phases: A design for each processing centre, aligned with the national standardised testing process. Implementation of ISBT128 flag characters on sample barcode labels. Implementation of the system in all processing centres without disruption to routine operations. The connection of testing instruments (Alinity s, Panther ART and Alinity ci) to enable fully automated sample management.

Results: The laboratory automation design was a consultative process between ARCL and the vendors to deliver a standardised testing process while allowing for local structural differences. Due to the unavailability of some instrument connection modules at the time of design, ARCL incorporated bulk input and output modules, enabling automation of pre and post-analytical processes for testing some tubes off-track. The use of animation was employed to deliver a scaled 3D walk through of each laboratory design. This proved successful to enhance engagement by enabling staff to visualise the future state of the laboratory. The impact of implementation on routine operations was minimised through a sequential implementation strategy, both within and between processing centres to allow operations to continue with minimal impact. Initial implementation with a single tube and instrument connection to the Alinity s only, provided adequate time for familiarisation with the system, prior to implementing additional tubes and the connection to Panther ART. The sequential site by site implementation strategy enabled the inclusion of improvement opportunities and system configuration changes into the next site. The Laboratory Automation Systems were successfully implemented into all 4 ARCL processing centres by November 2023, achieving the ARCL business objectives.

Summary / Conclusions: The implementation of the a3600 Laboratory Automation into the ARCL Testing Laboratories was successful, well-received by staff and had minimal impact on routine operations due to the development of clear future state design goals, good communication and a carefully considered implementation strategy. Significant improvements have been realised, with the laboratories demonstrating increased efficiency and testing capacity and improved safety through reduced manual processes. The use of a dedicated national project team to develop standardised system designs and the sequential approach to implementation, both within and between processing centres was key to a successful implementation without affecting the quality and safety of the Australian blood supply.

P352 | Effectiveness of receiving information regarding transfusion transmissible infectious diseases and medication prescriptions from related organizations to assure blood safety

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Background: Donor interview and post-donation information are crucial to ensure blood safety. As these measures rely on the accuracy of the donor's feedback, the Korean Red Cross Blood Services receives, as an additional safeguard, information on diagnosis of transfusion transmissible infectious diseases (TTID) and on prescription of medications prohibited from blood donation from the Korea Disease Control and Prevention Agency, the Ministry of National Defense, and the Health Insurance Review and Assessment Service since 2007. The information is databased in our Blood Information Management

System and is used to disqualify donors during interview and for lookback.

Aims: To confirm the effectiveness of the system by investigating cases that resulted in disqualification of donors and prevention of release of relevant blood products.

Methods: Two categories of data received from 2021 to 2023 from related organizations were analyzed. The first category is information about 16 TTIDs. The second is prescription information for 8 medications. Number of donors deferred and blood products prevented from being released were analyzed.

Results: During the study period, 47,278 TTID diagnosis information were received. Hepatitis C (66.5%) was the most common TTID followed by hepatitis A (24.2%), acute hepatitis B (4.0%), and malaria (3.4%). Among the 304,362 donors, found to be ineligible at interview, 227 (0.07%) donors were deferred based on TTID information. Hepatitis A (109 donors, 49.3%) was the most common diagnosis leading to deferral of the donor followed by hepatitis C (28.1%) and malaria (14.0%). Four donors were deferred due to HIV infection diagnosis. 31,204,110 prescription information were received over the 3 years, with Finasteride, Dutasteride, and Isotretinoin accounting for 97% of the received data. Based on the prescription information, 27,924 donors were disqualified during interview, accounting for 9.2% of the total ineligible donors during the study period. Isotretinoin (36.1%) was the major reason followed by Dutasteride (29.6%) and Finasteride (25.7%). Regarding lookback, 1365 units of blood products, accounting for 0.02% of the total number of blood products during the study period, were subjected to lookback based on TTID information received after donation. Of these, 13 units were still in the inventory and were discarded. Among 625 units (45.8%) already released to hospitals, 10 units (1.6%) were recovered and discarded. 5752 units (0.05%) of blood products were processed during the deferral period after receiving prescription information. Of these, 5,606 units (97.5%) were still in the inventory and were discarded. 129 units (2.3%) were already released before receiving the relevant information; 34 units (26.4%) were recovered and discarded.

Summary / Conclusions: Information about TTID and medication prescription was more useful for disqualifying ineligible donors. A significant number of ineligible donors, who were in the deferral period for medications, could be identified and disqualified. Using this system, donors diagnosed with hepatitis A, which is not included in donor screening, could be deferred. This system was also useful to identify donors infected with HIV and HCV. However, as there may be errors in the information received, and as information sharing is not real-time, delays in receiving the information does occur. Nevertheless, this system plays a significant role in ensuring the blood safety as there has not been a single confirmed case of TTID since 2006.

P353 | Biotin supplement intake and detection of high plasma levels in blood donors - assessing the risk of immunoassay interference

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Background: Daily requirements for Biotin or vitamin B7 can be met through a balanced diet. In recent years, however, people are taking biotin supplements due to theoretical benefits for skin, nails and hair. Excessive exogenous biotin levels in blood coming from these supplements can potentially interfere with results of immunoassays that use the free capture biotin-streptavidin methodology, including some infectious disease assays for donor screening. This interference may be relevant in the blood bank setting due to the potential for false negative results. Biotin intake among blood donors has not yet been evaluated.

Aims: Determine the overall frequency of blood donors self-reporting biotin supplement intake as well as their plasma biotin concentration and related metabolites.

Methods: From August 2022 to March 2023, prospective blood donors from five different cities in Colombia were given a survey to obtain information on the consumption of biotin-containing supplements and on socio-demographics. Plasma biotin, biotin sulfoxide (BSO) and bisnorbiotin (BNB) concentrations were determined in samples collected right before blood donation from 100 donors who self-reported biotin consumption and gave consent to participate in the study. Measurements were conducted at an international reference laboratory using

P353 - Table 1. Odds ratio (OR) for taking biotin supplements in Colombian blood donors (n = 1899).

Variables		Crude OR (95% CI)	Adjusted OR (95% CI)
Sex	Male	1	1
	Female	4.91 (3.01 - 8.00)	5.12 (3.13 - 8.39)
Postgraduate education	No	1	1
	Yes	4.57 (2.21 - 9.44)	5.62 (2.61 - 12.10)

P353 - Table 2. Distribution of plasma biotin concentration in 100 analyzed samples from Colombian blood donors self-reporting biotin intake.

Range of biotin concentration	Total (n)
<5 µg/L (limit of detection)	88
5.0 - 9.9 µg/L	3
10.0 - 19.9 µg/L	6
20.0 - 29.9 µg/L	3
Total	100

solid phase extraction followed by liquid chromatography-mass spectrometry (LC-MS/MS). Proportions were compared using the Chi-squared test or Fisher's exact test. A multivariate logistic regression analysis was conducted with biotin consumption as the dependent variable. Statistical significance was set at p -value of <0.05 .

Results: A total of 1,899 blood donors were surveyed and 122 reported biotin intake (6.4%). Being female and having post-graduate education were strongly associated with biotin consumption (Table 1). Elevated biotin levels (>10.0 $\mu\text{g/L}$) were detected in 9% of donor samples analyzed (Table 2). BNB was quantified in 6 samples, including all those with elevated biotin levels. BSO was below the level of detection in all analyzed samples.

Summary / Conclusions: Reported use of biotin was relatively common in this blood donor population (6.4%). Assuming no biotin detection among those who did not report intake, the estimated frequency of donor samples containing biotin exceeding 10 $\mu\text{g/L}$ was 0.58%. Blood centers are likely to experience biotin positive samples. Although manufacturers of vulnerable infectious disease tests have made efforts to redesign their assays to make them more robust against biotin interference, there are commercial assays available with interference thresholds close to or below the maximum biotin concentration found in this series. To mitigate the clinical impact of an analytical error, blood centers should be aware of the prevalence of elevated plasma biotin and the threshold at which interference occurs for the donor screening assays they use.

P354 | Blood donation screening of transfusion-transmissible viral infection using two different Nucleic Acid Testing (NAT) platforms—a single tertiary care oncology centre experience

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Background: Nucleic acid testing (NAT) is used to screen transfusion transmissible infections (TTIs) in donated blood samples and provide an additional layer of blood safety. In this study, we describe our experience in screening viral TTIs using two formats of NAT: cobas[®] MPX2 polymerase chain reaction-based minipool NAT (PCR MP-NAT) and Procleix Utrio Plus transcription-mediated amplification based individual donor-NAT (TMA ID-NAT).

Aims: The objective was to assess the role of two formats of NAT: individual donor-NAT (ID-NAT) & minipool-NAT (MP NAT) for human immunodeficiency virus-1 (HIV-1), hepatitis C virus (HCV) and hepatitis B virus (HBV) and its role in blood safety

Methods: Data routinely collected as a part of blood bank operations were retrospectively analysed over a period of 70 months for TTIs. Blood samples were initially screened for HIV, HBV, HCV, syphilis by chemiluminescence and malaria by Rapid card test. In addition to serological testing, all samples were further screened by TMA-based

ID-NAT (ProcleixUltrio Plus Assay) during Jan 2015–Dec 2016, and by PCR-based MP-NAT (Cobas[®] TaqScreen MPX2) during Jan 2017–Oct 2020.

Results: A total of 48,151 donations were processed over 70 months, of which 16,212 donations were screened by Procleix Utrio Plus TMA ID-NAT and 31,939 donations by cobas[®] MPX2 PCR MP-NAT. Replacement donors and male donors outnumbered voluntary donors and female donors respectively. The overall NAT yield rate of MP-NAT was 1:2281 compared to 1:3242 with ID-NAT, during the respective time period. ID-NAT detected 5 HBV infections missed by serology, whereas MP-NAT detected 13 HBV infections and 1 HCV infection missed by serology. The proportion of donations that were both seroreactive and NAT reactive was higher with MP-NAT (59.8%) compared to ID-NAT (34.6%).

Summary / Conclusions: Cobas[®] MPX2MP-NAT had higher overall NAT yield rate compared to Procleix Utrio Plus IDNAT and confirmed a higher proportion of seroreactive donations. Due to the ease of operation, simple algorithm, cobas[®] MPX2 PCR based MP-NAT can be an effective solution for blood screening in India.

P355 | Impact on false positive rates following the introduction of new assays for donor infectious disease screening

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Background: False-positive infectious screening test results have significant implications for blood centers and donors. Blood components are discarded and additional procedures for donor notification, management, and supplemental testing are required, representing a financial burden for blood centers. On the other hand, donors may be permanently deferred from future donation, impacting the blood supply. For those who are able to be reinstated, donor anxiety related to the process can dissuade future donation attempts. Highly specific screening assays are desirable to mitigate these effects.

Aims: To evaluate and compare false positive rates (FPR) and specificity between Abbott Alinity s assays and Abbott PRISM/Architect, using data from 3 years before and 3 years after the implementation of the Alinity s system as the sole blood screening platform at the Madrid blood center.

Methods: Since 2020, screening for HIV Ag/Ab, HBsAg, Anti-HCV and Syphilis has been performed using exclusively the Abbott's Alinity s system in our center. Before 2020, these markers were screened by the Abbott PRISM (HIV, HBsAg & Anti-HCV) and Architect i-4000 (Syphilis), followed by a transition period (2018-2019) when the 3 platforms co-existed. The false

P355 - Table 1

	Platform	PRISM	Alinitys
	Period	2015-2017	2020-2022
	Total donations tested	743,976	714,318
Anti-HCV	FP / FPR (%)	704 / 0.095	278 / 0.039
	Specificity (%)	99.90	99.96
Anti-HIV	FP / FPR (%)	262 / 0.035	224 / 0.031
	Specificity (%)	99.96	99.97
HBsAg	FP / FPR (%)	58 / 0.008	133 / 0.019
	Specificity (%)	99.99	99.98
Grouped FPR (%)		0.14	0.09

reactive rate (FRR) and specificity of the HIV Ag/Ab Combo, HBsAg and HCV assays run by either PRISM or Alinity s, were calculated for 2015-2017 (PRISM) and 2020-2022 (Alinity's). For syphilis, these metrics were calculated for 2017 (Architect) and compared to the period 2020-2022 (Alinity s). Repeat reactive results were confirmed by NAT (HIV, HCV, HBV), immunoblot (HCV & HIV), supplemental HBV serology testing and TP-PA for syphilis.

Results: Table 1 shows total analyzed donations and performance measurements for HIV, HCV and HBsAg for the two platforms/periods. Notably, anti-HCV FPR was reduced almost 2.5 times with the introduction of Alinity s. HIV had a discrete improvement but HBsAg FPR was slightly higher for Alinity s. Despite this, the overall reduction in the FPR for the 3 markers was 38%. The FPR for syphilis was 0.08% for Architect and decreased to 0.053% for Alinity's.

Summary / Conclusions: The introduction of the Alinity s assays led to a decrease in the global FPR, mainly driven by an increased specificity for anti-HCV. A positive impact on unnecessary deferrals of donations and donors have been achieved with the introduction of this new screening platform.

P356 | Cleaning effect with improvement of specificity between May 2022 and December 2023 after changing ID serology screening assays to another supplier at the Hungarian National Blood Transfusion Service

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Background: Changing blood screening suppliers can have impacts on specificity and the rate of donations to be discarded. It is well known that specificity values of ID screening assays can be intermittently lower and it is important to investigate the degree and time period until specificities improve

Aims: To monitor the specificity of the Roche infectious diseases screening assays Elecsys[®] HIV DUO, Anti-HCV II, HBsAg II, Anti-HBc II and Syphilis on cobas e801 analysers over 20 months from the

introduction of the assays with 589.621 donations (90% RT (regular test) /10% FT (1-time) donors) in comparison to routine data before the switch

Methods: Calculation of specificity for different time periods, comparing it to available data from previously used routine assays and respective data from the instruction for use. Specificity was calculated by using established confirmatory assays to validate whether a sample is confirmed positive or false reactive

Results: We grouped the number of donations screened in similar time intervals for a better statistical comparability with each approx. 230.000-350.000 donations. The initial specificity of the Elecsys assays ranged from 99.83% (Elecsys Anti-HCV II) to 99.96% (Elecsys HBsAg II) in the first 8 months and continuously improved over time to specificities between 99.92% and 99.99% for the respective assays (for anti-HBc, only a reactive rate can be calculated). Routine data from a previous long-term supplier showed very similar specificity values of 99.94%-99.99% in 336.000 donations from May 2021 to April 2022

Summary / Conclusions: A continuous increase in specificity for the Elecsys ID screening assays can be seen over a time of 20 months. All values exceed the data from the instructions for use and are in good comparison with data from a previously used screening platform/assays. A single month snapshot of December 2023 may serve as a potential final specificity result to be confirmed in the following months. A possible reason is the continuous removal of false reactive and confirmed positive donors over time, thus "cleaning" the donor pool, which leads to an improved specificity. After approx. 1-1.5 years, the level of specificity reaches very high values making the Elecsys ID screening assays an excellent tool for blood

P356 - Table 1: Results

	Number of donations screened	Elecsys HBsAg II	Elecsys Anti-HCV II
Package insert data		99.98	99.85
05/2022-12/2022	234,098	99.96	99.83
01/2023-12/2023	355,442	99.99	99.94
12/2023	28,396	99.99	99.96

P356 - Table 2: Results

	Number of donations screened	Elecsys HIV Duo	Elecsys Syphilis	Elecsys Anti-HBc II
Package insert data		99.87	99.93	99.94
05/2022-12/2022	234,098	99.86	99.91	98.84 ¹
01/2023-12/2023	355,442	99.92	99.96	98.98 ¹
12/2023	28,396	99.94	99.96	98.65 ¹

¹ anti-HBc reactives were not confirmed; all HBsAg/HBV NAT negative; FT donors only (n = 2009-36467).

screening centers reducing cumbersome retesting and confirmation procedures

P357 | Abstract withdrawn

P358 | Impact of nucleic acid amplification testing on blood safety to detect hepatitis B virus, hepatitis C virus, and human immunodeficiency virus from a regional blood center in the Philippines

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Background: Blood safety is of utmost importance in transfusion service; and in the Philippines, there are 5 mandatory transfusion-transmitted infections (TTI) needed to be tested before the release of blood product for transfusion, namely: Hepatitis B virus (HBV), Hepatitis C virus (HCV), Human Immunodeficiency Virus (HIV), Syphilis, and Malaria. The approved methodology for blood screening is serology including chemiluminescent microparticle immunoassay (CMIA). Initial reactive samples are required to be sent for confirmation testing at the Transfusion - Transmitted Infections - National Reference Laboratory in Manila, Philippines. Recently, nucleic acid amplification testing (NAT) is piloted in certain blood centers in the country. The SOCCSKSARGEN Regional Blood Center, which is temporarily relocated at Cotabato Regional and Medical Center in Cotabato City, Philippines is one of the pilot sites.

Aims: The study aimed to assess the impact of NAT on HBV, HCV, and HIV on blood safety in the SOCCSKSARGEN Regional Blood Center.

Methods: Data was collected retrospectively for 31 months (01 June 2021 to 31 December 2023) wherein blood donor samples were tested simultaneously for HBV, HCV, and HIV by CMIA (Architect i2000SR) and ID-NAT (Grifols Procleix Panther) methods. All initially reactive to NAT were subjected to discriminatory testing for HBV, HCV, and HIV (d-NAT). Invalid NAT tests for discriminatory due to insufficient sample quantity and machine/sample error were excluded in the computation of reactivity rates.

Results: A total of 65,755 blood samples were screened where 59,478 (90.45%) were non-reactive to CMIA. In the 6277 (9.55%) serology reactive samples, the total reactivity rate for HBV, HCV, and HIV were 8.21% ($n = 5396$), 0.66% ($n = 434$), and 0.38% ($n = 251$), respectively; and 8 were co-infections. The remaining 0.31% ($n = 204$) were invalid due to insufficient sample quantity and machine/sample error. Only 49,020 (74.55%) samples were screened using NAT due to the unavailability of NAT reagent and machine breakdown. The reactive results in this group were 6.14% ($n = 3,011$) for both CMIA and NAT (Group 1), 0.01% ($n = 48$) for NAT only (Group 2), and 4.96% ($n = 2,429$) for CMIA only (Group 3). In group 1, the HBV, HCV, and HIV d-NAT reactivity rates were 1.94% ($n = 952$), 0.004% ($n = 2$), and 0.02% ($n = 10$), respectively. All samples in this group were tested for only 1 or 2 discriminatory tests based on the CMIA reactivity due to insufficient sample quantity. In group 2, the HBV, HCV, and HIV NAT yield rates were 0.07% ($n = 35$), 0.01%

($n = 6$), and 0.004% ($n = 2$), respectively; while 13 samples were invalid due to insufficient sample quantity. Six (6) HBV & HCV and 2 HBV & HIV co-infections were observed in this group. In group 3, the HBV, HCV, and HIV reactivity rates are 3.89% ($n = 1,907$), 0.70% ($n = 342$), and 0.37% ($n = 180$), respectively, which were sent for confirmatory testing. Overall, the true positive rates for HBV, HCV, and HIV, based on the 49,020 total number of samples where discriminatory testing were done, was 5.80% ($n = 2,844$), 0.016% ($n = 8$), and 0.06% ($n = 31$), respectively.

Summary / Conclusions: Our study reported significant reactive rates among blood donors especially for HBV and incremental detection of reactive results by NAT which were non-reactive by CMIA. Hence, inclusion of NAT in combination to serologic testing in blood screening adds another layer to blood safety which can impact the lives of many potential recipients.

P359 | Seroprevalence of syphilis during the period of the COVID-19 pandemic among blood donors of Oran University Hospital

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Background: Syphilis, a blood-borne bacterial disease caused by *Treponema pallidum*, is currently on the increase. Serology provides an indirect approach, but is not always easy to interpret in the presence of COVID-19 virus and associated antibodies.

Aims: To assess the prevalence of *Treponema pallidum* among blood donors at the blood transfusion center of Oran University Hospital during the period 2020-2023 and compare it with data from the pre-COVID19 period.

Methods: Retrospective descriptive study over a four-year period (2020 to 2023) among blood donors at the blood transfusion center of Oran, aged between 18 and 65 years. Sampling on citrated tubes: if positive or doubtful, sampling is repeated on dry tubes. Data collection: Donation Applicant Form and Donation Biological Qualification Registers. Methods and reagents used: TPHA: CYPRESS DIAGNOSTICS. ELISA: Syphilis Total Ab Kit. Biorad. Data were entered and analyzed by SPSS version 22.0.

Results: The mean age of donors was 36.3 ± 7.3 years. The highest proportion of donors was between 18 and 37 years of age (58%). Their sex-ratio (M/F) = 3.5. Almost all donors (98%) reported COVID-19 infection. The average seroprevalence of syphilis confirmed on over this period was 0.45%: 1.6 times higher than HBV, 1.3 times higher than HCV and 3.5 times higher than HIV. In the four years prior to COVID19 (2016-2019), this prevalence was 0.39%: 1.6 times higher than for HBV and HCV, and 5.6 times higher than for HIV. The prevalence of false positives was 0.15%, verified on dry tube samples and controlled by the absorption technique for non-specific reactions. COVID-19 infection could be responsible for these interferences and certain cross-reactions, as could autoimmune diseases and other mononucleosis syndromes. Syphilis seropositivity was significantly associated with age ($p < 0.001$), gender ($p = 0.001$) and donor status ($p < 0.001$): the majority of seropositive donors (70%) were occasional.

Summary / Conclusions: The interference of COVID-19 infection with the results of indirect serological tests for syphilis must be taken into account when microbiologically qualifying blood donations.

P360 | Prevalence and incidence of transfusion-transmissible infections among blood donors in Madrid, Spain from 2015 to 2022

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Background: Monitoring time trends in infectious disease rates among blood donor provides a mechanism to assess the safety of the blood supply and the effectiveness of donor deferral criteria as well as other screening measures. Variations in population risks can also be reflected on changes in blood donor infectious disease rates.

Aims: To assess the prevalence and incident of HIV, HBV, HCV, and Syphilis infections from 2015 to 2022 at the Madrid Transfusion Center.

Methods: A retrospective analysis of blood donation data was performed to determine the prevalence of markers for transfusion transmitted infections (TTI). The prevalence of confirmed-positive test results for HIV, HBsAg and HCV were evaluated for each year for first-time donors (FTD) from 2015 through 2022. Syphilis antibodies were evaluated from 2017 to 2022. Incident cases were also recorded, and risk of seroconversion was calculated for repeat donors (RD). Before 2020, TTI markers were screened by Abbott PRISM (HIV, HBsAg & Anti-HCV) and Architect i-4000 (Syphilis). During 2018-2019, the Alinity s system was gradually introduced and since 2020 it is the only screening platform in use. Repeat reactive

P360 - Table 2: Incident cases (seroconversion risk /100,000 confirmed-positive donations):

Year Repeat donors	HIV	HBsAg	HCV	Syphilis
2015 124,714	13 (10.4)	3 (2.4)	0 (0)	NA
2016 134,332	11 (8.2)	8 (6.0)	0 (0)	NA
2017 133,671	8 (6.0)	1 (0.7)	4 (3.0)	38 (28.4)
2018 138,001	13 (9.4)	0 (0)	5 (3.6)	58 (42.0)
2019 136,982	11 (8.0)	1 (0.7)	4 (2.9)	71 (51.8)
2020 129,773	7 (5.4)	8 (6.2)	2 (1.5)	54 (41.6)
2021 134,989	5 (3.7)	10 (7.4)	2 (1.5)	71 (52.6)
2022 135,878	7 (5.2)	0 (0)	1 (0.7)	72 (53.0)
2015-2022 1,068,340	75 (7.0)	31 (2.9)	18 (1.7)	364 (45.0)

results were confirmed by NAT (HIV, HCV, HBV), immunoblot (HCV & HIV), supplemental HBV serology testing and TPPA for syphilis.

Results: Measured prevalence rates for infectious disease markers among FTD (Table 1), and seroconversion risk for RD (Table 2). Although a consistent pattern was not found across the entire analyzed time frame, the prevalence of HBV decreased while Syphilis increased. Among RD the proportion of incident cases have decreased for HIV and HCV.

Summary / Conclusions: Although prevalence and proportion of incident TTI continue to be lower than those in general population, the prevalence rate for Syphilis showed a continuing pattern of increase. Demographic aspects unique of the Madrid region might explain this trend. However, these results suggests that efforts should be focused on donor education about TTI and sexually transmitted infections, particularly for first time donors.

P360 - Table 1: Prevalence First-time donations with positive test (prevalence rate/100,000 confirmed-positive donations):

Calendar year Total donations /FTDs	HIV	HBsAg	HCV	Syphilis
2015 248,381/64,091	13 (20.03)	82 (127.9)	22 (34.3)	NA
2016 249,292/58,627	8 (13.6)	61 (104.0)	36 (61.4)	NA
2017 246,303/56,308	12 (21.3)	52 (92.3)	26 (46.2)	129 (229.1)
2018 245,450/48,272	2 (4.1)	40 (82.9)	20 (41.4)	125 (258.9)
2019 244,016/46,820	7 (15.0)	44 (94.0)	19 (40.6)	133 (284.1)
2020 233,438/34,154	9 (26.4)	32 (93.7)	12 (35.1)	125 (366.0)
2021 237,887 / 35,961	5 (13.9)	24 (66.7)	14 (38.9)	119 (330.9)
2022 242,993 / 40,323	5 (12.4)	34 (84.3)	20 (49.6)	125 (310.0)
2015-2022 1,947,760 / 384,556	61 (15.9)	369 (96.0)	169 (43.9)	756 (288.6)

P361 | Comparison of the specificity of a multiplex nucleic acid test for HIV, HCV, and HBV performed on two versions of an automated analyzer

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Background: The cobas® MPX test* is a qualitative test for detection of Human Immunodeficiency Virus Type 1 (HIV-1) Group M RNA, HIV-1 Group O RNA, Human Immunodeficiency Virus Type 2 (HIV-2) RNA, Hepatitis C Virus (HCV) RNA, and Hepatitis B Virus (HBV) DNA in human plasma and serum. The assay has been in routine use for donor screening using the automated cobas 6800/8800 Systems (6800/8800 Systems), which provide medium to high throughput testing. Recently, the lower throughput cobas 5800 System (5800 System) has been developed. The cobas 5800, 6800, and 8800 Systems utilize the same cobas MPX test reagents, which are provided in ready-to-use cartridges. A previous study demonstrated that the analytical sensitivity of the cobas MPX test on the 5800 System was equivalent to that on the 6800/8800 Systems (Zhu X et al., Vox Sang 2022;117(Suppl. 1):151-152).

Aims: The aim of this study was to compare the specificity of the cobas MPX test between the 5800 and 6800 Systems.

Methods: Plasma samples from donations seronegative for HIV, HCV, and HBV were pooled into 6-member minipools (MPs) using the cobas Synergy software and a Hamilton Microlab® STAR pooler. The MPs were tested by the cobas MPX test using the 6800 System. The same MPs were also tested by the cobas MPX test using the 5800 System. Reactive pools were to be resolved by testing the individual members of the pool, with additional testing performed to resolve any results that were discordant between the two Systems. Assay specificity, with 95% Clopper-Pearson exact confidence interval (CI), was calculated.

Results: 2015 MPs comprised of samples from 12,090 donations were tested by the cobas MPX test on both the 5800 and 6800 Systems. All pools were nonreactive for all targets on both systems. Pool specificity of the cobas MPX test on both systems was 100% for all targets (2015/2015; 95% CI 99.82%-100%). The specificity of the cobas MPX test in the 12,090 donations screened in MPs was 100% for all targets (12,090/12,090; 95% CI = 99.97%-100%).

Summary / Conclusions: In this study, the specificity of the cobas MPX test was 100% on both the 5800 and 6800 Systems. These results, along with the equivalent sensitivity established in the prior study, support the comparable performance of the cobas MPX reagents on the new cobas 5800 System and the previously developed cobas 6800/8800 Systems. The cobas MPX test can be used for donor screening on any of the three analyzers, depending on laboratory throughput needs. * The cobas MPX test and cobas 5800/6800/8800 Systems are not available for use in all countries.

P362 | Performance evaluation study of immunoassays in blood donor screening in Salta, Argentina

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Background: Testing of blood donations for infectious diseases plays a key role in maintaining the safety of blood transfusions. Salta Regional Blood Center routinely screens all regular and first-timedonors with serology assays for HIV (HIV antigen/antibody), HCV (anti-HCV), HBV (Anti HBc and HBsAg), HTLV (anti HTLV I and II antibody) and Chagas (antibody). Advances in the development of blood screening tests have resulted in improved assay sensitivity and specificity, which reduces the risk of transmission of infectious diseases, providing high-quality blood derived products. A method comparison was carried out between April and August 2023.

Aims: The aim of this study was to assess the specificity performance of the Elecsys infectious diseases assays using electrochemiluminescence immunoassay (eCLIA) on the Roche cobas® e 801 platform in parallel with the routine chemiluminescence microparticle immunoassay (CMIA) on the Abbott Architect i2000.

Methods: Specificity was evaluated in 1242 serum samples from first time unselected blood donors, routinely screened on Abbott Architect i2000s platform for HIV Ag/Ab 1+2, HCV Ab, HBc Ab, HBsAg, HTLV-I/II and Chagas antibodies (Architect HIV Ag/Ab Combo, Anti-HCV, Anti-HBc II, HBsAg Qualitative II, rHTLV-I/II and Chagas). After routine testing, the samples were processed on cobas® e 801 platform for the same parameters (Elecsys® HIV Duo, Anti-HCV II, Anti-HBc II, HBsAg II HTLV-I/II and Chagas). All tests were performed according to the manufacturer's instructions. Results were interpreted as reactive, borderline or non-reactive; borderline results were considered as reactive for the respective methods. Concordant negative results were considered true negatives. Samples with an initial reactive result were repeated in duplicate and any discrepant result between methodologies were resolved using confirmatory tests. Samples with repeat reactive results but subsequently verified negative by confirmatory tests were interpreted as false reactive. The specificities of the infectious disease immunoassays on cobas® e 801 and Abbott Architect were calculated.

Results: The specificity results on cobas® e 801 were 99.60% for Elecsys HIV Duo, 100% for Elecsys Anti-HCV II, 100% for Elecsys Anti-HBc II, 99.84% for Elecsys HBsAg II, 99.92% for Elecsys HTLV and 99.92% for Elecsys Chagas. For Abbott Architect, the respective specificity for Architect HIV Ag/Ab Combo was 99.84%; 99.44% for Architect Anti-HCV, 99.84% for Architect Anti-HBc II, 99.76% for Architect HBsAg Qualitative II, 99.84% for Architect rHTLV-I/II and 99.92% for Architect Chagas. The total number of false positives were 9 for Elecsys e 801 and 17 with Abbott Architect, representing 0.7% and 1.37% of discarded blood donations, respectively.

Summary / Conclusions: The observed specificity performance data of the Roche cobas® e 801 Elecsys® assays were very similar to the respective Abbott Architect assays used in routine at the Salta Regional Blood Center, complying with the regulatory demand for

≥99.5% specificity. The findings demonstrate that Roche cobas® e 801 Elecsys® assays are highly specific and reliable tools for blood donor screening. The total number of false positives was 9 for Elecsys and 17 for Architect. This suggests that ECLIA technology may offer an advantage over CMIA technology in minimizing overall false-reactive cases, thereby reducing the burden of confirmatory testing, the risk of donor deferrals, and the loss of blood units.

P363 | Potential Impact of Improvements in turnaround time

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Background: Blood component shortages continue to act as global barriers to effective patient care. As represented by insufficient component quantity or time in available inventory (time on shelf or TOS), these shortages stem from a myriad of well-characterized systemic issues. State-of-the-art blood screening methods are available in increasing abundance outside high-income countries (HIC), and blood donor screening time to result remains a factor of high importance. The useful qualities of donor screening assays are well-documented, but time to result (TTR) remains a limitation. According to accepted benchmarks, it is estimated that total screening TTR for an individual donor is 265 minutes. The time saved by faster donor screening has the potential to increase component shelf life with positive impact on availability and patient care, as well as laboratory operations.

Aims: We aim to demonstrate the use of theoretically improved screening methodology as a potential diagnostic and donor screening assay with positive theoretical impact on TOS and blood availability.

Methods: PubMed search was performed using the keywords *Laboratory Turnaround Time, algorithms, screening, transfusion transmitted disease testing, and blood bank* with the filters: *full text, original article, review, metanalysis, and not older than 5 years*. Testing is assumed to take place at the same site on a benchtop apparatus. The theoretical assay process was laid out to ensure that assay performance leads to a result that, like existing donor screening, is actionable within a donor screening algorithm. Statistical analysis for significant difference between the observed theoretical mean TTR (TTTR) and hypothetical decreases in mean available TTR (ATTR) was performed.

Results: The theoretical benchtop apparatus tested a total of 10 samples in succession; each test completion time was 132.5 minutes (mean/median/mode 132.5). Time to result for existing screening methodology was established at 265 minutes (mean/median/mode 265 minutes). The mean TTTR was 50% (132.5/265) of mean ATTR. When compared to hypothetical, reproducible ATTR performance of 265 minutes in an independent sample population of 10 samples, the 50% TTTR reduction from 265 minutes represented a significant difference ($p < 0.01$).

Summary / Conclusions: These theoretical results demonstrate the continued potential of novel methodology implementation in donor screening. Significant decreases in TTR such as a theoretical 50% decrease in TTTR can lead to decreases in time to component release

into inventory, increases in TOS, and improved lab operations. Given the unpredictable timing of demand for components in many settings (e.g. obstetrics, trauma, and surgical theater), development of a device with this TTR and reproducibility may enable blood services to bring components to shelves more quickly, better withstand blood shortages, and positively impact patient care. In addition to donor screening and patient care implications, further opportunity to demonstrate improvements in operational productivity may be considered.

P364 | Abstract withdrawn

P365 | Evaluation of efficacy and suitability of a TTI testing algorithm to at Regional Blood Transfusion Center in South India

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Background: Every unit of donated Blood is required to be tested for TTIs by screening tests. Besides efficacy, cost-effectiveness and availability of tests, another important factor in choosing a methodology is the need to balance discard of false positive units and risk to patients due to missing detection of true negatives. This study is planned to evaluation & validate a standard two-step viral TTI screening algorithm by using an additional set of tests directed towards these viral TTIs to confirm the findings.

Aims: To review the efficacy of an existing TTI testing protocol/algorithm, by using multi-step confirmatory testing, to supplement the findings of the protocol. Also to suggest the safe & cost effective TTI testing algorithm.

Methods: At our center routine viral TTI testing involves Chemiluminescence and NAT. CLIA is done on Alinity i system (Abbott Diagnostics) and ID-. NAT is performed on Grifols Panther system. Any unit testing reactive by chemiluminescence (and/or NAT) once, is tested on Alinity I TWO more times. The samples testing positive ATLEAST TWICE out of the total 3 tests are included in the study [Level 1 testing]. These samples then undergo the following confirmatory tests [Level 2 testing]: HBV: HBsAg qualitative Confirmatory assay, Anti HBS (quantitative) & Anti HBc (Abbott Diagnostics). HCV: Line immunoassay (LIA). HIV: Western blot. All of these units are also tested for viral load for the respective viral marker.

Results: The study conducted from April-Oct 2023 & total blood screening done at the center were 15861. During the study period, total sero-reactives were HCV: 81, HBV: 86, HIV:31. The samples included in the study were HCV: 49, HBV: 49, HIV: 14. Out of these, ID NAT reactive units were: HCV:2, HBV: 36, HIV: 6. After performing additional confirmatory testing: HCV: LIA positive- 2, viral load- 2 (The same 2 cases are positive by NAT, LIA and viral load). LIA showed indeterminate results for 4 cases. HBV: The confirmatory assay for HBV was positive for 39 cases, which were also positive for anti-Hbc antibody. Anti Hbs >5mIU/mL was seen in 3 cases. HIV: The western blot was positive for the 6 cases that were positive for NAT

and viral load was positive in 5 of those cases. We found 3 NAT in the study period & all three were HBV.

Summary / Conclusions: All donors deemed reactive by the institutional algorithm were found to be true positive by the 3-step validation protocol. Besides this, taking into consideration other factors such as cost, Chemiluminescence/CLIA along with NAT can be deemed most safe & suitable protocol both in the contexts of patient safety and reduction of blood unit discard. The additional confirmatory tests conducted indicate the same.

P366 | The impact of molecular testing on the prevalence of transfusion transmissible infections

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Background: Nucleic acid testing (NAT) is a molecular technique for screening blood donations to reduce the residual risk of transfusion transmitted infections (TTIs). A major benefit of NAT screening is narrowing the time from infection to detection (window period) of the infective agent (hepatitis B, C and HIV) as well as a reduction in the transmission of occult TTIs infection.

Aims: The aim of this research is to determine whether molecular testing (NAT) increases the prevalence of TTI by doing a comparative analysis of the results of serological and molecular techniques.

Methods: In the first 10 months of 2023 a total of 43,900 blood samples from 25,320 blood donors were tested for TTI markers with a chemiluminiscent microparticle immunoassay (CMIA) for the detection of HBsAg, anti-HCV, anti-HIV1/2 and, as well as molecular technique (Nucleic Acid Amplification Test-NAT) for detection of HBV DNA and HCV/HIV/HEV RNA. Confirmatory testing was done with neutralization test for HBsAg and Immunoblot test for HIV and HCV and with discriminative NAT testing. All HEV RNA positive samples were tested with the ELFA serological technique for the detection of IgM/IgG anti-HEV.

Results: The estimated TTI seroprevalence was as follows: 60 (0.24%) for HBV, 3 (0.01%) for HCV and 1 (0.004%) for HIV. The estimated NAT-prevalence was determined: 63 (0.25%) for HBV, 3 (0.01%) for HCV, 2 (0.008%) for HIV and 26 (0.1%) for HEV. From the total of 65 HBV positive samples, 2 (3.08%) were only HBsAg positive, 5 (7.7%) only NAT positive, and 58 (89.2%) were positive by both techniques. Out of 2 HIV positive samples, 1 (50%) was only NAT positive and 1 (50%) was positive with both techniques. Out of 26 HEV RNA positive samples, anti-HEV antibodies were only present in only 2 (3.2%).

Summary / Conclusions: The prevalence of HBV DNA and HIV RNA is bigger than the prevalence of HBsAg and anti-HIV/p24 respectively. There were no differences in the prevalence between serological and molecular markers for HCV.

P367 | Abstract withdrawn

P368 | Correlation between false positive rates of HIV ELISA for HIV and COVID-19 antibody levels in blood donors in Zhejiang Province of China

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Background: Since January 2023, there has been a significant increase in the false positive rate of HIV Ag/Ab ELISA test performed at Zhejiang Provincial Center for Blood Screening Laboratory in China. This increase was primarily due to the rising false positive rate of the fourth generation HIV-Ag/Ab ELISA test kit produced by Bio-Rad (hereafter referred to as HIV BR). At the same time of the increased false positive rate in HIV testing, the data from the National Health Control Center shows that the COVID-19 infection rate in China reached its peak in December 2022 and the epidemic was basically over before the Spring Festival in January 2023, with an infection rate close to 90%. Based on the changes in antibody levels, it takes 20-30 days from the time of COVID-19 infection for the antibody level to reach its highest point. According to the antibody changes pattern, antibodies can maintain a relatively high level for 3-6 months. The spike protein chain of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) shares structural similarities with other viruses such as dengue fever and Zika virus. This protein can mediate cross-reaction between SARS-CoV-2 and these viruses, so it may interfere with immune testing and cause false positive diagnosis.

Aims: This study aims to investigate the Correlation between False Positive Rates of HIV ELISA for HIV and COVID-19 Antibody Levels in Blood Donors in Zhejiang Province of China

Methods: The study period was from February 1, 2023 to May 31, 2023. We selected 301 plasma samples from repeat blood donors who had been infected with SARS-CoV-2 or received the SARS-CoV-2 vaccine, and analyzed the samples.

Results: The results showed that the false positive rate of HIV ELISA for HIV increased significantly at the beginning of the year and reached its peak at the end of February, reaching a rate of 0.68%. Then, it gradually decreased from March onwards and returned to normal levels by the end of May. Meanwhile, the antibody level of COVID-19 in blood donors also peaked in February and then fluctuated downward. Statistical analysis showed that the changes in the two were significantly correlated ($p < 0.01$), indicating a positive correlation between the false positive rate of HIV ELISA for HIV and COVID-19 antibody.

Summary / Conclusions: Due to the limitations of ELISA methodology and the potential high false positive rate problem of HIV Ag/Ab detection kits for other pathogens, especially the fourth generation HIV-Ag/Ab ELISA kit which is prone to false positives when detecting specific pathogens, we focused specifically on the false positive

performance of such kits produced by the French Bovine Leukemia Vaccine during the COVID-19 outbreak. Given the structural similarities between the spike protein of SARS-CoV-2 and other viruses, as well as the possible cross-reactive effects of antibodies, we speculate that this correlation may be caused by the cross-recognition of antibodies produced after SARS-CoV-2 infection with the HIV antigenic sites.

P369 | To compare rapid diagnostic tests and electrochemiluminescence with enzyme linked immunosorbent assay for transfusion transmissible infections screening in blood donors

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Background: The primary responsibility of transfusion services is to provide safe blood to be transfused to the patients. Different type of methods are used for the screening of HIV, HCV and HBV infections in the donated blood, these include Rapid Diagnostic tests, Enzyme-linked immunosorbent assay (ELISA), Chemiluminescence assay (CLIA) and RT-PCR. Rapid testing is used for field Surveys, emergency screening, and home testing. ELISA is better where batch tests of donor samples are done, CLIA is suitable for BTS in emergency conditions due to short Turn around time (TAT).

Aims: 1. To compare the two different serological testing platforms i.e. Rapid diagnostic tests with Chemiluminescence testing technology for screening of Transfusion transmissible infections Infections in blood donors. 2. To compare the results obtained from these two different serological testing platforms with the gold standard test for blood donor screening ELISA testing in the context of Sensitivity, Specificity, PPV and NPV.

Methods: This study was conducted in the Department of Transfusion Medicine of a tertiary care institution of Northern India, on both voluntary and replacement donors donating at the Blood donation center and Voluntary Blood donation camps. 800 blood donor samples were tested for the detection of Transfusion Transmissible Infections by two different methods, Rapid Diagnostic Test, and CLIA (Chemiluminescence assay).

Results: A total of 800 (98.6 % male, 1.4 % female) whole blood donors, with a median age of 31.8 years (18-60). The prevalence percentages were also compared with prevalence as per the annual report 2021-22 of the department. The sensitivity of RDTs and CLIA for all three TTI's was 100% whereas the specificity of RDTs and CLIA for all three TTI's was as follows - 100% and 99.75% respectively for HBsAg, 99.87% and 99.75% respectively for Anti-HCV, 99.87% and 100% respectively for Anti-HIV. The positive predictive value of CLIA as compared to RDTs was lower for HBsAg testing (71.43% vs 100%) and Anti-HCV (75% vs 85.71%) but higher for Anti HIV (100% vs 50%) respectively. The accuracy and the Youden index calculated of RDTs and CLIA were similar and comparable for all the three tests.

The Cohens kappa coefficient shows a substantial to perfect agreement of RDT and CLIA for the three TTI's with the gold standard test (Elisa testing).

Summary / Conclusions: In our study we conclude that the overall performance of rapid diagnostic testing and CLIA testing was comparable with ELISA testing. The overall sensitivity and specificity obtained for RDTs and CLIA was comparable to ELISA testing for all TTIDs evaluated. Rapid diagnostic testing should be used in resource constrained settings where the cost is a limitation or in low annual collection centers. It can also be utilized for use for donor notification testing prior to donor referral at higher centers. CLIA testing is also comparable in our study ELISA and can be shifted to at high throughput centers where in routine ELISA testing is being done till date. The result obtained is more objective and can be obtained in shorter TAT. More false positive tests in CLIA are acceptable than missing out true positive samples because it may affect blood safety. ELISA testing is usually preferred over Rapid testing in day to day practice as objective results can be obtained on ELISA and quality control can be performed which can further help in better monitoring and managing quality testing for routine testing in the TTI Lab.

P370 | Abstract withdrawn

P371 | A proper lookback demands an unblemished traceability system - from vein to vein

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Background: In a transfusion service, an impeccable vein-to-vein traceability system from donor vein to patient vein is essential to conduct proper retrovigilance of any blood component.

Aims: The aim of this study is to review the fate of blood components from donors seroconverted for hepatitis B virus (HBV), hepatitis C virus (HCV), human immunodeficiency virus type 1 and 2 (HIV), syphilis, and human T-cell lymphotropic.

Methods: This is a retrospective and descriptive study of blood donors with confirmed infection for HBV, HCV, HIV, syphilis, or HTLV I/II over the past 10 years (from 2014 to 2023) in La Rioja, an autonomous community in Spain. As a serological donor screening assay, we conducted a chemiluminescent microparticle immunoassay determining: hepatitis B surface antigen (HBsAg), antibody to HCV, HIV p24 antigen, and antibody to HIV type 1 and 2, antibody to *Treponema pallidum*, and antibody to HTLV I/II (in donors with risk factors for this virus). Confirmatory studies included: nucleic acid amplification techniques (NAT) and specific HBsAg neutralization to HBV, NAT and immunoblot to HCV, HIV, and HTLV I/II, and immunoblot and reagent test to syphilis. We considered whether donors were new or regular/known, along with their distribution by gender and age. For regular or known donors with confirmed infection, we investigated the fate of

blood components from the last donation with negative serology and NAT results, as well as serological and post-transfusion data for the recipients.

Results: During the studied period, 100.752 donations were obtained. In 41 of them (0.04%), we confirmed one of the described infections occurred: 14 for HBV, 3 for HCV, 5 for HIV, 18 for syphilis, and 1 for HTLV I/II. Of these, 26 donors were new and 15 were regular or known. Regarding gender, 25 were males and 16 were females. The distribution by years of donors with confirmed infection is shown in the table below:

P371 - Table 1

Age	number of donors
18-30	6
31-40	15
41-50	8
51-60	9
>60	3

P371 - Table 2

	Red blood cells	plasma	buffy coat
Sent to industry	0	11	0
Negative result in the recipient	6	1	4
Not used or expired	5	2	10
Recipient deceased	4	0	1
No response from recipient's result	0	1	0

The retrovigilance study yielded the following data of 45 transfused blood components, from the last negative donation of seroconverted donors:

Summary / Conclusions: (1). The profile of a donor with confirmed infection process is a male aged 31 to 40 who donates for the first time. (2). The predominant infectious process is syphilis, accounting for 43.9% of the total. (3). In our traceability system no flaws were found, allowing for proper "vein-to-vein" traceability of blood components. (4). During this period no infections associated with transfusion have been transmitted.

P372 | Transfusion-transmissible infections among blood donors in a regional blood center

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Background: Transfusion is a life-saving procedure, and is used across a wide range of disorders and conditions. To ensure the quality and safety of blood products, donors undergo appropriate screening and

are tested for transfusion transmissible infections, and the donated blood is subsequently processed and stored according to accepted standards. The Philippine Blood Center currently serves as the regional blood center in the National Capital Region, and caters to the blood inventory needs of hospitals across the Philippine capital.

Aims: This study aims to determine the prevalence of positivity for transfusion transmissible infections that are tested in a regional blood center. These include nucleic acid testing for hepatitis B, hepatitis C and HIV, as well as serologic testing for malaria and syphilis.

Methods: We performed a review of all available test results for transfusion transmissible infections in the Philippine Blood Center. Archived test results from January 2016 until December 2023 were retrieved from the blood center's Technical Division.

Results: Over a period of eight years, our data show an overall prevalence of 0.05% for malaria, 0.66% for syphilis, 2% for hepatitis B, 0.43% for hepatitis C, and 0.17% for HIV. Through the years, the positivity for hepatitis B has been decreasing, whereas the trends have been largely unchanged for remaining TTIs.

Summary / Conclusions: The low positivity for transfusion transmissible infections reflects the effectiveness of screening procedures. Such procedures are standardized and uniformly implemented across the country. A study looking into the positivity rate among all blood service facilities can provide a better picture of blood safety, and such a study is likewise envisioned by the Philippine Blood Center. With the establishment of a national Hemovigilance Unit, national data can be shared, consolidated, analyzed, and translated into policy.

P373 | Abstract withdrawn

P374 | Analysis of exclusions of blood donors with repeatedly reactive serological results

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Background: Spanish Royal Decree 1088/2015 stipulates that in the event of a reactive serological result in a screening test, we must be re-tested in duplicate using the same assay (once on the donation unit and once on the extracted tube). Similarly, the royal decree requires that in the donation that is reactive in one or both of the repeats, blood components are discarded and confirmatory tests are performed, using a different methodology from that used for screening.

Aims: The aim of this study is to investigate exclusions in blood donors with reactive serology results from screening techniques but negative confirmatory tests, a situation necessitating donor exclusion

P374 - Table 1

Age	18-30	31-40	41-50	51-65
Number of donors	19	11	29	45

and the discarding of blood components from that donation and any previous donations, if they exist.

Methods: The study is conducted on blood donations obtained in the last 5 years (from 2019 to 2023) in the Autonomous Community of La Rioja. We define a repeatedly reactive result (RR) as one that shows positivity with the screening technique in the tube extracted during the blood donation and in at least one repetition of that same donation but yields a negative result in the confirmation techniques. As a serological donor screening assay we perform a chemiluminescent microparticle immunoassay to hepatitis B virus (HBV), hepatitis C virus (HCV), human immunodeficiency virus type 1 and 2 (HIV), syphilis and human T-cell-lymphotropic virus type 1 and 2 (HTLV I/II). As confirmatory techniques we perform: nucleic acid amplification techniques (NAT) and specific HBsAg neutralisation to HBV, NAT and immunoblot to HCV, HIV y HTLV I/II and immunoblot and reagent test to syphilis. Data regarding observed infectious pathology and its distribution by sex and age are collected. We take into account whether the donor has donated on other occasions or is new donor.

Results: The number of donations obtained in La Rioja during this period is 49,524, of which 104 (0.20%) show RR results. From the entirety of RR results: 11.53% are HBV, 34.63% are HCV, 16.34% are HIV, 29.81% are syphilis, and 7.69% are HTLV I/II. Of these, 66.34% are male and 33.66% are female.

The age distribution is shown in the table below:

Furthermore, 78.85% have donated at least once before, while 21.15% are new donors.

Summary / Conclusions: (1). The 0.20% of blood donations are discarded and their donors excluded. (2). The majority of donors with RR results are regular donors, male over 50 years old, and HCV RR results are the most frequent. (3). It is possible that some age-associated factor (inflammatory, rheumatologic, medications, etc.) may be responsible for these results. (4). It is challenging for professionals to explain to donors that despite not having an infectious pathology, they cannot donate again until the results turn negative. (5). This action does not impact transfusion safety but does affect donor loss. Therefore, we pose this question: should we reconsider some modification of the current regulations to change this situation without compromising transfusion safety?

Transfusion transmitted infections—Hepatitis B (HBV)

P375 | Ten-year follow-up program for donors with occult hepatitis B virus infection in Dalian, China

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Background: Occult HBV infection (OBI) constitutes a risk of HBV transfusion-transmission that is associated with low levels of viral DNA undetectable with NAT and potentially infectious. The viral mechanisms associated with OBI appear to be multifactorial but poorly understood, and the long-term clinical outcome in OBI donors is unknown.

Aims: Donors with OBI confirmed by negative HBsAg repeat testing and partial sequencing of HBV genome were followed-up to assess the evolution of HBV serological markers and the genetic evolution of the infecting HBV strains.

Methods: A follow-up program of OBI donors has been implemented in the Dalian blood center, China. HBsAg+ donors were included in follow-up as controls. Donors were called back at 6-months intervals and a whole blood sample was collected. HBsAg, anti-HBc, and anti-HBs were tested with CLIA. Viral load (VL) was measured with an in-house qPCR (LoQ: 20 IU/mL). Viral particles were concentrated from 6 mL plasma by precipitation with 12% PEG. HBV whole genome, Pre-Core/Core, Pre-S/S, and BCP regions were amplified and sequenced.

Results: Between 2011 and 2023, sequential blood samples (2-8/donor) were collected from 98 OBI donors and 15 HBsAg+ controls over an average period of 56 months (range: 5-149) and 30 months (range: 13-67), respectively. OBI donors were 79% males, with an average age of 38.5 years (18-55), and 15% reported having been vaccinated against HBV. 56% were repeat blood donors, 35% first time donors, and 9% platelet donors. HBsAg+ controls showed no significant changes in clinical and virological markers. During follow-up, OBI donors had ALT levels constantly <50 IU/L and undetectable HBsAg. At index time, 37% were anti-HBc+ only, 45% anti-HBc+/anti-HBs+, and 13% anti-HBc-/anti-HBs+, of which 4 seroconverted to anti-HBc 4-64 months post-index. Primary OBI was confirmed in 4 donors. No change over time in anti-HBs levels was observed in 37 (52%) donors (median level: 40 IU/L [range: 10-1000]), while anti-HBs was transiently detectable in 19 (27%) donors, 9 (13%) donors showed a transient (≥ 5 -fold) increase, and 6 (8%) seroconverted to anti-HBs. Over time, HBV DNA was detected continuously in only 16% of OBIs, transiently at least twice in 31% of cases, and only indexically in 53% of cases. 22 NAT non-reactive OBIs in follow-up were confirmed HBV DNA reactive after viral particle concentration. At index and in follow-up, viral load was undetectable or <200 IU/mL in OBIs. The median VL in controls was 117.5 IU/mL (range: 43-3 $\times 10^8$). At least one HBV sequence was available at index time and/or in follow-up for 76 (77.5%) OBIs and 13 (87%) controls. HBV genotypes were B (12%), C (84%), and D (2%) in OBIs, and B (85%) and C (15%) in controls. Pre-S/S aa sequences divergence in an individual at two-time intervals (range: 8-91 months) was analyzed in 19 OBIs including 11 anti-HBs+. Results showed 0% to 2% nucleotide variability over time in OBIs irrespective of anti-HBs status. A similar intra-donor diversity was observed in HBsAg+ controls.

Summary / Conclusions: Anti-HBs and HBV DNA variations observed in OBIs over time suggested a continuous balance between viral replication and host immune control. Comparable very limited rates of viral genetic changes over time were observed in OBI and non-OBI donors. No adverse clinical sign was associated with OBI during follow-up.

HBV DNA-containing particles persist at barely detectable level in the blood and remain potentially infectious, supporting permanent deferral of donors with OBI.

P376 | Abstract withdrawn

P377 | Abstract withdrawn

P378 | Detection of early acute, late acute, and occult hepatitis B infections by use of an assay with enhanced sensitivity in the National Blood Centre, Kuala Lumpur (NBCKL)

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Background: In 2019, the Ministry of Health in Malaysia launched the National Strategic Plan for Hepatitis B and C (NSPHBC) to diagnose 90% of the population, reduce hepatitis-related mortality by 65%, and provide treatment to 90% of those who need it by 2030. The plan emphasized the need for increased screening for viral hepatitis through laboratory testing, especially in blood transfusion settings. The National Blood Centre in Kuala Lumpur (NBCKL) conducts serology and NAT tests to screen all blood donations routinely for HBV. NBCKL had reported 0.063% of 232,295 donations tested positive for HBV in 2021, and 0.073% (269,386 donations) in 2022. In October 2022, the Transfusion Microbiology Laboratory (TML) of NBCKL switched from using the Architect i2000sr to the Alinity i system for transfusion-transmitted infection (TTI) serology screening. The new system introduced by Abbott Laboratories also had a new improved assay called the HBsAg Next Assay (HBsAg Nx), which has better detection capabilities than its predecessor, HBsAg Qualitative II assay (HBsAg Qual II).

Aims: This study evaluated the improved sensitivity of the HBsAg Nx assay in detecting early acute, late acute, and occult Hepatitis B infections. The findings shall offer NBCKL comprehensive insights into the performance of the newly introduced assay.

Methods: This cross-sectional study utilized 253 archived plasma samples which were Abbott HBsAg Qual II Non-Reactive collected between December 2019 and January 2023. These HBsAg Qual II NR archived samples were further categorized as 166 OBI (ID-NAT detected & Anti-HBc RR), 54 NAT yield (ID-NAT detected & Anti-HBc NR), and 33 late acute (ID-NAT not differentiated & Anti-HBc RR). All were tested with HBsAg Nx Assay. The initially reactive samples underwent duplicate testing. Consistently reactive samples (in 1 or 2 replicates) to HBsAg Nx were further confirmed using the HBsAg Next (HBsAg Nx) Neutralisation assay (Abbott). Additionally, the clinical evaluation of HBsAg Nx assay was also carried out. Descriptive analysis is depicted by the proportion (%) of plasma samples in each group (NAT yield, Occult B infection (OBI), and late acute), with categorical data presented as frequency (percentage).

Results: The clinical evaluation of HBsAg Nx showed 100% sensitivity with known positive HBV samples and 99.86% specificity with

random samples. Among the 253 archived samples (HBsAg Qual II Non-Reactive, NAT Reactive), 30 (11.9%) tested reactive with HBsAg Nx assay. These 30 samples were confirmed positive with the HBsAg Nx Neutralisation test. Out of these 30 confirmed HBsAg positive samples, 23 were OBI, 6 NAT yield, and 1 late acute. The incremental detection rates by the HBsAg Nx assay were 13.9% for OBI, 11.1% for NAT yield, and 3% for late acute cases.

Summary / Conclusions: The HBsAg Nx assay showed improved HBsAg detection by 11.9%, particularly in samples previously classified as OBI, NAT yield, and late acute, where HBsAg was initially absent with HBsAgQual II on Abbott Architect. The adoption of HBsAg Nx assay may benefit countries relying only on serology for TTI screening. For centres using both serology and NAT testing, improved detection reduces non-concordant cases, allowing shorter follow-up for donors with concordant results and significant time and cost savings.

P379 | Five-year analysis of prevalence, trends and demographic characteristics of HBV infection among blood donors at National Blood Centre Kuala Lumpur, Malaysia

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Background: Screening donated blood for HBV is mandatory due to blood transfusion being a potential route of HBV transmission. Understanding the prevalence, trends, and demographic patterns of HBV infection among blood donors is essential for ensuring the safety of blood products. Malaysia introduced the hepatitis B vaccination program for infants in 1989, and it may likely influence the epidemiology of HBV infection in Malaysia.

Aims: This study aimed to analyze the prevalence, trends and demographic characteristics of HBV infection, including occult HBV infection (OBI), among blood donors who donated at The National Blood Centre Kuala Lumpur (NBCKL) Malaysia over a five-year period.

Methods: A retrospective analysis was conducted for HBV infection cases from 2019 to 2023. Data on HBV infection cases were collected prospectively from 2019 onwards. The total number of donations ($n = 957,492$) were obtained via Blood Bank Information System (BBIS). HBV infection cases were categorized into concordant (both serology reactive and NAT detected), NAT yield (serology non-reactive but NAT detected), and serology yield (serology positive but NAT non-reactive). Additionally, OBI were further identified by performing anti-HBc test on NAT yields. NAT screening utilized the Grifols Procleix Ultrio Elite assays on Panther instrument, while serology screening and anti-HBc test were performed with Abbott Alinity. The prevalence rates were calculated, and comparisons were made among different HBV infection cases and among five different age groups (below 20, age 21 to 30, age 31 to 40, age 41 to 50, and above 50). Trends over the five-year period were also examined.

Results: Over the five-year period, 743 HBV infection cases were identified, and overall prevalence rate was 0.078%. The prevalence rates for concordant ($n = 576$), serology yield ($n = 58$), NAT yield ($n = 109$), and OBI ($n = 68$) cases were 0.060%, 0.006%, 0.011%, and 0.007% respectively. It was observed that for the overall HBV infection cases, yearly prevalence rate decreased steadily from 2019 at 0.095% to 2021 at 0.074%, but rose to 0.081% in 2022, and subsequently declined in 2023 (0.056%). The majority of the cases occurred in the 31 to 40 years old group ($n = 273$, 37%), followed by 41 to 50 years old group ($n = 219$, 30%). Only 3% ($n = 25$) belonged to below 20 age group. The 31 to 40 age group had the highest prevalence rate at 0.029%, followed by 41 to 50 age group at 0.023%, while the lowest was 0.003% for below 20 age group. As for OBI, the prevalence rate peaked at 0.009% in 2021, but later dropped to 0.005% in 2023. Distribution of OBI cases were almost equal among age groups 31 to 40 (32%, $n = 22$), 41 to 50 (32%, $n = 22$) and above 51 (30%, $n = 20$). Notably, their prevalence rates were also almost equal, 0.0023% for both 31 to 40 and 41 to 50 age groups, and 0.0021% for above 51 age group. No OBI cases belonged to below 20 age group.

Summary / Conclusions: This study provides valuable insights into the prevalence, trends, and demographic characteristics of HBV infection, including OBI, among blood donors donated at NBCKL. The observed fluctuations in prevalence rates over the five-year period underscore the dynamic nature of HBV transmission. These findings underscore the importance of ongoing surveillance and targeted interventions to address HBV infection, particularly among adults in their 30s and 40s. The hepatitis B vaccination program for infants in 1989 likely plays a role in the observed epidemiology of HBV infection at NBCKL.

P380 | Characterization of anti-HBs titers in blood donors with anti-HBc reactive results in an upper-middle-income country hospital blood bank

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P380 - Table 1. Serologic profile of anti-HBc reactive donations.

State HBV Infection	Total
Occult HBV Infection (OBI)	0 (0%)
Resolved Infection (HBV NAT-; HbsAg- / anti-HBc+ CMIA & ECLIA; anti-HBs+ ECLIA; anti-HBc IgM- ECLIA)	145 (89%)
Chronic HBV (HBV NAT+; HbsAg+; anti-HBc+ CMIA & ECLIA; anti-HBc IgM- ECLIA; anti-HBs [ECLIA]-)	1 (0.6%)
Anti-HBc+ only (both: CMIA & ECLIA)	9 (5.5%)
False Reactive anti-HBc	8 (4.9%)
Total	163 (100%)

Background: Colombian regulations require both a test for hepatitis B surface antigen (HBsAg) and a test for total antibody to hepatitis B core antigen (anti-HBc) for blood donor screening. However, this strategy results in a significant loss of blood donors due to anti-HBc reactivity impacting the blood supply. The Japanese Red Cross successfully introduced the evaluation of anti-HBs titers to accept some HBsAg-/HBV NAT-/ anti-HBc+ donations.

Aims: To characterize the serological profile of anti-HBc reactive donations from a Colombian hospital blood bank and evaluate the impact of introducing anti-HBs titers in reducing the discard of anti-HBc reactive units.

Methods: We tested 29,185 donations from May 2022 to January 2024 for HBsAg and anti-HBc using a Chemiluminescent microparticle immunoassay (CMIA). In addition, HBV molecular screening by multiplex Nucleic Acid Test (NAT) was performed in mini pools of 6. Anti-HBc reactive donations were subsequently tested by supplementary assays using Electrochemiluminescence immunoassay (ECLIA): total anti-HBc, Anti-HBc IgM and anti-HBs titration. Units with anti-HBs titers ≥ 200 mIU/mL were identified as potentially safe for clinical use.

Results: The reactive rate for anti-HBc by CMIA was 0.56%, equivalent to 163 reactive donations. Women made 51.5% of these donations, 92.6% were whole blood donations, and 13.5% came from repeat blood donors. The average age of anti-HBc reactive donors was 45.55 (SD: ± 11.8) years. The agreement between CMIA and ECLIA was 95.1% for anti-HBc detection. Table 1 shows the serologic profiles of anti-HBc reactive donations. In total, 124 (76.1%) anti-HBc reactive donations had titers >200 mIU/mL and 11% had lower titers < 30 UI/L for anti-HBs. All donors were non-reactive for anti-HBc IgM (ECLIA).

Summary / Conclusions: We observed that 76.1% of anti-HBc only reactive donors had levels of anti-HBs above the threshold for considering their blood safe for transfusion. No case of Occult Hepatitis B infection (OBI) was identified in this population, despite an intermediate prevalence of HBV in Colombia. The present study provides information to reconsider existing algorithms for the management of HBV donor and donations in the country.

P381 | Prevalence of occult hepatitis B Virus infection and HBV DNA detection among Slovenian blood donors

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Background: Blood donations are screened for HBsAg and HBV DNA (NAT screening) to detect hepatitis B virus. However, there is still a residual risk of HBV transmission by blood components from "window period" donations in early acute infection and donors suffering from occult HBV infection (OBI). To determine the prevalence of OBI in Slovenian blood donors, we performed anti-HBc screening on blood donations, with anti-HBs testing on anti-HBc reactive donations.

Since >100 IU/L of anti-HBs antibodies are supposed to be protective against HBV infection by blood transfusion, donations with anti-HBs <100 IU/L can be a concern regarding transfusion transmission if there is HBV DNA present in the blood.

Aims: Our aim was to determine the prevalence of OBI donors in the Slovenian blood donor population, after the introduction of mandatory HBV DNA screening in the year 2007 and universal vaccination of children in 1998. With an additional enrichment method for HBV DNA negative donations, that were anti-HBc reactive with <100 IU/L anti-HBs, we wanted to find out, if there's any residual risk still present in these donations.

Methods: 5706 blood donations were screened for anti-HBc with Abbott Architect Anti-HBcII Assay on Architect i2000SR System from January till March 2019. Reactive donations were additionally tested with Abbott Architect Anti-HBs Assay. We used ultracentrifugation at 250,000 g for 3 h at 4°C for the enrichment of HBV DNA in anti-HBc positive samples, with <100 IU/L anti-HBs. Pellets were resuspended in nuclease-free water and tested with Procleix Ultrio Elite Assay on Procleix Panther System. In case of a reactive result, confirmation was performed with Cepheid Xpert HBV Viral Load Assay on the GeneXpert instrument.

Results: Among screened blood donations there were 66% males and 43% females, from 18 to 65 years old. Out of 5706 anti-HBc screened donations, we gained 73 (1.28%) anti-HBc reactive results. We didn't detect HBV DNA in any anti-HBc reactive donation with our NAT screening. There were >100 IU/L anti-HBs present in 37 (50.68%) anti-HBc reactive donations. 36 (49.32%) anti-HBc reactive donations had <100 IU/L anti-HBs and were further ultracentrifuged. After ultracentrifugation of all 36 donations, we got one reactive result with Procleix Ultrio Elite Assay. Viral load for this sample was also detected with quantitative assay result <10 IU/mL HBV DNA. There were no anti-HBs antibodies present in this donation.

Summary / Conclusions: Compared to previous results of anti-HBc prevalence in blood donors in Slovenia, there's been a drop in OBI donations. In the year 1998, we had 3.8% and in 2007 we had 3.0% anti-HBc reactive donations. Now younger donors are already vaccinated and protected against infection, while some OBI donors were already deferred from donating due to HBV DNA detected with our routine NAT screening. Even with HBV DNA and HBsAg negative blood donations after the mandatory screening, there is still a residual risk of HBV transmission with transfusion from OBI donations without anti-HBs or with <100 IU/L anti-HBs present in the blood. With ultracentrifugation, we managed to detect low-level HBV DNA in 1/36 anti-HBc reactive donations with <100 IU/L anti-HBs. Since there were no neutralizing antibodies available in the donation, this donation could pose a potential risk for transmission of HBV through the donated blood products.

P382 | Results with full testing algorithm for hepatitis B in the donor population of Albania

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Background: Hepatitis B has a high seroprevalence in our donor population (4%). Nucleic Acid Testing (NAT) yield has also been reported high, overall 1:217 donations. This high NAT yield suggests for a high prevalence of donations with occult hepatitis B (OBI). For better investigation of OBI and for a better follow up of our periodic donors we added this year also anti-HBc and anti-HBs testing for all NAT reactive donations.

Aims: Evaluating our first results with full testing algorithm for hepatitis B and based on results establishing an accurate follow up of periodic donors that result NAT reactive.

Methods: All donated blood in our blood centre is first tested with serological methods for HBsAg, anti-HCV, HIV Ag/Ab Combo and syphilis TP on Abbott Architect system. Serology negative donations are further tested individually with NAT multiplex agent (MPX- HBV, HCV, HIV) on the Roche Cobas 6800 system and triplex test (Procleix Ultrio Elite Assay- HBV, HCV, HIV) on Panther Grifols system. All initially reactive donations with NAT are discarded. Samples are tested in triplicate and subsequently with anti-HBc and anti-HBs in order to establish for permanent deferral of the donor for follow up and eventually readmission. We included in this study all the samples tested during October-December 2023 when we introduced for the first time anti-HBc and anti-HBs testing for all NAT reactive donations. Total number of donors tested 8636.

Results: Out of the 8636 samples tested, 66 resulted HBV DNA reactive. Divided by age group we had 2 (19-30 years old), 26 (31-45 years old) and 38 (46-60 years old). Among these 62/66 (94 %) were anti-HBc positive. Only 4/66 (6 %) resulted anti-HBc negative. All four anti-HBc negative donors were less than 40 and 2 of them less than thirty years old, part of the vaccination program for hepatitis B that has begun in the mid-1990s in our country. 29/66 (44%) samples were only initially reactive, and among them 25/29 (86.3%) were anti-HBc positive and 4/29 (13.7%) were anti-HBc negative. 25 donors that were only initially reactive and anti-HBc positive, were all in the age group 38-60 years old. Among them, only 8 had also anti-HBs positive and 17 had anti-HBs negative. The remaining 37 samples (56%) were repeatedly reactive and anti-HBc positive.

Summary / Conclusions: NAT has increased safety of transfusions in our country and specially for immunocompromised patients. 97% of NAT reactive donors are above 30 years old, that means not vaccinated for hepatitis B. There is a high prevalence of anti-HBc in our donor population that correlates with high seroprevalence of hepatitis B in Albania. Full testing algorithm for hepatitis B with anti-HBc and anti-HBs gives the possibility for accurate follow-up and readmission of donor for donation.

P383 | Analysis of anti-HBc, anti-HBs, and NAT markers in HBsAg-negative blood donors

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Background: The screening of hepatitis B (HBV) in Brazil consists in HBSAg, anti-HBc and nucleic acid amplification test (NAT). Globally, HBV transmission by blood products is recognized as a transfusion risk.

Aims: To analyze the results of anti-HBc, anti-HBs, and NAT markers in HBsAg-negative donors called for re-draw to confirm positive or indeterminate results.

Methods: Positive donations for HBV markers (HBsAg, Anti-HBc, and NAT-HBV) from January to December 2019 at the Hemotherapy Service of Hospital São Vicente de Paulo, Passo Fundo/RS, were included. Confirmatory tests for HBV were analyzed. HBV screening was performed using chemiluminescence (Abbott), and NAT HBV Bio-manguinhos. Donors with positive results underwent a new sample collection, the tests were repeated, and reactive or indeterminate samples were further tested for HBSAg, anti-HBc IgG and IgM or total and anti-HBs using electrochemiluminescence (Roche) in a supporting laboratory.

Results: Of the 13,537 donations recorded, 165 (1.2%) were positive for any HBV markers. Out of serologically ineligible donors, 122 (73.9%) returned for re-draw and result confirmation. A total of 110 donors with negative HBSAg were selected for analysis. Among these, 59 (53.6%) were positive and 14 (12.72%) were negative for anti-HBc IgG; 1 (0.9%) was positive and 72 (65.45%) were negative for anti-HBc IgM; 32 (29.0%) were positive and 5 (4.5%) were negative for total anti-HBc. Regarding anti-HBs, 93 (84.5%) donors were positive and 17 (15.4%) were negative. Analyzing donors with positive anti-HBc, 80 (87.9%) had positive anti-HBs and 11 (12.1%) had negative anti-HBs. Among donors with negative anti-HBc, 13 (68.4%) had positive anti-HBs, and 6 (31.6%) had negative anti-HBs. Regarding NAT, all analyzed donors had a non-detectable result. Anti-HBc is the most prevalent marker on blood donors in the Southern region of Brazil (1.36%), as shown in Hemoprod (2019), a document compiling national production data. In some countries, anti-HBc is not a mandatory donor screening marker; many countries use HBsAg as the sole HBV marker. Studies report that the total anti-HBc test has contributed to reducing post-transfusional HBV infection incidence. However, in endemic areas for hepatitis B, its use may lead to a decrease in blood component availability due to donor rejection. Some authors recommend anti-HBc IgM as an alternative in these areas. Additionally, some countries use anti-HBs in cases of positive anti-HBc to determine the course of the donation.

Summary / Conclusions: Hepatitis B remains a public health issue in various countries. Further studies are needed to define the best

strategy for HBV screening in donors, ensuring transfusion safety without compromising blood component availability. Moreover, it is necessary to consider the specificities of each location concerning the endemicity of this disease, as well as the possibility of implementing NAT testing.

P384 | The potential of hepatitis B virus infection in NAT non-discriminated reactive blood donors in Southern Taiwan

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Background: Since 2013, Taiwan Blood Services Foundation (TBSF) had implemented mini pool (MP) nucleic acid testing (NAT) to screen for HBV, HCV, and HIV, which significantly reducing transfusion-transmission risks. However, NAT reactive with non-discriminated reactive (NDR) results have been observed during the donor screening process, possibly linked to occult HBV infections (OBIs). Moreover, among the NAT reactive donors, the occurrence of NDR has increased over the past years, increasing the residual risk of transfusion-transmission HBV infection.

Aims: This study is aimed to further analyze the molecular and serology test among NDR blood donors in southern Taiwan, evaluating potential results risk for transfusion-transmission HBV infection.

Methods: From April to November 2023, the Kaohsiung Blood Center recruited 656,427 blood donors. Some of them are NDR donors and were invited to participate in further experiments. These experiments included molecular testing (consisting of repeated Ultrio Elite Triplex NAT and repeated Ultrio Elite discriminatory HBV (dHBV) testing) and HBV serological testing. HBV DNA positive was defined as any reactive result of dHBV testing.

Results: Out of the 626 NAT-reactive blood donors, 69 were identified as NDR donors. Among these NDR donors, 20.3% had a history of NDR or were HBsAg+/ Neutralization- in previous donations. Among the 30 enrolled NDR donors, 76.7% tested positive in Triplex NAT and/or dHBV tests, and 73.7% were HBV DNA positive. The average S/CO values of Triplex NAT and dHBV were 11.94 ± 2.34 and 19.19 ± 3.74 , respectively. Comparison with the manufacturer's package insert suggests that HBV DNA viral load in serum could be very low. Additionally, 83.3% tested positive for Anti-HBc, 56.7% for Anti-HBs, 36.7% for Anti-HBe, and 26.7% for Anti-HBc+/Anti-HBs+. Furthermore, 13.3% had Anti-HBs levels greater than 100 mIU/mL and none of the individuals tested positive for HBsAg and HBeAg. Notably, 56.7% of enrolled NDR donors were HBV DNA+/Anti-HBc+/HBsAg-, and among HBV DNA positive donors, 77.3% were Anti-HBc positive.

Summary / Conclusions: The majority of HBV DNA-positive cases among NDR donors highlight a residual threat of transfusion-transmitted HBV infection. Further experiments are necessary to confirm and address this potential risk. Additionally, over half of the HBV DNA+/ Anti-HBc+ /HBsAg- results in NDR donors might have occult

HBV infection, and implementing anti-HBc testing in NDR donors could be a beneficial strategy to enhance blood safety.

P385 | Liver fibrosis in Chinese blood donors with occult hepatitis B virus infection

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Background: Occult hepatitis B infection (OBI) which means hepatitis B surface antigen (HBsAg)-negative/hepatitis B virus (HBV) DNA-positive is significantly associated with HCC, but the impact of OBI on the severity of liver fibrosis remains unclear.

Aims: A total of 122 OBI blood donors and 284 healthy blood donors were selected and tested to investigate whether OBI status was associated with liver fibrosis severity.

Methods: The serum samples of OBI blood donors and healthy blood donors were tested for viral load, HBV markers, liver fibrosis indexes and the values of liver stiffness measurement (LSM) were tested by Fibroscan. The PBMCs of OBI blood donors and healthy blood donors were sent for transcriptome sequencing after HBcAg stimulation.

Results: OBI blood donors had significantly higher levels of LSM values than healthy blood donors without OBI ($p < 0.05$). And the proportion of liver fibrosis patients ($LSM \geq 7.3 \text{ kPa}$) in OBI blood donors was higher than that of healthy blood donors (8.2% vs 3.8%, $p < 0.05$). The sequencing results showed that after HBcAg stimulation, the expression levels of pro-inflammatory factors such as IL-1 α , IL-6, IL-17A, IL-17F, IL-22 were significantly upregulated in PBMCs from OBI blood donors compared with that from healthy blood donors.

Summary / Conclusions: OBI status appears to indicate a higher risk of liver fibrosis.

P386 | Abstract withdrawn

P387 | Utilization of automated real-time PCR technique n HBsAg negative donors enhances the safety of blood transfusions

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Background: The prevalence of HBV infection in Vietnam is notably high, with rates reaching up to 15%. In Ho Chi Minh City, the rate of occult hepatitis B infection (OBI) among blood donors is estimated to be around 0.06%. The main concern regarding HBV transmission through blood transfusion lies in

donations that test negative for HBsAg, as they might have been collected during the OBI phase or the HBsAg negative window phase of the infection.

Aims: This study aimed to: (1) assess the utilization and effect of real-time PCR testing, and (2) calculate the rate of HBV DNA positivity among HBsAg negative voluntary blood donors at the Cho Ray blood transfusion center of Cho Ray Hospital in Ho Chi Minh City, Vietnam.

Methods: In this study, 64,423 voluntary blood donors were included between February 2023 and June 2023. Initially, all first-time donors underwent pre-donation screening using an immunochromatographic Alere Determine HBsAg 2 rapid test (Abbott) and allowed to donate blood when tested negative. Subsequently, the rapid test negative first-time donors in addition to the repeat donors further tested using Elecsys[®] HBsAg II electrochemiluminescence immunoassay (ECLIA) on cobas e 6000 system (Roche). The positive samples were recorded and identified, while the remaining HBsAg negative samples underwent a subsequent mini-pool six testing utilizing a real-time cobas[®] MPX nucleic acid test (Roche Diagnostics GmbH, Mannheim, Germany) on a fully automated cobas[®] 6800/8800 system. HBV DNA mini-pool six test followed by individual testing if the pool tested positive.

Results: Out of 64,423 voluntary blood donors, 26,963 (41.85%) were first time, whereas 37,460 (58.15%) were repeat donors. Among the first-time donors, 745 (2.76%) cases tested positive for HBsAg through the rapid test. The remaining first-time donors and repeat donors, totaling 63,678 individuals, underwent HBsAg testing using the ECLIA technique. A total of 159 (0.25%) donors were found to be HBsAg positive. Out of these donors, 145 (0.23%) were first time and 14 (0.02%) were repeat donors. Lastly, the cobas[®] MPX test was performed on samples of 63,100 HBsAg negative donors. This test successfully identified an additional 36 (0.057%) HBV DNA positives. Among these, 22 (0.035%) were first time and 14 (0.022%) were repeat donors. The cobas[®] MPX HBV DNA positive results indicate that 1 in every 1,753 donors may be in the early window period of the infection or OBI.

Summary / Conclusions: The present study reveals that the utilization of the cobas[®] MPX test on a fully automated cobas[®] 6800/8800 system has successfully identified an additional 36 HBV positive donors that were overlooked by other serological methods. Consequently, the utilization of an automated real time PCR technique in donor screening improves the identification of low viral loads during the HBsAg negative phase of HBV infection. This approach can enhance the safety of blood transfusions by ensuring the accurate identification of HBsAg negative but HBV DNA positive infected individuals.

P388 | Evaluation of a HBsAg qualitative immunoassay in the Central Blood Transfusions Service of Indonesian Red Cross (CBTS-IRC)

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Background: At the Blood Center, testing all donor blood for the presence of infectious diseases plays an important role in maintaining the safety of blood transfusions. In Indonesia, blood donor serology tests are mandatory for HIV, HBV, HCV and Syphilis. High specificity and sensitivity testing with automated assays is essential for this purpose. Alinity-i HBsAg Next Qualitative Immunoassays to detect the quality of hepatitis B surface antigen (HBsAg) in donor serum or plasma as a screening test to prevent transmission of HBV to blood recipients.

Aims: To enable the performance of Alinity-i HBsAg Next Qualitative Immunoassays in terms of sensitivity and specificity using the Central Blood Transfusion Service Protocol - Indonesian Red Cross (CBTS-IRC), to verify whether it can be used for blood donor examination at the Blood Center. Next, the evaluation results can be used as a reference for donor blood screening examinations at other Blood Centers of Indonesian Red Cross.

Methods: A total of 900 samples were characterized (500 negative, 400 HBsAg positive), 10% of the total samples were included in Lot to Lot verification using 3 different reagent lots. Precision testing is carried out using 1 weak positive sample and is run 20 times in one process. Precision results with target acceptance criteria CV ≤5%. The Between Day test was carried out using 1 weakly positive HBsAg sample and was carried out repeatedly within 20 days. The sensitivity and specificity were calculated using the following acceptance criteria: Sensitivity > 99.5%, specificity ≥99.8%.

Results: Sensitivity and specificity of Alinity-i HBsAg Next Qualitative Immunoassays using one set of reagents found sensitivity of 100% and specificity of 100%. Lot to lot verification is within the acceptable limit of 100%. The precision (%CV) obtained was 2.98%, which is within the acceptance criteria of CV≤5%. Between Day (%CV) obtained 2.1% which is within the acceptance criteria of CV≤5%.

Summary / Conclusions: In our evaluation, the Abbott Alinity-i HBsAg Next Qualitative Immunoassays is suitable for HBsAg screening of donor blood at the Blood Center of Indonesian Red Cross.

P389 | Automated mini pool nucleic acid testing - experience of routine blood donor screening at a hospital based blood centre

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Background: Nucleic acid testing (NAT) has revolutionized blood screening, enabling the detection of viral infections with enhanced

sensitivity. The latest automated NAT platform, cobas[®] 5800 system, offers advanced capabilities for accurate and efficient donor screening for transfusion transmitted infections.

Aims: This study aimed to evaluate the performance of mini-pool NAT (MP-NAT) on the cobas[®] 5800 system in routine blood donor screening at a hospital based blood centre.

Methods: A total of 7592 routine blood donor samples were screened and 7549 seronegative samples were subjected to MP-NAT using the cobas[®] 5800 system. HBV Profile and viral load testing were done for the NAT yield samples. HVB reactive samples with very low sample/cut-off ratio were also subjected to minipool testing followed by Hepatitis B profile testing and HBV viral load analysis. The results of these tests were interpreted individually and correlated further.

Results: 3 HBV NAT yields were detected. Subsequent HBV profile revealed that two of these cases exhibited evidence of past hepatitis infection with current infectious potential, characterized by Anti-HBc Total positivity, negative Anti-HBc IgM, and positive Anti-HBe antibodies. Viral load testing further confirmed the presence of HBV in these samples, with viral loads found as low as less than 20 IU/ml. Additionally, MP-NAT successfully detected samples with very low serology signal-to-cutoff (S/Co) ratios, with values as low as 1.2, and corresponding viral loads of less than 20 IU/ml.

Summary / Conclusions: These findings underscore the very good sensitivity of MP-NAT on the cobas 5800 system in identifying HBV infections, including donors with very low viral load, thus emphasizing its critical role in transfusion safety. Further in depth analysis of such NAT yield cases and cases with very low reactivity on serology platform will aid in understanding the actual sensitivity of minipool NAT Testing

P390 | Evaluation of a novel HBsAg assay for blood donor screening in the Philippines

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Background: Hepatitis B virus (HBV) remains a significant public health concern, particularly in the context of Transfusion Transmissible Infections (TTI). Accurate screening of blood donors for Hepatitis B surface antigen (HBsAg) is crucial to maintaining high standards in blood safety. The emergence of greyzone testing has heightened the need for robust assays capable of precisely quantifying HBsAg levels. In this study, we aimed to evaluate the performance of the Sysmex HBsAg assay against the current predicate in the Philippines, utilizing confirmatory datasets sent to the Research Institute for Tropical Medicine (RITM).

Aims: This study aims to assess the performance of the novel HBsAg assay in comparison to the current standard, focusing on its quantitative capabilities in the greyzone testing range. The objective is to determine its sensitivity for HBV screening among blood donors.

Methods: We analyzed 60 samples at RITM, with a gender ratio of 52 males and 8 females, median age 33. The novel HBsAg assay results were compared with the current standard, considering the assay's retest recommendations for samples falling within the greyzone.

Results: The assay demonstrated results between 3.08 to over 2500 IU/mL, while the current standard showed a range of 188 to 7502 S/CO. Notably, the novel assay covered a wider dynamic range against the predicate, including samples above 100 IU/mL. With 100% overall concordance, all cases were positively identified using both serology assays. Only two samples met retest criteria (0.03 to 5 IU/mL), highlighting the assay's high sensitivity for blood donor screening.

Summary / Conclusions: The novel HBsAg assay exhibits excellent performance compared to the current predicate offered in RITM, offering accurate results in the greyzone. Achieving 100% overall concordance and minimal retests underscores its high sensitivity for HBV screening in blood donors. These findings support the integration of the novel assay into the TTI screening process in the Philippines.

P391 | An effective method for separation of VERO cells from flask bottom for use in flow cytometry and fluorescent microscopy in herpes virus identification as hepatitis B model

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Background: To assess the effectiveness of the inactivation method on the model virus and cell culture, the results of the experiment are based on changes in cell morphology, which can sometimes fail to distinguish between the natural cell damage and caused by the virus's cytopathic effect. Therefore, the use of methods demonstrating virus replication in cell culture can be very helpful in distinguishing between viral replication with and without cell damage and natural cell destruction. Therefore, in this study, we used the hepatitis B model virus, i.e., the HSV virus, and compare the use of nanoparticle methods with inverted microscopy methods to investigate viral infections to provide a more sensitive and accurate method that is also cost-effective. One of the key points is effective separation with cell separating agents from the surface that may increase sensitivity in identification. Due to the adhesive nature of the cell line used (VERO) and to prevent the cells to re-adhere during the flow cytometry and fixation process, Trypsin was not used. Therefore, a method for preparing cells for flow cytometry and preparing indirect immunofluorescence slides was designed. In this method, EDTA with a specific concentration and pH was used to detach the cells from the bottom of the cell culture plate or flask.

Aims: Our goal is to prepare cells for flow cytometry and to prepare indirect immunofluorescence slides using EDTA with a specific concentration and pH to dissociate and detach cells from the bottom of the cell culture plate or flask with higher efficiency compared to the conventional method of using trypsin.

Methods: (1). Initially, an EDTA solution of 15 mM was prepared using the mentioned method. Then, 1/5 cc of this solution was added to each 25 cm² flask of 90% confluency of VERO cells. (2). The flasks were placed on a specific shaker at 400 revolutions per minute (rpm) for 1.5 min. (3). They were transferred to a 37°C incubator and left for 20 min. (4). The flasks were removed from the incubator, and to ensure complete dissociation of the cells from the bottom of the flask, they were gently tapped around the flask. After checking the cells under a microscope, it was ensured that the cells were completely dissociated and detached. (5). The trypsin method was performed in the usual manner.

Results: The cell dissociation status after treatment with 15 mM EDTA and trypsin is as follows respectively: Immediately after treatment: 70% and 100%. Dissociated after one hour: 100% and 70%. Dissociated after two hours: 100% and 60%. Dissociated after three hours: over 95% and 50%. Quantum dots and FITC, used as conjugates in both indirect immunofluorescence and flow cytometry, exhibited appropriate characteristics and sensitivity, allowing for the detection of VERO cells infected with the virus at high levels, with minimum detectability of 1 infectious particle per milliliter for Quantum dots and 10 infectious particles per milliliter for FITC.

Summary / Conclusions: The optimized separation method using 15 mM EDTA with higher efficiency compared to the conventional trypsin separation method can be utilized for flow cytometry and indirect immunofluorescence assays.

P392 | Prevalence of occult hepatitis B infection among blood donors in Thailand

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Background: Early window period (WP) infection and occult HBV infection (OBI), characterized by the presence of HBV DNA in plasma with undetectable HBsAg, is a potential risk of HBV transmission through blood transfusion. OBI prevalence varies between different geographic areas as the population can be infected with different HBV genotypes.

Aims: The purpose of this study is to evaluate the prevalence of OBI among Thai blood donors and the characteristic of OBI by serological methods.

Methods: Six thousand and seventy blood donors were screened for HBsAg, Anti-HBc, Anti-HBs by Alinity i (Abbott Laboratories)

and nucleic acid test (NAT) by Cobas 6800 system (Roche Diagnostics). Samples with initially reactive results were repeated in duplicate.

Results: The prevalence of occult hepatitis B infection (OBI) was 0.18%. Seropositive OBI accounted for 0.15% while seronegative OBI was 0.03%. There were 0.26% of donors who had a positive result for both nucleic acid test for HBV and HBsAg. Anti-HBc only positive was observed in 15.22% of blood donors. Most NAT yield cases of hepatitis B infection were presented as "anti-HBc" only. In our study, NAT yield with "anti-HBc" only was about 0.15%, compared with 0.02% in NAT yield with positive anti-HBs which is because of its relatively short life span. The prevalence of positive anti-HBc in donors was 15.22%, thus anti-HBc should not be recommended as a surrogate test in Thai blood donors.

Summary / Conclusions: The prevalence of OBI in Thai blood donors was 0.18%. Anti-HBc only positive was 15.22% and should not be used as a surrogate test for blood donors in Thailand.

P393 | Results of screening antibodies to the nuclear antigen anti-HBc among donors in the Karaganda region

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Background: Mandatory screening of donors for the anti-HBc marker in Kazakhstan was introduced after amendments were made to the order of the Ministry of Health of the Republic of Kazakhstan dated August 2. Screening for antibodies to detect the nuclear antigen of the hepatitis B virus (anti-HBc) in the Karaganda region has been carried out since October 4, 2022.

Aims: Evaluate the results of screening for antibodies to detect the nuclear antigen anti-HB-core among donors in the Karaganda region from October 4, 2022, to December 2023.

Methods: The study included 19,056 donors from 18 to 71 years old. All samples were examined for the presence of HIV-1/2 antibodies, HBsAg, antibodies for HCV, syphilis and additionally for the presence of antibodies for the nuclear antigen anti-HBc. The testing was done with quantitative determination of antibodies to the surface antigen anti-HBs in samples that had an a-HBc reactive result using chemiluminescence immunoassay method on an Alynity, Vitros analyzer. If the anti-HBs values of a-HBc reactive donors were in the range of 10-100 mIU/ml, then all prepared products were rejected, and the donors were excluded from donation indefinitely. Negative blood samples for decreed markers of 18,352 donors were tested for the presence of HIV, HCV and HBV nucleic acids by PCR in pools of six samples using Cobas Ampliprep and Cobas Taqman. Data for statistical analysis was obtained from the Infodonor IS. A comparative statistical analysis was carried out using a spreadsheet editor MS Excel.

Results: Over a 15-month study of screening for antibodies for the HBV nuclear antigen (anti-HBc), with 19,056 participants, was

conducted. Where 14,421 (75.7%) were men and 4,637 (24.3%) were women. Anti-HBcore was detected in 1930 out of 19056 samples (10.1%), with acceptable anti-HBs values of <10 IU/l or > 100 IU/l - 1539 (79.7%) and unacceptable for anti-HBs, with the values were in the range of 10-99.9 IU/l - 391 (20.3%). 186 out of 19056 primary reactive samples containing HBsAg were identified, in which 47 (25.2%) were confirmed. The frequency of detection of anti-HBc depended on the age of the donor: the lowest detection rate was observed among donors born in 2001-2005 and amounted to 0.6%, while the highest among donors born in 1950-1960 and amounted to 32%. The PCR method tested 18,352 samples, a reactive reaction was detected in 7 pools, in each pool there was 1 sample with antibodies for the HBV nuclear antigen (anti-HBc), but all indicators of surface anti-HBs were within acceptable limits. In 4 samples HBV DNA was detected, while the indicators were for anti-HBs <10 IU/l, in 3 samples HBV DNA was not determined. One sample depicted an indicator for anti-HBs <10 IU/l (3.50), the remaining two for anti-HBs > 100 IU/l. Thus, to ensure the infectious safety of donor blood and its components, all prepared components from these 3 samples were rejected and disposed, donors were excluded from donation indefinitely.

Summary / Conclusions: Significant limitations of our study are that virus concentrations were not measured, and vaccination records were not included in our research. In Kazakhstan, today the WHO recommendations for the elimination of hepatitis B have been implemented in terms of ensuring safety measures for the collection of blood and its components. However, a problem with testing donors using existing methods and their further treatment from hepatitis B is yet to be addressed in local state hospitals in Kazakhstan.

P394 | Abstract withdrawn

Transfusion transmitted infections—Hepatitis C (HCV)

P395 | Abstract withdrawn

P396 | Analysis of anti-HCV immunoassay sample to cutoff ratios in blood donors with positive results to aid recipient lookback risk assessments

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Background: High sample to cutoff (s/co) ratios on the Abbott PRISM chemiluminescent immunoassays have been shown to be reasonably predictive of confirmed positive results (Kiely, Transfusion, 2010). Although s/co ratios cannot be used for interpretation, they can be used as a communication aid for blood donors with positive results,

especially in low prevalence populations with a low positive predictive value and for risk assessment in recipient lookback decisions. On 26 October 2020, Australian Red Cross Lifeblood replaced the PRISM analyser with the Alinity s analyser, for the serological screening of blood donors for anti-HCV, anti-HIV, anti-HTLV and HBsAg. The change in testing analysers meant the s/co ratios, specifically for the anti-HCV assay, required re-analysis.

Aims: An analysis of Alinity s Anti-HCV assay s/co ratio distributions was performed to determine whether high s/co ratios are predictive of confirmed infection and lower s/co ratios more likely to be false positive and if Alinity s/co ratios can be used to aid recipient lookback risk assessments.

Methods: The s/co ratios for anti-HCV repeatedly reactive samples collected between 25 October 2020 and 29 July 2021, and between 1 January 2022 to 31 December 2022 were analysed with the final donor status assigned at the completion of HCV testing. The s/co ratios for confirmed infection anti-HCV positive donors (i.e. had a previous reported RNA positive result) were plotted against their years of recovery or clearance, to give an indication of s/co signal strength. The s/co ratios for repeat donors who were anti-HCV equivocal (reactive on two immunoassays, immunoblot negative, HCV RNA non-reactive) and anti-HCV indeterminate (reactive on two immunoassays, immunoblot indeterminate, HCV RNA non-reactive) were then plotted on the same graph with their inter-donation interval (interval between their prior anti-HCV negative result and their positive result) to determine if their s/co ratios correlated to the confirmed infection anti-HCV positive donors, suggestive of seroconversion with cleared HCV RNA.

Results: The s/co ratios from 1111 donations repeatedly reactive for anti-HCV were analysed. Of the 87 anti-HCV positive donors, there were 33 instances where the date of infection clearance was recorded. Correlation was used to determine the relationship between the s/co and years since recovery or clearance. There was a strong negative correlation between these two values ($r = -0.69$, $n = 33$, $p < 0.0001$), indicating recency of confirmed infection had a strong correlation with higher s/co ratios. When the s/co ratios for the anti-HCV equivocal ($n = 26$) and anti-HCV indeterminate ($n = 12$) donors and their inter-donation interval were plotted on the same graph, the s/co ratios were significantly lower and not consistent with acute infection. In addition, there was no correlation pattern.

Summary / Conclusions: The analysis showed blood donors with anti-HCV equivocal and anti-HCV indeterminate results who had s/co less than 4.0 and no definitive risk factors with shorter inter-donation intervals, were unlikely to represent acute infection and more consistent with false positivity. Alinity s/co ratios can be used to assess potential false positivity in recipient lookback risk assessments.

P397 | Analysis of HCV confirmatory test results in blood donors

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Background: Hepatitis C is a disease widely distributed throughout the world, it is estimated that 3% of the world's population is infected. In Brazil the prevalence varies from 1% to 2%, depending on the region. Testing blood donations for HCV antibodies and nucleic acid testing (NAT) has been the standard practice in Brazil.

Aims: To analyze the results of confirmatory tests for HCV in donors from a Hemotherapy Service in Southern Brazil.

Methods: Donations with reactive or indeterminate anti-HCV results were analyzed from January to December 2019 at the Hemotherapy Service of Hospital São Vicente de Paulo, Passo Fundo/RS. HCV screening was performed by the chemiluminescence method, using the Architect Anti-HCV ABBOTT kit, on the Architect i1000 and i2000 platforms, as well as the NAT-HCV Biomanguinhos. All donors with a reactive or indeterminate result were summoned by letter to collect a new sample, which was again tested by chemiluminescence. The samples that remained reactive or indeterminate were sent to an external laboratory for the 3rd generation HCV Immunoblot (RIBA) test.

Results: Of the 13537 donations during the study period, 107 (0.8%) had reactive or indeterminate anti-HCV results, of which 104 (97.2%) had non-detectable NAT (ND) and 3 (2.8%) had detectable NAT (D). The 107 donors were summoned for a new sample collection, 67 (62.6%) returned and of these, 56 had a reactive or indeterminate anti-HCV result, 1 with detectable NAT and reactive Immunoblot, 4 with non-detectable NAT and reactive Immunoblot and 51 with non-detectable NAT and non-reactive Immunoblot, as well as 11 donors who had a non-reactive anti-HCV result in the second sample with non-detectable NAT.

Summary / Conclusions: Regarding the classification of the donors' status, only 1 (1.4%) was positive in the three tests carried out in this study, while 76% of the donors were positive only in the anti-HCV test by chemiluminescence. This data is alarming, given that these donations were discarded due to a false-positive result, in addition to the need to call in these donors, provide counseling, as well as to perform confirmatory tests, contributing to an increase in the cost of blood therapy processes and causing anxiety in donors. Lucey et al., 2022, recommended some measures to optimize pre-analytical processes that can reduce false-positive results, including ultracentrifugation, as well as observing the presence of hemolysis in the samples. With the aim of improving the cost-effectiveness of HCV testing in

P397 - Table 1. Results of supplementary tests in 67 blood donors who returned to the blood center for follow-up

anti-HCV neg in follow-up sample	Active infection (IB +/NAT+)	Resolved infection (IB +/NAT-)	False positives (IB-/NAT-)
11	1	4 (6.0%)	51 (76.1%)

donors, Cheng et. al., 2023, concluded that removing the anti-HCV test and maintaining only the NAT test in donor screening contributed to reducing costs without compromising transfusion safety in Australia. The anti-HCV test by chemiluminescence showed high rates of false-positive results in our study, increasing the rate of blood component disposal and contributing to increased costs in Hemotherapy Services. We highlight the importance of critically analyzing donor testing strategies and looking for ways to reduce false-positive rates, thus avoiding the disposal of blood components.

P398 | Serologic pattern of screening assay for antibody to hepatitis C virus in Hong Kong blood donors

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Background: Notwithstanding the excellent performance of the current immunoassays for anti-HCV screening in healthy blood donors in terms of analytical sensitivity and specificity, positive predictive value remains low. Therefore, a sequential +/- confirmatory testing strategy is adopted in attempt to rule in or rule out the reactive primary screening immunoassay findings.

Aims: To characterize the serologic pattern of anti-HCV repeatedly reactive samples subsequent to primary screening immunoassay (IA1).

Methods: All donations were screened simultaneously for anti-HCV and HCV RNA by Alinity s anti-HCV (Jan 2020 - Aug 2021) or anti-HCV II (Aug 2021 - Dec 2023) assays (IA1) (Abbott, Germany) and Procleix Ultrio Elite assay on Panther system (RNA) (Grifols, USA) respectively. IA1 repeatedly reactive/RNA negative samples were further tested by anti-HCV supplementary immunoassay (IA2) using Murex Anti-HCV version 4.0 (DiaSorin, South Africa). IA1/IA2 reactive RNA negative samples as well as IA1/RNA reactive samples were subject to confirmatory test by a strip immunoassay (IB) using INNO-LIA HCV Score (Fujirebio, Belgium).

Results: From January 2020 to December 2023, 830,346 donations (340,728 donations by Alinity s anti-HCV assay; 489,618 by anti-HCV II assay) were tested; 462 (0.056%) were repeatedly reactive for anti-HCV by IA1; of which, 58 (12.6%) were RNA reactive and 404 (87.5%) RNA negative, which were then tested by IA2; of which 342 (84.7%) were IA2 negative and 62 (15.4%) IA2 reactive. Therefore, these 62 IA1/IA2 reactive/RNA negative and 58 IA1/RNA reactive samples were tested by IB. Of the 62 IA1/IA2 reactive/RNA negative samples, 28 (45.2%) were IB positive, 27 (43.5%) indeterminate and 7 (11.3%) negative. All (100%) of the 58 IA1/RNA reactive samples were IB positive. Both IB reactive groups (i.e. IA1/RNA/IB reactive and IA1/IA2/IB reactive RNA negative) had the highest Alinity s anti-HCV/anti-HCV II S/CO ratio (median: 20.96 and 27.46 respectively, $p = 0.81$). Their S/CO ratios were statistically higher than the other groups: IA1/IA2 reactive IB indeterminate group (median: 2.65), IA1/IA2 reactive IB negative group (median: 1.47) and IA1 reactive IA2/RNA negative group (median: 1.79) ($p < 0.001$). However, S/CO ratios were not different between the latter 2 groups ($p = 0.28$). The frequency of

confirmed positive HCV donations using Alinity s anti-HCV and anti-HCV II assays during the 2 study periods were 0.059% and 0.054% respectively ($p = 0.32$). In general, anti-HCV II gave higher S/CO ratios in all the sub-groups mentioned above ($p < 0.001$). Nil cases of IA1/IA2 reactive IB negative were observed by anti-HCV II assay whereas there were 7 cases with anti-HCV assay. Subsequent to the IB test algorithm, 86 donations were confirmed anti-HCV positive, giving a prevalence of 0.01%; 67.4% were HCV RNA reactive and 32.6% negative.

Summary / Conclusions: The findings of this study revealed that Alinity s anti-HCV/anti-HCV II assay S/CO ratios were not useful in differentiating between biological false reactive samples (IA1 reactive/IA2 negative) and anti-HCV confirmed positive cases (IA1/IA2/IB reactive) due to overlapping ranges of S/CO values. The IB negative and indeterminate groups were more heterogeneous in nature, in that they could represent non-specific reactivity or progressing seroreversion, etc. Again, S/CO ratios alone were not helpful to predict the underlying cause of the reactive IA1 results without clinical follow-up of the donors.

P399 | Evaluating the efficacy of HCV rapid screening kits in resource-constrained environments

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Background: The prevalence of Hepatitis C Virus has impacted the pool of available blood donors. In an era where blood supply is already limited, the evaluation of HCV kits poses a significant challenge for medical laboratory scientists as they strive to meet the heightened demand for safe blood.

Aims: The objective of this investigation was to assess the effectiveness of HCV testing kits.

Methods: The research took place at the Makhdoom Diagnostic Centre and Blood Bank in Mandi Bahaulddin. A sample of 500 blood donor specimens was analyzed, comprising 384 positive and 116 negative samples for Anti-HCV. Three rapid screening devices and one ELISA were assessed against Polymerase Chain Reaction (PCR) as the reference standard for HCV detection. Specificity, sensitivity, positive predictive value (PPV), negative predictive value (NPV), Positive Likelihood Ratio (PLR), and Negative Likelihood Ratio (NLR) were calculated using PCR results as the benchmark.

Results: A total of 210 blood donors participated in the study, and blood was collected from each participant and processed accordingly. The majority of participants were male, comprising 97.4% ($n = 487$), while females constituted only 2.6% ($n = 13$). The mean age of the participants was 37 years. HCV PCR served as the gold standard and

was performed on the Cobas 6800 platform. Three ICT kits (ABBOT, Abon, and SD Bioline) and one ELISA kit (Cenix) were assessed for Anti-HCV. The ELISA kit (Cenix) demonstrated a sensitivity of 99.2%, while the ABBOT ICT kit exhibited a sensitivity of 98.7%, followed by the Abon ICT kit (98.2%) and the SD Bioline ICT kit (97.7%). The specificity of the ELISA kit (Cenix) was determined to be 95.7%, while the ABBOT ICT kit showed a specificity of 98.3%, followed by the SD Bioline ICT kit (98.3%) and the Abon ICT kit (96.6%). The positive predictive value (PPV) for the ELISA kit (Cenix) was 98.7%, and for the ABBOT and SD Bioline ICT kits, it was 99.5%, followed by the Abon ICT kit (99.0%). The negative predictive value for the ELISA kit (Cenix) was 97.4%, for the ABBOT ICT kit it was 95.8%, followed by the Abon ICT kit (94.1%) and the SD Bioline ICT kit (92.7%).

Summary / Conclusions: In our study, the sensitivity and specificity of the ELISA kit (Cenix) were acceptable as of the gold standard PCR for Anti-HCV detection. Among the ICT kits, the Abbot devices demonstrate the highest sensitivity and specificity, surpassing other options. Therefore, considering the effectiveness of these kits, particularly in resource-limited countries, they can be reliably employed for disease screening in blood banks.

P400 | Hepatitis C virus infection testing of blood donors from South-Eastern region of Serbia

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Background: Hepatitis C virus (HCV) can cause a serious mostly asymptomatic chronic liver disease that may progress to liver cirrhosis and hepatocellular carcinoma. In order to prevent transmission of hepatitis C infection via blood transfusion anti HCV antibody screening of blood donors is mandatory in Blood transfusion institute of Nis since 1994. Nucleic acid testing (HCV NAT) is introduced as mandatory procedure in our country since 2019 as a part of blood transfusion service reorganization in Republic of Serbia.

Aims: Analysis of serological and molecular test results in Hepatitis C virus infection detection in blood donors of South-Eastern region of Serbia in period 01.01.2020 – 31.12.2023.

Methods: EDTA-anticoagulated blood donor samples were routinely screened for presence of anti HCV antibodies using different assays: chemiluminescent microparticle immunoassay (CMIA) Alinity s anti HCV Abbott, chemiluminescent immunoassay Advia Centaur anti HCV Siemens or enzyme-linked immunosorbent assay (ELISA) Monolisa HCV Ag/Ab Ultra V2 Biorad. All blood samples were also screened by NAT technology (Cobas MPX). Following confirmatory testing procedure, repeated reactive blood donor samples for presence of anti HCV antibodies were tested using Inno-lia HCVscore test, Fujirebio.

Results: During study period a total of 157,311 blood units were screened for presence of anti HCV antibodies, 117 blood donor samples (0.074%) were found repeat reactive (RR) for presence of anti HCV antibodies (36 samples were from first time donors). Eighteen

samples (0.011%) were confirmed positive, 9 samples (0.005%) were indeterminate and 90 samples (0.057%) had negative result after confirmatory testing procedure. Twelve confirmed positive samples have been also found positive for presence of HCV RNA.

Summary / Conclusions: Analysis of our test results shows that prevalence and incidence of Hepatitis C virus infection in blood donor population of South-Eastern Serbia is low. Therefore confirmatory testing is necessary to distinguish true and false positive screening test results and suppress loss of eligible blood donors due false reactive anti HCV antibody screening results. At the same time introduction of mandatory HCV RNA testing is important step for increasing blood transfusion safety in our country.

P401 | Significance of anti-HCV S/CO values in positive qualitative range

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Background: The WHO EMRO estimated that a total of 22 million persons are infected with HCV. According to WHO, the prevalence of HCV is estimated to be less than 0.5% in the UAE. More than 149 nationalities are donating blood in Dubai Blood donation center which reveals unexpected reactivity to screening tests. In order to ensure blood safety and prevent transfusion transmitted infection including Hepatitis C infection, serological and molecular screening for all blood donations are mandatory in our blood bank. The use of s/co ratios may minimize the amount of supplemental testing that needs to be performed while improving the reliability of reported test results.

Aims: Abbott Alinity s Anti-HCV II assay was validated in our blood center and approved as a test of record in February 2022. To investigate the prevalence of HCV infection in blood donors at Dubai blood donation center. To evaluate the utility of anti-HCV signal-to-cutoff ratios (S/CO ratio) as a predictor of active infection. To identify potential HCV parallel testing algorithms.

Methods: Between 1st of February 2022 to 31st December 2023 a total of 117,452 Donors from 159 different nationalities were tested by Abbott Alinity s Anti-HCV II in parallel with NAT multiplex assay using Roche Cobas 6800 system. For the purpose of this study all repeatedly reactive samples by Alinity Anti-HCV II were tested by Inno-Lia HCV Score form Innogenetics. All donor demographic data (age, gender, nationalities) were collected from Edelphyn Blood bank software system, and correlated with laboratory test & reactivity pattern results.

Results: The prevalence of Anti-HCV in our center is 0.11%. The HCV prevalence was significantly higher in male donors regardless of the HCV infection status. The prevalence was significantly higher in Egyptian and Pakistani donors (0.89%) but lower than prevalence reported by WHO in their home countries (2.49%, 2.33% respectively). The Alinity s Anti-HCV II assay showed 100% sensitivity and 99.94 specificity. The Alinity s Anti-HCV II S/CO ratio distribution of HCV active infection group (0.04%) was significantly higher than the HCV

indeterminant group (0.03%) and Biological false reactive group (0.06%) but significantly lower than the anti HCV positive group (0.07%). Few confirmed positive samples gave low values while some biologically false positive samples (BFP) gave high values. Out of the 36 anti-HCV indeterminate results, 23 (63.89%) were due to isolated reactivity to the NS3 band followed by 14 (38.89%) to capsid C1. The bands reactivity intensity between +/- and 1+. For reactivity pattern of 43 active HCV infection results showed 43 (100%) reactivity to NS3 followed by 41 (95.35%) for C1, C2 and NS4 while E2 reaction was observed for 32 (74.42%) of the samples. 78 chronic/resolved HCV infection results showed the highest reactivity to NS4 bands 50 (64.10%) followed by C1 35 (44.8%), E2 34 (43.59%) and NS3 31 (39%)

Summary / Conclusions: The prevalence of Anti-HCV in our donor population is low (0.11%), this reflects the robust screening strategy for donor selection in our center, but also the national medical fitness screening program which includes HCV testing for expats for certain job categories. S/cut-off ratios are predictive of confirmed HCV results and rarely not correlated to the HCV infection status. Implementation of second HCV immunoblot assay could help for immunoblot indeterminate cases for a better counselling of the impacted blood donors. Alinity s Anti-HCV II demonstrate excellent sensitivity and specificity performance.

Transfusion transmitted infections—HIV

P402 | Abstract withdrawn

P403 | A survey on the use of antiviral drugs among blood donors in Shenzhen from 2019 to 2023

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Background: The introduction of the first AIDS pre-exposure prophylaxis (PrEP) drug in China in December 2020 has raised concerns about the potential escalation of HIV transfusion-transmission risk associated with the use of antiretroviral therapy (ART) in the country. The current utilization of ART drugs by blood donors in China is not well understood.

Aims: This study aims to assess the potential risks of antiretroviral therapy drugs on blood safety among blood donors in Shenzhen, China. Additionally, the study seeks to investigate the development of an efficient and cost-effective method for detecting ART drugs.

Methods: The study utilized high pressure liquid chromatography coupled with tandem mass spectrometry (HPLC-MS/MS) to quantify the concentrations of 8 antiretroviral therapy (ART) drugs in five distinct groups comprising approximately 440,000 blood donors over the period of 2019–2023. These groups included individuals in group A (n = 3) who were regular consumers of known ART drugs, group B (n = 86) consisting of repeat blood donors, group C (n = 73)

comprising anti-HIV positive/anti-TP negative blood donors, group D (n = 358) consisting of anti-TP positive/anti-HIV negative blood donors, and group E (n = 25) comprising anti-HIV positive/anti-TP positive blood donors.

Results: The results showed that baseline concentrations of ART drugs were not detected in 86 samples of group B. Positive antiviral drug samples were found in 3 out of 73 samples in group C (4.1%), 0 out of 358 samples in group D, and 1 out of 25 samples in group E (4.0%) in the resolution test using 1:2 pooled plasma samples. The ART drugs remained detectable even after dilution at 1:6. Tenofovir, lamivudine, and efavirenz were detected in 3 blood donors, while lamivudine, lopinavir, ritonavir, and zidovudine were detected in 1 blood donor. In conclusion, ART drugs were found in blood samples testing positive for anti-HIV and anti-TP antibodies.

Summary / Conclusions: The presence of antiretroviral therapy (ART) drugs was identified in blood donors in Shenzhen who tested positive for both HIV and *Treponema pallidum* (TP). The population at high risk for HIV infection prevention and treatment faces challenges in accessing antiretroviral therapy (ART) medications. Pooled testing offers a more cost-effective and efficient method for investigating medication usage within a large sample population. Keywords: ART drugs, HIV, TP, detection rate, pooled testing.

P404 | Biological qualification of blood unit at Brescia Blood Bank (BBB) - description of three cases of HIV seroconversion

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Background: The biological qualification of blood products make use of the latest technologies in infectious serology and molecular biology, as well as standardized and strictly regulated operational and decision-making algorithms. Thanks to high levels of sensitivity and reliability of analytical methods, we are able to intercept any positivity to virological markers tested on blood donors. The following describes three cases, detected in 2023 at BBB who tested HIV- positive.

Aims: The aim of BBB is to ensure transfusion safety patient and monitor the health of the periodic donor.

Methods: Tests performed at BBB: serology screening: Abbott Alinity HIV Ab/Ag combo (CMIA); NAT screening: Roche Cobas MPX (Real Time-PCR). Confirmatory tests performed by the Reference Laboratory (RL): quantitative HIV Ab/Ag (QT): Diasorin Liaison XL Murex HIV Ab/Ag (CMIA); serology confirmation: Biorad, NEW LAV-BLOT HIV (immunoblotting); HIV-RNA QT: Roche Cobas HIV quantitative Nucleic Acid Test (Real Time-PCR and molecular hybridization).

Results: Three HIV positives were detected on periodic donors with screening tests, confirmed by RL. **Don. 1** female 62 years old, **Don. 2** female 45 years old, **Don. 3** male 48 years old. At the time of donation, the three subjects, having failed to declare risk factors, were admitted

to donation. **Don. 1:** 1st sample on 11/17: NEG screening serology, NAT POS; 2nd sample on 11/21: NEG serology, NAT POS and HIV-RNA QT POS (31600 cp/ml); 3rd sample on 11/24: POS screening serology, HIV Ab/Ag QT POS, NEG immunoblotting, p24/Ag POS, NAT POS, HIV-RNA QT POS (1.7×10^6 cp/ml). **Don. 2:** 1st sample of 11/28: POS screening for both serology and NAT; 2nd sample of 11/28: NAT POS, HIV-RNA QT POS (9900 cp/ml), immunoblotting POS and p24/Ag NEG. **Don. 3:** Sample of 11/13: POS (RR) for both serology and NAT; 2nd sample of 11/15: NAT POS, HIV-RNA QT POS (21600 cp/ml), immunoblotting POS and p24/Ag NEG.

Summary / Conclusions: It is interesting to note the similarity between the cases **Don. 2** and **Don. 3:** the positivity of all screening and confirmatory serologic and molecular parameters, as well as the absence of the early HIV-p24/Ag marker attest to seroconversion. For **Don. 1**, we note, that HIV screening serology on the first two samples was negative, while NAT was positive, confirmed by HIV-RNA QT test; this means that the infection was still in the early asymptomatic stage, before seroconversion. Serological tests on the sample, 7 days after the first finding (3rd sample), was positive, also NAT confirmed. However, the infection was still at an early stage, demonstrated by HIV-p24/Ag positivity, together with immunoblotting negativity and a sharp increase in viremia. These cases of HIV positivity, highlight the high reliability and efficiency of the analytical methods and procedures present in our BBB, designed to preside over the quality and safety of blood products collected for transfusion therapies. In particular, the case of **Don 1** demonstrates the system's ability to intercept the onset of new positivities in the periodic donor population, even in the very early stages of infection.

P405 | HIV infections among the blood donors in the Regional Blood Center in Poznan, Poland in years 2018-2022

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Background: Testing donors for the infection with HIV is compulsory and is included in the hemovigilance procedures that aim to minimize the risk of occurrence of serious adverse reactions in donors, serious post transfusion adverse reactions and also function as way of epidemiological monitoring of donors. Hemovigilance on the part of donors also includes epidemiological data for donors with confirmed positive results of STDs, including HIV.

Aims: The aim is to analyze the number of confirmed HIV infections among the blood donors in the area of activity of Regional Blood Donor Center in Poznan, Poland in years 2018-22

Methods: The analysis was made using data from the computer system which is in operation in Regional Blood Donor Center in Poznań, Poland and epidemiological data regarding HIV infections.

Results: In recent years we have observed rising number of HIV infection. In 2018 in Poland there were 1275 cases of new infections, in 2019 – 1615, in 2020 – 840, in 2021 – 1173, and in 2022 – 2384. In the Greater Poland Province (the area of activity of Regional Blood

Donor Center in Poznan) Poland in 2018 there were 152 cases, in 2019 – 131, in 2020 – 76, in 2021 – 131, in 2022 – 213. In accordance with current recommendations screening tests (serological method) for the presence of the HIV markers as well as NAT testing are performed for every donation for first-time donors (FTD) and repeat donors (RD). Screening tests and NAT testing are performed to detect the status of the donor. In case of the positive result of one or both tests, the status is described as reactive and further confirmatory testing in the Institute of Hematology and Transfusiology in Warsaw is performed. The analysis of the data for years 2018-2022 has shown single HIV infections in the group of donors: in 2018 out of 100,331 donations 2 donors were infected (1 FTD, 1 RD, in 2019 out of 102 373 donations – 3 donors (1 FTD, 2 RD), in 2020 out of 90 406 donations – 0 donors; in 2021 out of 102 995 – 4 donors (1 FTD, 3 RD) and in 2022 out of 106 561 donations – 5 donors (4 FTD, 1 RD). In total there were 14 donors (7 FTD, 7 RD) in years 2018-2022).

Summary / Conclusions: The analysis has shown the occurrence of HIV infected people in the group of blood donors. In reference to the number of tested samples the result is comparable to the rate in general population. The analysis has proved the increase in the number of HIV infections among the donors and the similarity with the general trend in the country. (1). Testing donors for the HIV infection increases the safety of blood recipients and as a compulsory measure in blood establishments it is fully justified in the Polish population. (2). In the area of activity of Regional Blood Donor Center in Poznań we have not observed so far the so called test seekers i.e. donors registering for the blood donation with the intention just to get tested. (3). We have observed no correlation in the group of donors with confirmed positive test results for HIV regarding being first-time or repeat-donor. (4). An upward trend regarding the number of HIV infections in the group of donors can be observed despite many years of wide public educational activities regarding the risk of virus transmission by means of transfusion.

P406 | A novel approach for detecting breakthrough infections following Pre-Exposure Prophylaxis (PrEP) administration.

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Background: Pre-exposure prophylaxis (PrEP) and post-exposure prophylaxis (PEP) are utilized as preventive measures against HIV infection; however, instances of drug failure resulting in breakthrough infections may still occur. Timely detection and confirmation of HIV infection, followed by prompt initiation of treatment, are crucial steps in managing the condition effectively.

Aims: The objective of this study is to establish a magnetic bead enrichment strategy for the detection of human immunodeficiency virus deoxyribonucleic acid (HIV DNA) in peripheral blood, and to assess the sensitivity improvement of this method for detecting HIV

DNA in HIV-infected patients following early antiretroviral treatment (ART).

Methods: Whole blood was collected peripherally at four timepoints from a single patient in the acute retroviral syndrome (ART) HIV window period. Peripheral blood mononuclear cells (PBMCs) were isolated using a Ficoll gradient, and CD4⁺ T lymphocytes were enriched from the total PBMCs through negative selection. The concentration of HIV DNA in both the magnetic beads enriched group and the whole blood group was determined using an HIV DNA detection kit.

Results: CD4⁺ T cells were isolated by magnetic beads and identified by FCM for purity, their purity was (96.4 ± 2.6)%. The viability was (95.9 ± 2.9)%, as demonstrated by trypan blue staining. The person on continued ART treatment in this study have significantly greater reduction in HIV viral load and have undetectable HIV plasma RNA at follow up timepoint 4. No HIV DNA was detected in the whole blood group at all 4 timepoints. The quantitative results of HIV DNA in the CD4⁺ T lymphocyte group of the magnetic bead enrichment group were 73.4, 429.3, 137.1, 449.9 copies/10⁶ CD4⁺ T cells respectively.

Summary / Conclusions: The utilization of magnetic bead enrichment can enhance the sensitivity of detecting low copy HIV DNA in blood samples, thereby facilitating the early confirmation of HIV WP infection and breakthrough infection following antiretroviral therapy.

Transfusion transmitted infections—bacteria

P407 | Detection and molecular typing of *Morganella morganii*, a cause of transfusion-transmitted bacterial infections

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Background: The Japanese Red Cross Society (JRCS) is responsible for all blood services-related aspects in Japan, from collecting to delivering blood products. All platelet concentrates (PCs) are derived from apheresis donations and approximately 800,000 PC bags are supplied annually. JRCS has not implemented bacterial screening or pathogen reduction technologies; however, the incidence of transfusion-transmitted bacterial infection (TTBI) is low because of several preventive measures such as, initial blood flow diversion, pre-storage leukoreduction, short shelf life (3.5 days), and visual inspection. Only few TTBI cases caused by PC occur annually. In 2022, two PC bags from a single blood donation caused *Morganella morganii* (*M. morganii*)-related TTBI in two patients on the third day of blood collection, causing serious illness in one patient and death in the other. Visual abnormalities were not reported in the PC bags in either case. *M. morganii*, a Gram-negative, rod-shaped bacterium belonging to *Enterobacteriaceae*, is found in the intestinal tract. In Europe, a fatal TTBI caused by *M. morganii* was reported after a pooled PC transfusion.

Aims: JRCS currently plans to implement bacterial screening using culture methods. This study aimed to confirm *M. morganii* detection via the culture testing protocol to be implemented and verify methods for identification at strain levels using molecular typing tests.

Methods: From a TTBI case, we spiked 1 mL of bacteria suspension containing 10 or 100 CFU of *M. morganii* into day 1 PC bags (*N* = 10) and stored the bags at 20–24°C with agitation for 40 h; for detection, 8 mL PC samples were inoculated into aerobic and anaerobic culture bottles and cultured using BacT/ALERT VIRTUO (VIRTUO). During sampling, PC samples were spread on agar plates and cultured to count the bacterial colonies. *M. morganii* genotypes isolated from PC and patients were analyzed using pulsed-field gel electrophoresis (PFGE) and whole-genome multilocus sequence typing (wgMLST), comparing approximately 3600 gene sequences to confirm a causal relationship.

Results: After bottle culturing with VIRTUO at 36°C for 10 days, eight of the ten PC bags were culture-negative. On day 6 or 7 after bacterial spiking, PC samples were collected from the eight culture-negative PCs and again cultured with VIRTUO. All bottles tested culture-negative. In only two bags, *M. morganii* increased beyond 10E+5 CFU/mL 40 h post inoculation and tested positive within 4 h of incubation with VIRTUO. No aggregation was observed in bags with a normal appearance. PFGE patterns of *M. morganii* isolates from PC and the two patients were identical. Furthermore, analysis using wgMLST showed high homology (>99.99%).

Summary / Conclusions: The *M. morganii* strain used in this study was presumed sensitive to innate immune mechanisms such as the complement system, as it lost its proliferative ability in 8 of 10 bags during storage, proliferating in only two PC bags. These results suggest that some Gram-negative species contaminate PCs but are undetectable or do not cause TTBI because they do not proliferate. Our study verified that *M. morganii* could be detected via VIRTUO using the current screening protocol when grown in storage. Strain identities were confirmed via PFGE and wgMLST. We expect cultural screening to be effective in identifying PCs contaminated by bacteria that generally do not cause visual abnormalities, such as *M. morganii*.

P408 | Environmental bacterial screening in the blood center

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Background: Septic transfusion reactions (STRs) from bacterial contamination of platelets are a persistent cause of transfusion-associated mortality. Although the route of contamination often goes unidentified, bacterial contamination is frequently attributed to donor bacteremia or skin microbiota and is largely a consequence of room temperature storage. Consequently, mitigation strategies are aimed at the collection (e.g., donor temperature, screening questions, skin preparation), pathogen reduction and detection of bacterial growth by culturing and rapid tests. Despite implementation of the United States Food and Drug Administration (FDA) guidance on bacterial risk control strategies, 7 STRs occurred between 2018 and

P408 - Table 1: Environmental bacteria data before and after enhanced cleaning and procedural mitigations.

	Pre-cleaning Swabs CFU/cm ²	Post-cleaning Swabs CFU/cm ²	1 month Post-enhanced Cleaning CFU/cm ²
Site 1	14546	85	27
Site 2	282850	2148	<1
Site 3			3
Site 4			3

2021 from contaminated platelets units in the United States involving *Acinetobacter calcoaceticus-baumannii* complex alone or in combination with *Staphylococcus saprophyticus* and/or *Leclercia adecarboxylata* (Villa, Transfusion, 2023). An emerging source of concern is environmental bacteria, and the FDA has proposed that the source may be upstream of platelet manufacturing (Kracalik, Emerging Infectious Diseases, 2023).

Aims: Protocols for bacterial screening in the blood center environment are inconsistent between centers, and only 40% of centers conduct screening (Ramirez, Vox Sanguinis, 2023). To investigate the impact of environmental bacteria on STR occurrences, the largest Blood Center in the United States initiated prospective environmental bacterial screening.

Methods: Environmental bacterial screening was initiated in 2023. The screening was conducted before and after enhanced environmental cleaning and is conducted monthly at 4 blood manufacturing sites. The screening protocol captures both quantitative (CFU/cm² (log)) and qualitative (bacterial identification) data.

Results: Baseline bacterial testing was conducted at 2 of the 4 sites prior to initiating enhanced environmental cleaning procedures. Baseline testing included environmental swabbing before and after cleaning of selected areas. Pre-cleaning bacterial load was 14546 and 282850 CFU/cm² at the sites and decreased to 85 and 2148 CFU/cm², respectively, after cleaning the swabbed areas. The bacterial load was further reduced to 27 and < 1 CFU/cm² after updated cleaning protocols were implemented in the first month of screening. While cleaning proved effective for *Acinetobacter calcoaceticus-baumannii* complex alone or in combination with *Staphylococcus saprophyticus* and/or *Leclercia adecarboxylata*, *Bacillus spp* may persist in low quantities.

Summary / Conclusions: Environmental bacterial screening represents the continuous efforts to mitigate the bacterial risk in the blood supply and provides a method for evaluating mitigations implemented. Enhanced cleaning protocols clearly demonstrated a reduction in environmental bacteria in the blood center locations.

P409 | Increased incidence of syphilis among blood donors poses blood donation at risk in Japan

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Background: The number of syphilis cases has been increasing in recent years in Japan. In 2022, more than 10,000 cases were reported for the first time in the history of Japan. Although measures such as testing of all donated blood for syphilis, and the notification of positive blood donors are implemented in Japan, because of the persistence of antibodies in the plasma even after appropriate treatment, the increased incidence of syphilis in the population may importantly affect blood donor selection and the stable blood supply.

Aims: To analyze the incidence of syphilis among the Japanese blood donors and the characteristics of the infected individuals in an attempt to understand the risk posed to the blood collection and supply.

Methods: Blood samples from 39,199,047 blood donations in Japan in the period 1 January 2015 to 31 December 2022 were analyzed. After an initial screening for *Treponema pallidum* antibody (anti-TP), those positive were confirmed by the rapid plasma reagin (RPR). All RPR positive samples were eligible for the study. The confirmed syphilis positive blood donors were evaluated according to gender, age and donation frequency.

Results: In 2015, 321 samples (6.5/100,000 blood donations) were RPR positive. From 2016 to 2018, the incidence increased, reaching 9.7/100,000 (458 samples) in 2018. Although almost constant in the period 2018–2021, it further increased to 11.1/100,000 (553 samples) in 2022. No evident regional differences were observed, with RPR positivity increasing in all testing facilities in Japan. Whereas the confirmed syphilis cases increased 4.8 times in the general population, the increase among blood donors was less significant (1.7 times in the same period). Interestingly, during the COVID-19 pandemic (2020), whereas the incidence of confirmed syphilis cases in the general population significantly decreased compared to the previous year, the incidence of RPR positive blood donations did not change. RPR positivity was higher among male first-time donors, especially those over 20 years, peaking in the 30's. In absolute numbers, the highest values were observed among male repeat donors, especially in the 40's or over. Among female donors, the highest incidences were observed among first-time donors in the 20's and 30's, but in absolute numbers, repeat donors in the 20's and 30's represented the majority. Although in the general Japanese population, the highest absolute number of syphilis is reported among women in the 20's, the highest numbers among blood donors were observed in male.

Summary / Conclusions: Congruent with the increase of syphilis cases in the Japanese population, the number of RPR positive blood donors has also increased, but at a lower rate. Although not as high as in the general population, among female donors, the highest numbers and incidences are observed among those in the childbearing age (20's

and 30's), which implicates the risk of congenital syphilis. In addition, the high numbers of syphilis among repeat donors pose risk of losing loyal blood donors. Although our present results indicated that blood donors seem to behave differently from the general population, and the donor medical interview seems to be effectively excluding high risk groups from donating, it is a critical situation that needs to be urgently managed.

P410 | *Cutibacterium acnes* does not enhance the release and accumulation of proinflammatory factors in platelet concentrates during storage

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Background: *Cutibacterium acnes*, a commonly isolated bacterial contaminant of platelet concentrates (PCs), is often transfused into patients before contaminated product can be retrieved due to the slow growing nature of this bacterium in culture media used during routine bacterial screening. *C. acnes* plays an immunomodulatory role in immune responses by eliciting the release of proinflammatory factors (PF) such as cytokines. A few mild transfusion reactions have been attributed to *C. acnes* contamination, and this number is considered an underestimate as it is often difficult to discern if mild symptoms stem from the transfusion event or an underlying condition. Currently, it is unknown whether *C. acnes* contamination of PCs can elicit the release of PF from platelets during PC storage. This is pertinent since the accumulation of PF in PCs during storage has been linked to non-infectious transfusion reactions like febrile non hemolytic transfusion reactions (FNHTR) and to transfusion related acute lung injury (TRALI). Since *C. acnes* contaminated PCs are often transfused, it is important to elucidate if patients are exposed to elevated concentrations of PF thereby increasing their risk of non-infectious transfusion reactions.

Aims: Determine if *C. acnes* contamination of PCs results in an enhanced pro-inflammatory PC profile during storage.

Methods: Four *C. acnes* PC isolates belonging to different phylotypes, and one *Staphylococcus aureus* PC isolate were used in this study. In biological triplicates, four ABO-matched buffy coat PC units were pooled and split into 6 units of equal volume, and four of these PC units were inoculated with one *C. acnes* isolate each [10 colony forming unit (CFU)/mL], the fifth was inoculated with *S. aureus* (25 CFU/unit, positive control), and the sixth unit was inoculated with sterile saline solution (negative control). The units were stored under standard PC storage conditions (20-24°C/agitation/5 days) and sampled on days 0, 3 and 5 to determine bacterial concentration. Additionally, flow cytometry was used to assess platelet activation (CD41a – platelet marker, Annexin V and CD62p – platelet activation markers), and pro-inflammatory cytokine content (IL-1 β , IL-6, IL-8, IL-

10, IL-12, and TNF α), while soluble CD40L (sCD40L) concentrations were determined using ELISA.

Results: The concentration of all four *C. acnes* isolates remained stable (10 CFU/mL) until day 5, while *S. aureus* proliferated to a concentration of 10⁸ CFU/mL by the end of storage. The platelet activation profile, pro-inflammatory cytokine content, and sCD40L content of *C. acnes* inoculated PC units were comparable to the negative control during storage. On the other hand, *S. aureus* contaminated units displayed a significant increase in activation ($p \leq 0.01$), and IL-8 concentration (32.8 ± 0.5 pg/mL) by day 5 compared to the control. sCD40L concentrations gradually increased to $\sim 16.0 \pm 1.7$ ng/mL by day 5 in the negative control and *C. acnes* inoculated units, however, a significant reduction ($p \leq 0.01$) in sCD40L content (6.7 ± 0.2 ng/mL) was observed in *S. aureus* inoculated units on day 5.

Summary / Conclusions: PC units contaminated with *C. acnes* do not enhance the accumulation of PF during storage. Therefore, transfusion of *C. acnes* contaminated product may not pose an enhanced risk of inflammatory reactions when transfused to patients. Future work focused on the inflammatory response of mammalian cell lines to plasma derived from *C. acnes* contaminated PCs will help to elucidate residual risk.

P411 | A novel syphilis treponemal TmpA peptide antigen diagnostic assay using red cell codecytes in routine immunohematology column agglutination testing platforms

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Background: Syphilis is surging globally, with substantial increases noted in several countries. Recently, the TTIMS Program in the US reported a marked increase in syphilis prevalence in donations, which is considered to be a reflection of overall increasing trends in the general population. This resurgence is also seen in low- or middle-income countries (LMICs). Even though the infectivity of syphilis in the blood may decrease after 72 hours of cold storage, the residual risk of transfusion-transmitted syphilis may still remain. The laboratory diagnosis of syphilis is complex, and no single diagnostic assay is able to accurately determine both latent and active infections. Although treponemal antigen-based assays outperform non-treponemal assays, most of the LMICs cannot afford the more expensive treponemal antigen-based assays for syphilis screening; instead, they use a low-cost non-treponemal assay (e.g., rapid plasma reagin, RPR). Thus, there remains a need, especially in LMICs, for low-cost, high-sensitivity, easy-to-use treponemal antigen-based syphilis diagnostics, ideally those suitable for use with existing laboratory resources. With the well established peptide sequence of the treponemal spirochete *T. pallidum*, we explore the development of a novel assay by creating syphilis peptide red cell codecytes (TmpA-codecytes) for use

on existing routine blood antibody screening platforms. The cost of the TmpA-constructs per assay volume of TmpA-kodeocytes is 1 cent (US).

Aims: We evaluated the feasibility of using Kode Technology to make TmpA-kodeocytes for use in two column agglutination testing platforms (CAT-BioRad and CAT-Ortho Clinical Diagnostics).

Methods: Candidate Kode Technology function-spacer-lipid (FSL) constructs were made from the TmpA protein of *T. pallidum*, using the peptide and FSL selection algorithms, and then used to make kodeocytes. From three TmpA-FSL constructs, one was found to be the most suitable for diagnostics. Preparation of kodeocytes involved mixing a solution of FSL construct (e.g., 2.5 µmol/L) with washed packed group O red cells, incubation at 37°C for 2 h, and storage in a red cell stabilizer solution at 4°C. Kodeocytes were rested overnight before testing and were used within 21 days without washing. Developmental kodeocytes were evaluated against a large range of syphilis antibody-reactive and non-reactive samples in CAT and compared against established methodologies. 150 reactive (R) and 2072 non-reactive (NR) blood donor samples based on treponemal antigen-based assays were used to evaluate the performance of the developed assay. The RPR was also run in parallel.

Results: Among 150 treponemal antigen-based assay R samples, 146 were TmpA-kodeocyte reactive (97.3% correlation), compared to 58.0% with the RPR assay for the same samples. Against the 2,072 expected Syphilis NR samples, the agreement rate for TmpA-kodeocytes was 98.8%. External WHO/CDC syphilis quality control samples further validated the sensitivity of the TmpA-kodeocytes, as did the non-reactive results, with 157 samples reactive for non-syphilis pathogens and autoantibodies.

Summary / Conclusions: TmpA-kodeocytes are viable for use as cost-effective serologic reagent red cells for the detection of syphilis antibodies with a high level of specificity. This kodeocyte methodology potentially allows for the introduction of reverse algorithm testing into low-volume resource-constrained laboratories, as it utilizes existing transfusion laboratory infrastructure without adding any extra expenditure.

P412 | Abstract withdrawn

P413 | Culture-positive platelet donations at a hospital-based blood bank

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P413 - Table: Type of organisms identified in culture-positive platelet units (n = 27)

Type of Bacteria	Frequency (%)
<i>Staphylococcus epidermidis</i>	8 (29.8)
<i>Staphylococcus haemolyticus</i>	2 (7.4)
<i>Staphylococcus capitis</i>	1 (3.7)
<i>Staphylococcus hominis</i>	1 (3.7)
Coagulase-negative staphylococcus	2 (7.4)
<i>Micrococcus luteus</i>	2 (7.4)
<i>Bacillus</i> species	2 (7.4)
Other	9 (33%)

Background: Bacterial contamination of platelets stored at room temperature poses the highest risk of infection and fatality among blood components. Mitigation procedures involve optimizing arm disinfection, diverting initial blood samples, using pathogen inactivation, and bacterial detection through immunoassays, metabolic activity assays, or bacterial culture. Our center performs platelet culture using BacT/ALERT system on all apheresis, buffy coat and random platelet components donated in-house. BacT/ALERT is an automated microbial detection system that is commonly used for bacterial detection. For platelet-rich plasma and buffy coat platelet units, pooled samples from individual platelet units (pooled samples from three platelet units) are inoculated into one aerobic pediatric culture bottle and incubated for 5 days. For apheresis units, sampling is performed per unit without pooling. If positive BacT/ALERT results on the pooled sample, repeat testing from individual units or related components is performed. Initial bacterial identification often utilizes Gram staining, followed by confirmation and further characterization through automated systems like Phoenix and MALDI-TOF mass spectrometry. Sample inoculation is performed minimum of 12 hours after donation or 16-24 hours after processing the whole blood unit (whenever is feasible).

Aims: This retrospective study aims to determine the prevalence of culture-positive platelet units in blood donations in our center. Additionally, the study aims to identify the types of organisms found in culture-positive units and assess their clinical significance.

Methods: A retrospective review of the blood bank's records (spanning from 2018 to 2023) was performed. We analyzed the number of positive platelet samples, identified the organisms present, and assessed their clinical significance. Ethical approval was obtained from the institutional research ethics committee.

Results: A total of 5009 platelet units were cultured during the study period. The initial positive BacT results rate of all culture bottles/sample pools was 1.9% with half being *Staphylococcus* species (14, 52%). All rest of identified organisms were environmental opportunistic pathogens (Table). Only 5 platelet units were

confirmed positive after repeat testing (1:1000 units); one platelet unit due to gram positive bacilli (which was discarded), and 4 platelet units due to *Micrococcus luteus* (all inoculated on the same day). These 4 platelets units were transfused prior to the finalization of initial culture results, and repeat cultures were performed from related components. There were no clinical sequelae observed in the recipient patients, and blood cultures in these patients tested negative.

Summary / Conclusions: This study confirms a very low rate of positivity of platelet unit cultures at our center, with the most common cause being *staphylococcus* species. While many of the identified bacteria are of an environmental origin, some can be pathogenic with their pathogenic potential depends on various factors including the host's immune status and the site of infection. Continued assessment and monitoring is essential to ensure blood safety.

P414 | Microbial findings in vascular allografts before and after antimicrobial decontamination

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Background: Since 2020, South Danish Transfusion Service and Tissue Centre at Odense University hospital has been responsible for processing and storage of vascular allografts harvested from organ donors. Vascular allografts are used to replace cardiac valves and arteries in patients with mycotic aneurysms, endocarditis or infected autologous grafts. Given the prevalent contamination of grafts during procurement, establishing an effective protocol for microbial detection and decontamination prior to transplantation is imperative.

Aims: To describe microbial findings in vascular allografts both pre- and post-treatment with an antimicrobial solution and to explore patterns of identified microbial species.

Methods: Grafts were procured in the operation room immediately after harvesting of organs and transported at 4°C to the tissue center for processing and storage. Prior to cryopreservation, grafts underwent a 24-h incubation at 37°C in an antimicrobial solution targeting pathogenic bacteria and fungi. Until December 2021, Cambridge Antibiotic Solution (Bioscience) was used, and after that Base128 (Alchimia). Pre- and post-antibiotic treatment, a tissue sample from each graft were homogenized using a GentleMACS dissociator (Miltenyi Biotec), then transferred to two blood culture bottles (BACT/ALERT FA Plus and FN Plus), which were incubated in a BACT/ALERT® VIRTUO® (bioMérieux) until reported positive or for a maximum of 6 days if negative. Isolates from positive samples were subcultured and identified using MALDI-TOF MS.

Results: In all, 155 grafts from 29 donors were included from November 2020 to December 2023. Each donor provided 2-12

grafts. Before antibiotic treatment, microorganisms were detected in 41 homogenized tissue samples from 19 donors (66.5%). Regarding the isolated microbial genera, 46 (from 15 donors) belonged to species normally found on the skin (*Staphylococcus*, *Bacillus*, *Micrococcus*, *Propionibacterium*), 8 (from 5 donors) originated from the gut (*Candida*, *Enterobacter*, *Klebsiella*, *Lactobacillus* and *Hafnia*) and 2 isolates were from the oral flora (*Streptococcus*). Following antibiotic treatment, only two donors had culture-positive samples. *Paenibacillus urinalis*, considered susceptible to vancomycin in the BASE 128 antibiotic solution, was detected in a single sample from one donor. This graft was discarded following a risk-based assessment. *Streptococcus mitis* Group was found in three samples from the second donor, which were considered susceptible to the imipenem in the Cambridge Antibiotic Solution. None of these species were detected in the pre-treatment samples from the two donors. The overall discard rate was 0.6% (1 out of 155). In 15 of 19 (78.9%) donors with positive microbial findings, more than four grafts per donor were procured. In donors without microbial findings, more than four grafts per donor were procured in 2 of 10 donors (20%).

Summary / Conclusions: Microbes was detected in 41 of 155 grafts (26%), before antibiotic treatment. However, following antibiotic treatment, microbes could only be detected in four samples from two donors. This observation suggests that the antimicrobial decontamination process in our tissue bank is highly effective, thereby ensuring safe utilization of allografts in patient treatment. The majority of microbes detected are likely contaminants originating from the skin or gut of the organ donors rather than from the graft processing conducted at the tissue center.

P415 | Sociodemographic factors associated with reactivity in syphilis serology among Brazilian blood donors from January 2020 to February 2022

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Background: A concerning increase in syphilis has been observed worldwide. Traditional surveillance is limited by the scarce routine testing of asymptomatic infections. Blood donation screening is an

P415 - Table 1: Prevalence of current syphilis by donor characteristics and adjusted prevalence ratios (PR) estimating associations between selected variables and current syphilis.

		Prevalence per 100,000 (95% CI)	Adjusted PR (95% CI)
Gender	Female	583 (558-608)	1
	Male	580 (559-601)	1.15 (1.09-1.22)
Age (years)	≤24	548 (512-583)	1
	25-34	708 (675-742)	1.69 (1.55-1.83)
	35-44	497 (469-526)	1.42 (1.30-1.55)
	45-54	516 (479-553)	1.60 (1.44-1.77)
	≥55	630 (570-690)	2.07 (1.83-2.33)
Race	White	421 (398-444)	1
	Black	946 (876-1017)	2.00 (1.82-2.20)
	Mixed	644 (620-668)	1.39 (1.30-1.49)
	Asian	161 (74-248)	0.56 (0.33-0.97)
	Indigenous	608 (14-1202)	2.05 (0.77-5.43)
Education level	≤Primary	788 (720-855)	2.35 (2.07-2.68)
	High School/technical	520 (494-546)	1.72 (1.54-1.91)
	≥University degree	262 (238-285)	1
Donation type	Replacement	822 (789-854)	1.42 (1.34-1.51)
	Community	461 (443-479)	1
Donot type	First time	1213 (1174-1252)	5.10 (4.76-5.46)
	Repeat	238 (225-251)	1

opportunity to monitor asymptomatic syphilis infections, providing public health insights into transmission.

Aims: To define syphilis prevalence, incidence and factors associated with infection among blood donors in five Brazilian blood centers.

Methods: Donations from Jan. 2020-Feb. 2022 were screened with treponemal EIA (enzyme immunoassays) or chemiluminescence immunoassays, followed by alternate EIA and rapid plasma reagin (RPR) testing. Samples with positive or indeterminate results in the alternate EIA were categorized as current (if RPR reactive) or resolved syphilis (nonreactive RPR). Current syphilis cases were further categorized according to RPR titers (<1:8 or ≥1:8). We report the prevalence of syphilis in first-time donations (FTD) and repeat donations (RD), incidence in RD, and use a multivariable binomial regression model to assess factors associated with current syphilis.

Results: Of 862,146 donations, 10,771 (1.3%) were reactive or indeterminate on screening; 7562 available samples underwent additional testing. Of those, 5892 (77.9%) tested positive on the alternate EIA; 911 (12.0%) were resolved infections, 2990 (39.5%) were current syphilis with RPR < 1:8, and 1991 (26.3%) were current syphilis with RPR ≥ 1:8. The prevalence of syphilis infection, including resolved and current cases, was 2.5% among FTD and 0.6% among RD. Subgroups with the highest prevalence of current syphilis were black donors (946/10⁵ donations), donors with ≤primary education (788/10⁵ donations) and replacement donors

(822/10⁵ donations). In the multivariable model, age, race, education, donor type, and type of donation were significantly associated with current syphilis. Male donors had 1.15 times the prevalence of current syphilis compared to female donors (95% CI 1.09-1.22; Table). The incidence of syphilis in RD was 90/10⁵ person-years (95% CI 86-95).

Summary / Conclusions: The prevalence of syphilis was <3% among FTD and <1% among RD. We found wide variation according to demographics, with male gender, older age, black/mixed race, lower schooling, FTD, and replacement donations significantly associated with higher prevalence of current syphilis in the adjusted model.

P416 | Bacterial contamination of whole blood-derived platelet concentrates—results of a prospective multicentre study from Pakistan

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Background: Bacterial contamination of platelet concentrates (PCs) is a common cause of transfusion-related sepsis, which carries a high risk of morbidity and mortality. It is the most pressing challenge transfusion medicine professionals are now dealing with. The main cause of bacterial contamination is skin microflora. Additionally, storage conditions like gas-permeable blood bags kept at room temperature (20–24°C) and continuous agitation support bacterial growth. One study suggests that 1 in 2,000–2,500 platelet concentrates might have bacterial contamination. A number of strategies are used in advanced countries to lessen the possibility of bacterial contamination, such as optimal skin disinfection, transferring the first 10–30 ml of blood collected into a derivation pouch, using pathogen reduction technologies, and PC culture. According to Pakistan's National Quality Control Guidelines of 2020, at least 1% of PCs should be tested for quality control parameters (swirling, volume, yield, pH, and culturing).

Aims: To assess the prevalence of bacterial contamination and the characterization of the bacterial isolates in whole blood-derived platelet concentrates.

Methods: This prospective study included 1254 samples of 72 h post-donation PCs (whole blood-derived). PCs from three different sites were studied, including Peshawar Regional Blood Centre, Peshawar ($n = 766$), Mirpur Regional Blood Centre, Mirpur ($n = 425$), and Dr. Akbar Niazi Teaching Hospital Blood Bank, Islamabad ($n = 63$). Using aseptic technique, a 10 ml sample was obtained from the PC bag and inoculated into BD BACTEC™ aerobic/anaerobic platelet quality control testing culture bottles. The bottles were incubated in the BD BACTEC™ blood culture system for seven days at 37°C. Culture bottles indicating bacterial growth were subcultured, and microbiological tests were used to identify and classify bacterial strains. PCs not showing a positive signal after the 7-day incubation period were labelled as negative and not tested further.

Results: The study revealed that seven PCs were found to be contaminated, showing a contamination rate of 1 in 179 (0.55%). None of the platelet units with positive screening tests were ultimately transfused. The bacteria detected were consistent with skin microbial flora that are connected to non-fatal septic blood transfusion reactions. The majority (71.42%; $n = 5$) were coagulase-negative *Staphylococcus aureus*, with two cases of gram-positive *Propionibacterium acnes* (28.58%).

Summary / Conclusions: The rate of confirmed bacterial contamination of PC was comparable to that reported in developing countries, while it was on the higher side when compared with data from developed countries. This underscores the critical importance of implementing screening measures for bacterial contamination of blood products. Bacterial contamination can pose serious risks to transfusion recipients, including sepsis and other life-threatening complications. The blood transfusion authorities (BTAs) can play a pivotal role in ensuring compliance with national standards and guidelines to uphold the safety and quality of blood transfusion services.

P417 | Profile evaluation of blood donors with positive serology for syphilis

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Background: Syphilis, an ancient disease, is still a public health problem worldwide. Various strategies have been proposed to prevent transfusion transmitted syphilis, such as selection of low-risk donors and screening for the disease using efficient laboratory methods.

Aims: To evaluate the type of donation and the type of donor with positive serology results for syphilis in a hemotherapy service in the south of Brazil.

Methods: All donations with a reactive and/or indeterminate result for Syphilis were evaluated at the Hemotherapy Service of Hospital São Vicente de Paulo – Passo Fundo – RS from January to December 2019. Data were collected through the computerized system e-Delphyn and donor files and later inserted into an Excel spreadsheet. Tests to detect syphilis in donor samples were performed through chemiluminescence method, using the Syphilis TP kit from the manufacturer Abbott, on the Architect i1000 and Architect i2000 platforms. Donors with an initial reactive or indeterminate results were invited to collect a new sample for confirmatory tests.

Results: From the 13,537 donations during the period, 115 (0.8%) presented reactive or indeterminate results for syphilis. Regarding gender, 56 (48.7%) donors were male and 59 (51.3%) were female. Related to the type of donation, 36 (31.3%) were spontaneous and 79 (68.7%) were replacement donations. As for donor type, 86 (74.8%) were first-time donors and 29 (25.2%) were repeat donors. When evaluated together, first-time and spontaneous donors accounted for 25 (21.7%), repeated and spontaneous donors were 11 (9.6%), first-time and replacement donors were 61 (53.0%), and repeated replacement donors were 18 (15.6%). Donors were called for a new sample collection, and 68 (59.1%) returned, of which 17 (68.0%) were first-time and spontaneous donors, 6 (64.5%) were repeated spontaneous donors, 33 (54.0%) were first-time and replacement donors, and 12 (66.6%) were repeated replacement donors.

Summary / Conclusions: The positivity for syphilis among genders was similar. Serological inadequacy is more frequent in first-time donors, as it aligns with the results obtained in this study. Regarding the type of donation, there was a predominance of replacement donations. According to some authors, this type of donation has 5.6 times more chances of presenting positive serology than spontaneous donations. Replacement donations are generally carried out by family members who may omit information during clinical screening to complete the donation. The return rate for result confirmation is a noteworthy factor, as these donors may be unwell and contributing to syphilis transmission. It is essential, for the interruption of the transmission chain, that these individuals return, undergo confirmatory tests, receive guidance, and are referred for

treatment when necessary. Knowing the profile of donors with greater risk of presenting positive serology is important to direct recruitment campaigns for specific groups with lower chances of rejection in serological tests, contributing for transfusional safety and hemocomponents availability. These data are also essential for public health, guiding actions of awareness and prevention of sexually transmitted infections.

P418 | Impact of the introduction of specific treponemal antibody detection in syphilis screening in a center that had been using RPR for 30 years

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Background: Spanish legislation includes serological screening for syphilis among the requirements for donations of whole blood or its components. Spanish law allows donations from individuals with syphilis after one year of confirmed cure. In our transfusion center, the determination of non-specific antibodies (Ab) to cardiolipin (RPR, Rapid Plasma Reagin) as a marker of active infection has been performed since its inception until June 2021. Subsequently, the automated detection of specific treponemal Ab using the chemiluminescent micro-particle immunoassay (CMIA) was established as a screening technique for this infection. This has led to the detection of specific syphilis Ab in donors with negative RPR at the time of donation.

Aims: To analyze the results obtained in our center after implementing the determination of specific treponemal Ab., and its impact on the loss of donations and donors.

Methods: Descriptive and retrospective study of serological screening results for syphilis using RPR determination in the period from 2017 to 2020, compared with data obtained since the introduction of specific

treponemal Ab detection in the period from June 21, 2021 to Dec 2023. In the first period, donations with positive RPR were tested for the presence of specific treponemal Ab, and if these were negative, it was considered a false-positive (FP) result of RPR. In the second period, donations positive for specific treponemal Ab were analyzed by RPR and TPHA (Treponema Pallidum Haemagglutination Assay), considering: (i) active syphilis if RPR was positive, (ii) cured or latent syphilis if TPHA was positive and RPR negative, (iii) FP if both TPHA and RPR were negative.

Results: In the first period, 195,173 donations were analyzed, of which 85 (0.04%) were FP for RPR. In the second period, 75,262 donations were analyzed, of which 270 (0.35%) showed a reactive result (table 1). All 74 donors with cured/latent Syphilis were permanently excluded from donation, of which 50 (67%) were previous donors (table 2). At the beginning of the second period, the majority of diagnosed donors had donated during the first period (87.5%), and at the end of these 30 months, the majority of diagnosed donors with reactive serology are new donors (62.5%).

Summary / Conclusions: After the implementation of the detection of syphilis-specific antibodies in the screening of blood donations, a significant number of past cases of syphilis that were not detectable with the previous screening were identified and definitively excluded. With the introduction of specific Ab screening, the number of FP has increased more than 6-fold compared to those obtained with RPR. Nowadays, the transition has been completed and the loss of regular donors has practically disappeared, with positive results among new donors. In view of the current decline in donations, criteria should be agreed for the re-entry of donors with properly treated and cured syphilis. Consensus should also be sought on the acceptance of donors with FP results.

P419 | Presentation and analysis of blood donors with confirmed positive results for syphilis at the Blood Transfusion Institute of Serbia for the period 2020-2023

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Background: Although an ancient disease that was thought to be eradicated thanks to effective prevention and widely available therapy, Syphilis remains a major public health problem. Despite the fact that transmission of syphilis infection by transfusion has not been recorded for several decades, transfusion services still carry out testing of blood donors for this marker as mandatory, primarily as an indicator of risky sexual behavior. Data from the WHO, CDC, European Center for Disease Control, as well as national epidemiological services, in previous years record an increase in the incidence of this infection in the general population.

Aims: Presentation and analysis of the frequency of positive results of confirmatory testing for syphilis in the population of voluntary blood donors at the Blood Transfusion Institut of Serbia.

P418 - Table 1. Results of syphilis screening in the 2nd period

Year	Tested donations	Reactive Serology	FP	Cured / latent syphilis
2021*	25996	77 (0.29%)	45 (58.4%)	32 (41.6%)
2022	49266	106 (0.21%)	80 (75.5%)	26 (24.5%)
2023	48151	87 (0.18%)	71 (81.6%)	16 (18.4%)
Total	75262	270 (0.35%)	196 (72.6%)	74 (27.4%)

P418 - Table 2. Cured / latent syphilis: donors characteristics

Year	Cured / latent syphilis	Past donor	New donor
2021*	32	28 (87.5%)	4 (12.5%)
2022	26	16 (61.5%)	10 (38.5%)
2023	16	6 (37.5%)	10 (62.5%)
Total	74	50 (67.6%)	24 (32.4%)

Methods: A retrospective study was conducted in which the results of syphilis tests performed on 271 498 blood donors in the period (2020-2023) were analyzed. All tests were performed under the same conditions on automated platform using EIA tests (ELISA and CLIA). In donors with repeated reactive results of the screening test, in accordance with the national testing algorithm, confirmatory testing was carried out using immunoblot tests.

Results: From the total number of tested donors (271,498), a positive confirmatory (immunoblot) test for Syphilis was determined in 125 donors (0.0460%). Of the total number of confirmed positive donors, 109 (87.2%) were male, and 16 (12.8%) were female. If we look at the number of previous blood donations, 67 (53.6 %) confirmed positive results were detected in first time donors, the remaining 58 (46.6%) were detected in multiple donors. The percentage representation of positive donors was calculated according to age groups, for each individually observed year; overall, the age group of 31-40 years with 39 donors (31.2%) is the most represented. For 7 confirmed positive donors, we have information that they donated blood in the same year in which a positive confirmatory test was detected, where the shortest period since the previous (negative) blood donation was 5 months. In two donors, an associated infection is present (one HIV, other HBV, confirmed by PCR test). To all confirmed positive donors were sent a uniform letter with an invitation to the blood donors counseling center, in order to discuss the obtained test results and refer them to reference institutions for further diagnosis and treatment. 82 (68%) blood donors responded to the invitation. In the post-donation questionnaire, 5 donors provided information that they had previously been treated for syphilis, 55 donors did not exclude the possibility of risky sexual contact, and for 22 donors the risk factors were not defined.

Summary / Conclusions: Analysis of the data on the frequency of syphilis in the population of blood donors shows that 125 donors (0.0460 %) were positive for syphilis. Donors with a positive result of the confirmatory test are excluded from the further blood donation process. This research represents an important indicator of risky sexual behavior and points to the great importance of monitoring sexually transmitted diseases in the blood donor selection process.

P420 | Evaluation of doubtful or positive syphilis serology in blood donors

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Background: Syphilis testing is mandatory in biological qualification of blood donation to ensure transfusion safety. Accurate results are necessary to decide about the status of the donor and the validity of donation.

Aims: The aim of our study was to evaluate the result of positive or doubtful syphilis serology by different methods.

Methods: We conducted a prospective study over 5 months on the Military Blood Transfusion Center. It concerned 29 positives and 9 doubtful syphilis serology of blood donors tested by ELISA technique from Abia. These results were retested by at least one other method: hemagglutination assay (Fortress Diagnostics TPHA test) and or ELISA from Bio-Rad. False positive reactions were characterized and test specificity evaluated.

Results: Among positive ELISA (Abia) Syphilis serology, only 3 samples were found positive by the two other methods thus a percentage of 10.35% of true positive. False positive reactions were detected in 89.65% Abia ELISA tests (26 samples); 65.4% of them realized by two techniques and 34.6% by only TPHA reaction. Abia false positive values ranged in only 1 case between reaction cut off (0.381-0.385) and 0.5, in 12 cases (46.15%) in the Intervale of 0.5-1 and in 50% of cases >1. Nine Borderline Abia ELISA reactions (value ranging between 0.200 and cut off) were found negative when realized in 6 cases by TPHA and in 3 cases by ELISA from Bio-Rad. All doubtful or positive ABIA ELISA samples were excluded from donation. If considering as negative the results of all others tests when values are under cut off by Elisa Abia, the specificity of this method could be evaluated to less than 99.67%.

Summary / Conclusions: Doubtful or positive reactions should be retested by different method to avoid editing false positive result which can perturbate donors; in the other side, false negative results can be harmful for blood receiver that's why taking into account the specificity and the sensitivity of the method is primordial in the choice of the best laboratory testing tool.

Transfusion transmitted infections—parasites

P421 | Analytical sensitivity of a donor screening assay for *Plasmodium* ribosomal RNA and DNA

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P421 - Table: LODs for aRNA of each species.

Species	50% LOD aRNA particles/mL	95% LOD aRNA particles/mL
Pf	8.2 (6.8-9.5)	27.9 (22.5-38.5)
Pm	9.0 (7.5-10.4)	32.2 (25.8-44.7)
Pv	8.2 (6.7-9.7)	33.1 (26.3-46.5)
Po	9.5 (7.5-11.4)	59.0 (44.1-90.3)
Pk	6.6 (5.2-7.8)	23.7 (18.9-33.6)

Background: The investigational cobas® Malaria test* for use on the cobas 5800/6800/8800 Systems is a qualitative donor screening test for the detection of nucleic acid of the 5 major species of *Plasmodium* responsible for human infection: *P. falciparum* (Pf), *P. malariae* (Pm), *P. vivax* (Pv), *P. ovale* (Po) and *P. knowlesi* (Pk). For use of the assay, 1.1 mL of whole blood (WB) is collected into 7.7 mL of a lysis/storage medium (LM) which lyses cells and any parasites present and stabilizes the nucleic acid. The lysate is tested with the cobas Malaria test on the automated cobas 5800/6800/8800 Systems. The design is intended to detect ribosomal RNA (rRNA) and DNA.

Aims: To establish the analytical sensitivity of the cobas Malaria test.

Methods: The limit of detection (LOD) for intact infected red blood cells (iRBC) was established using RBC from fresh Pf infected cultures. The iRBC concentration was determined by microscopy, and the material serially diluted in *Plasmodium*-negative WB. 1.1 mL aliquots of specific concentrations of iRBC in WB were inoculated into 7.7 mL of LM and each concentration tested in 135 replicates by the cobas Malaria test. The analytical sensitivity for the rRNA of each species of *Plasmodium* was established using armored RNA (aRNA). aRNA are non-infectious, recombinant particles encoding a single copy of *Plasmodium* target rRNA sequence encapsulated by bacteriophage coat protein to protect the RNA from degradation. aRNA particles for each species were serially diluted in cobas omni Specimen Diluent and tested in 71 or 72 replicates. LODs (with 95% confidence intervals (CI)) were calculated by PROBIT analysis.

Results: The 50 and 95% LODs for intact Pf iRBC were 0.6 (0.5-0.7) and 2.9 (2.4-3.8) iRBC/mL, respectively. The LODs for the aRNA of each species are shown in the Table.

Summary / Conclusions: The 95% LOD for intact iRBC is consistent with the expected results for an organism containing a high number of target copies. A previous study reported 7400 copies of rRNA per ring-staged parasite (Seilie A et al., Am J Trop Med Hyg 2019). Given the high copy number, if an organism is captured in the sample it would be detected. Therefore, the LOD is the concentration providing for 95% probability of having at least 1 organism in a randomly drawn sample, which is an average of 3 organisms per sample volume in accordance with Poisson distribution. The aRNA studies establish a similar sensitivity of the assay for the RNA targets of the 5 species. The cobas Malaria test provides a new high sensitivity option for reducing the risk of transfusion-transmitted malaria. * The cobas® Malaria test is in development and not commercially available.

P422 | Laboratory detection of donors implicated in transfusion-transmitted malaria

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Background: Transfusion-transmitted malaria (TTM) is a rare but potentially fatal complication of transfusion in non-endemic areas (non-EAs). A previous publication (Niederhauser & Galel, Transfus Med Hemother 2022) systematically reviewed TTM in non-EAs. This

P422 - Table 1: Donor PCR results by sample type for TTM cases in US and Canada

Case #: Country, year, dnr risk, species	Fresh f/u sample	Retained sample from index donation		
		Blood segment	Plasma	Undefined sample type
1: US, 2010, FR, Pf	Pos			
2: US, 2011, FR, Pm	Pos			Neg
3: US, 2016, FR, Pf	Pos			
4: US, 2017, FR, Pf	Neg	Pos nested PCR, Borderline PET-PCR		
5: US, 2017, FR, Po		Neg		
6 (BMT): US, 2018, T, Pf	Pos			
7: US, 2020, FR, Pf		Neg		
8: Canada, 2022, FR, Pf	Pos			

P422 - Table 2: Donor PCR results by sample type for TTM cases in Europe

Case #: Country, year, dnr risk, species	Fresh f/u sample	Retained sample from index donation		
		Blood segment	Plasma	Undefined sample type
9: Netherlands, 2011, T, Pm	Pos		Neg	
10: France, 2012, FR, Pf	Pos		Pos	
11: France, 2015, FR, Pm	Pos		Neg	
12: Italy, 2019, M, Pm	Pos			
13: Austria, 2019, T, Pf	(dnr lab results not reported)			

current review analyzes the laboratory results of donors (dnrs) implicated in TTM in non-EAs.

Aims: To assess the detectability of *Plasmodium* infection by molecular or antibody testing of dnrs who caused TTM in non-EAs.

Methods: Cases of TTM in US, Canada, and Europe since 2010 were identified through publications in the prior review and additional publications since that review. Authors and labs were contacted to solicit missing details about sample types and lab methods where possible. Results of tests performed on retained samples from the index donation causing the TTM and/or fresh follow-up (f/u) samples were summarized.

Results: 12 cases of TTM and 1 case of bone marrow transplant (BMT)-transmitted malaria were reviewed, including 7 cases in the US, 1 in Canada, and 5 in Europe. The implicated dnrs included 9 former residents (FR) of an EA, 1 former missionary (M) in an EA, and 3 travelers (T) to EAs. The tables show all cases reviewed, and results of polymerase chain reaction (PCR) for various dnr sample types tested. PCR results were reported for 12 of the 13 implicated dnrs. PCR was positive in at least 1 sample from all dnrs tested, except for 2 cases (Cases 5 & 7). In those 2 cases, PCR was performed only on retained segments of the blood donation that had been refrigerated for several weeks. All dnrs tested on a fresh sample were PCR positive except Case 4 who was positive on a retained sample but not a fresh sample. One positive result was reported in a stored plasma sample. Where described, PCR assays were laboratory-developed tests (LDTs). Analytical sensitivity is available for the assays used in Cases 4-8, and is approximately 3000-6000 infected red blood cells per mL. ELISA results were reported for 7 dnrs. Of these, 3 (Cases 1, 6, 7) were positive and 4 (Cases 2, 9, 10, 11) were negative.

Summary / Conclusions: This review found that antibody ELISAs failed to detect 4 of the 7 TTM dnrs tested. PCR testing, largely or entirely by LDTs, was able to detect *Plasmodium* infection in all dnrs tested except for 2 dnrs tested only on samples likely to have deteriorated from prolonged refrigerated storage. Recently developed ribosomal RNA-based molecular dnr screening assays are reported to be approximately 1,000 fold more sensitive than these LDTs. Prospective dnr screening performed on fresh samples using these new more sensitive molecular tests holds promise as a potential method to further reduce TTM.

P423 | The Introduction of malaria antibody testing at the Irish Blood Transfusion Service

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Background: Malaria Antibody Testing (MAT) was introduced by the Irish Blood Transfusion Service (IBTS) in May 2023. Prior to the introduction of testing, donors who met the following criteria were

permanently excluded from donating blood; A donor who lived in any endemic malarial area for at least 3 months during the first 5 years of life, A donor who lived in any of the malarial areas of sub-Saharan Africa, Papua New Guinea or West Papua for a continuous period of 6 months or more at any time, A donor who experienced an unexplained fever during their visit to or in the 4 months after their return from a malarial area, A donor with a history suggestive of, or a confirmed diagnosis of malaria. With the introduction of MAT, all of the above donors are eligible to donate. A resident of a malarial area is a donor who spent 6 months or more continuously at any time of life in any malarial endemic area. A sample-only collection policy is used on the first attendance. Donors are eligible to donate 42 days later, providing there is no reactivity on testing. Donors will have a MAT on every subsequent donation. Visitors are deferred for 12 months from date of leaving the at risk area.

Aims: To review the introduction of MAT at the IBTS. To describe the results of testing and to review the donor demographics of those tested.

Methods: Selected samples are screened for malaria antibodies using the Trinity Biotech Malaria EIA. Reactive samples are repeated in duplicate, RR samples are referred to the Microbiological Services Laboratory (NHSBT, London) for confirmatory testing. All MAT results from May to December 2023 were reviewed to determine the RR rate. RR donors were classified according to their confirmatory results as False Positive, Inconclusive, Past Infection (serology only positive) and Current Infection (PCR positive).

Results: A sample was collected from 569 donors for MAT between May and December 2023, 536 donors were eligible to donate based on a negative result. Of the donors that were screened, 57.2% reported their ethnicity as White (328/569), 13.3% Indian sub-continent (76/569), 7.7% any other Asian (44/569), 6.8% Black (39/569), 3.3% Arab (19/569), 2.4% any other ethnicity (14/569), 1.9% as mixed (11/569) and ethnicity was not disclosed for 7.3% (38/569). 743 samples underwent MAT, 33 RR samples were recorded, giving a RR rate of 4.45%. Of these, 6.1% were false positive (2/33), 6.1% were inconclusive due to discordant confirmatory serology (2/33), 84.8% were past infection (28/33) and 3% positive (1/33). P. ovale DNA was detected in the positive sample. Of the donors classified as past or current infection (29/33), 65.5% reported a history of malaria (19/29), 31% were residents of a malarial area (9/29) and 3.4% had a history of unexplained fever (1/29). RR donors are permanently deferred, however they can be retested after 3 years. In certain circumstances, such as those donors with a rare red cell phenotype, donors may be retested after 1 year. Of the donors that tested RR for malaria antibodies, 48.5% reported their ethnicity as Black (16/33), 18.2% Indian sub-continent (6/33), 15.2% White (5/33), 9.1% mixed (3/33), 3% any other ethnicity (1/33), and ethnicity was not disclosed for 6.1% (2/33).

Summary / Conclusions: The introduction of MAT has facilitated the recruitment of more ethnically diverse blood donors. Continual monitoring of the performance of MAT and the epidemiology of malaria in the donor population is required for the development of evidence based screening policies.

P424 | Screening and testing of blood donors for Chagas disease based on recent travel destinations—results from an international survey conducted by the ISBT TTID working party

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Background: Chagas disease, also known as American trypanosomiasis, is a life-threatening disease caused by the protozoan *Trypanosoma cruzi*, which infects humans primarily through exposure to the feces of triatomine bugs. An estimated 11 million people may be infected worldwide, mainly in Latin America where Chagas disease is endemic. Some infected individuals show no symptoms but can transmit the parasite horizontally through blood donation or vertically to their fetus.

Aims: The current study aimed (1) to compare international practices among blood services regarding the screening and testing of donors who returned from an area where Chagas disease is endemic and (2) to determine the *T. cruzi* positivity rate among these at-risk blood donors.

Methods: On September 18, 2023, a worldwide survey was sent to employees of blood services on behalf of the ISBT TTID-parasites and SRAP subgroups. The survey questions pertained to (1) the endemicity of Chagas disease in the blood service's region; (2) whether a blood service screened and tested blood donors for *T. cruzi* based on the duration of the stay in an area where Chagas disease is endemic; (3) the years of implementation of the test for *T. cruzi*; (4) the test used; (5) the number of tests carried out since implementation; and (6) the number of confirmed-positive donations among donors who returned from an area where Chagas disease is endemic (and did not have other risk factors).

Results: Twenty-six respondents — 5 (19.2%) in Africa, 4 (14.4%) in Asia, 9 (34.6%) in Europe, 3 (11.5%) in North America, 1 (3.8%) in Oceania, and 4 (14.4%) in South America — completed the survey. Twenty-one (80.8%) operated in a non-endemic region. Of these, 9 (42.9%) tested blood donors for *T. cruzi*, 8 (88.9%) of which tested blood donors for *T. cruzi* due to the duration of the stay in an endemic country. The median year of introduction of the travel criterion for *T. cruzi* was 2010 (range = 2006–2016) among blood services. The participating blood services tested blood donors for *T. cruzi* after the following length of stays (in consecutive days) in an at-risk area: ≥180 days at 4 blood services (50.0%), ≥120 days at 1 blood service (12.5%), ≥30 days at 1 blood service (12.5%), and <30 days at 2 blood services (25.0%). All (except one blood service) used ELISA for screening, but the methods for confirmatory testing were more heterogeneous and included NAT, ELISA, Western Blot, indirect immunofluorescence, and line immunoblot assay. Among the screened centers, which tested >308,000 donations from donors with travel-related risk factors since implementing the

test, no donation tested positive based on the duration of the stay in an endemic country alone.

Summary / Conclusions: Given the absence of *T. cruzi*-positive donations reported by participating blood services, travelling to an endemic area does not appear to be a risk factor for Chagas disease, regardless of the duration of the stay in an at-risk area. The pre-donation questions related to the length of stay in an at-risk area (alone) might, therefore, be safely removed.

P425 | Transfusion-transmitted babesiosis due to travel escapes regional testing approach

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Background: Before *Babesia* nucleic acid testing (NAT) was implemented in endemic areas in May 2020, *B. microti* was considered the leading transfusion-transmitted (TTB) infection in the US. During the first two years, the regional testing approach was successful, and the number of TTB cases was dramatically reduced and eliminated for donations collected in endemic areas. Two cases were reported in 2021 from states bordering endemic areas from donors without relevant travel history. However, in 2023, three TTB cases due to travel were reported.

Aims: Provide an update on the efficacy of *Babesia* NAT testing limited to endemic areas.

Methods: All American Red Cross (ARC) donations collected in 13 endemic states and Washington DC are tested for *Babesia* using a licensed NAT assay targeting 18S RNA of four species of *Babesia* (*B. microti*, *B. divergens*, *B. duncani*, and *B. venatorum*). In non-endemic areas, the only prevention measure remains a question in the donor history questionnaire to assess if a donor had ever had a diagnosis or a positive test result for *Babesia*. Donors who tested positive for *Babesia* or answered “yes” to the history of babesiosis questions are deferred for two years.

Results: In the three years since test implementation, ARC has identified positive donors in five TTB cases reported in non-endemic states, corresponding to about 1 case every 1,900,00 unscreened donations. Of the five cases, three cases reported in 2023 were associated with travel. Two of the three donors were residents of non-endemic states (Michigan and Georgia), and the donations were transfused in Indiana and Florida. Both donors reported extensive travel to endemic states (Connecticut, New York, and Maine). The third donor was a resident of an endemic area (New Jersey) who donated during a vacation in a non-endemic state (Oregon).

Summary / Conclusions: *Babesia* testing implementation coincided with the beginning of the pandemic and a dramatic reduction in travel, which could have further reduced exposure to the parasite. But as travel resumes, cases of TTB associated with donors moving to and from endemic states are emerging. While *Babesia* testing has demonstrated high effectiveness, and no case of TTB has been attributed to a tested unit, the regional testing approach does not protect from infections acquired during travel or endemic area residents donating in non-endemic states. Making *Babesia* a nationwide required assay should be considered to eliminate the risk of TTB.

P426 | Detectable molecular screening for malaria in a non-endemic area of a candidate for blood donation - case report

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Background: In 2015, the global technical strategy for reducing malaria transmission was published by the World Health Organization (WHO, 2015), defining strategic pillars of how national programs should be sustained. Brazil launched, in the same year, the plan for the elimination of malaria caused by *Plasmodium falciparum* until 2035, considering the final five years only with transmission of malaria caused by *Plasmodium vivax* (Brazil, 2022).

Aims: To report a case of detection of *Plasmodium spp* by means of the Nucleic Acid Test (NAT) in a blood donor in a non-endemic area for malaria.

Methods: Case report of a non-endemic area - municipality of Passo Fundo, Rio Grande do Sul, Brazil. Male donor, 25 years old, resident of Passo Fundo/RS, of Venezuelan nationality, who had his first blood donation at the Hemotherapy Service of the São Vicente de Paulo Hospital on December 1^o, 2023. Serological tests of the donation were performed, showing a non-reactive results for Anti-HIV 1 and 2, Anti-HTLV I/II, Anti-HBc, HBsAg, Anti-HCV and Chagas disease; and presented with a reactive serological result for syphilis; all tests previously mentioned were performed in a primary tube using the chemiluminescence method via Architect i2000 Abbott kits. All donations are sent to the NAT HIV/HBV/HCV/Malaria test, performed by the SIT NAT located at the Center for Hematology and Hemotherapy of Santa Catarina (HEMOSC). The NAT as a screening for donors is part of a national program in partnership between the Ministry of Health (MS) and the General Coordination of Blood and Blood Products (CGSH). The kit implemented was the KIT NAT PLUS HIV/HBV/HCV/MALÁRIA BIO-MANGUINHOS. According to current legislation (Consolidation Ordinance No. 5/2017), in non-endemic regions of malaria, the criteria of unfitness of candidates for donation depends on the exposure to endemic areas. The Ministry of Health launched, in 2022, the National Plan for the Elimination of Malaria, whose objectives are to reduce mortality and severity of cases, the incidence of the disease, to keep the disease absent in places where the transmission has already been interrupted and eliminate it from Brazil (Brazil, 2022), reinforcing the importance of testing nationally as a result of the increase in immigration in the country.

Results: The NAT indicated a detectable result for malaria in a pool, which presented detectable results in the individual testing. The sample was forwarded for species identification, and *Plasmodium vivax* was identified. The blood components from the donation were discarded and the donor summoned for a new sample collection.

Summary / Conclusions: Epidemiological studies have highlighted the importance of malaria in endemic and non-endemic regions and

therefore the inclusion of the NAT is crucial in the prevention of transfusions contaminated by this pathogen, consequently increasing transfusion safety. The sensitivity of the tests used in the screening of donors is an important factor, since it was determined that inoculums with 10 parasites per unit of red blood cells are sufficient to transmit the infectious agent (PEREIRA, 2011). The NAT for the detection of the nucleic acid of *Plasmodium spp* aims to reduce the period of ineligibility for blood donors who were in areas considered endemic, from 12 months to 1 month, in addition to detecting the agent in asymptomatic carriers who, despite clinical/laboratory screening, are considered suitable for donation.

Transfusion transmitted infections—newly emerging and other transfusion related pathogens

P427 | CRISPR/Cas12a-based methods for the fast and simple detection of dengue virus 3

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Background: Despite the development of numerous molecular diagnostic methods, the need of simple system is high in the context of emerging infectious diseases. Furthermore, the problem of cross reactivity still remains concerning, because of the high degree of structural and sequence homology between flavivirus. Recent advances in CRISPR-based technology allowed the development of new diagnostic approaches: CRISPR/Cas12a system and its collateral cleavage activity of adjacent ssDNA reporters is a promising tool for viral diagnosis.

Aims: Aiming to develop new methods for rapidly amplify and detect Dengue virus 3 (DENV3), we combined in a single tube the CRISPR/Cas12a system with an isothermal reverse transcription recombinase polymerase amplification (RT-RPA). The result is visualized with naked-eyes, using a lateral flow strip or by the fluorescence emission directly in the reaction tube.

Methods: DENV3 is selected as model in this study given a recent outbreak (Martinique, 2020). The samples were tested in a single tube by pairing an isothermal RT-RPA with the CRISPR/Cas12a reaction. Amplicons were targeted by a crRNA guide that hybridizes specifically to DENV serotype 3. This recognition event activates the Cas12a trans-activity cleavage of ssDNA reporters. Two formats of detection were developed based either on a visual detection. On one hand, the result can be visualized on a lateral flow strip. On the other hand, a detection of fluorescence can be obtained. Assays were performed on

28 clinical DENV samples, 30 blood donors who had no history of arbovirus contact and reference titrated materials.

Results: No cross reaction were observed, either by fluorescence or lateral strip detection, showing the specificity of the crRNA guide to complementary DENV3 amplified genomes. The analytical sensitivity was the same for both methods (10TCID₅₀/mL). The diagnostic sensitivity and specificity were 85.71% (95% CI, 72.75-98.68%) and 96.66% (95% CI, 90.24-100.00%) respectively using the lateral flow detection. By detecting the fluorescence emission directly in the reaction tube, the sensitivity and specificity were 89.28% (95% CI, 77.83-100%) and 100% (95% CI, 81.75-100.00%) respectively.

Summary / Conclusions: This new analytical strategy provide perspectives for developing innovative assays for the molecular diagnosis of emerging viruses. The specificity and the rapidity of the test is of interest including differential diagnosis arbovirus in case of similar clinical signs or for rapid blood testing, meaning an improvement of the prevention and the surveillance of arboviral infections. Finally, one of the major advantages of this test is its flexibility, allowing to be reactive in case of a new emerging virus.

P428 | Blood donor screening in endemic regions—insights from a 2023 arbovirus epidemic wave study in Brazil

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Background: Dengue fever, a mosquito-borne viral infection, poses a significant public health threat in endemic regions worldwide. With the transmission of dengue through blood transfusions documented, ensuring the safety of the blood supply is of utmost importance. This abstract explores the importance of dengue screening protocols for blood donors in endemic regions where the prevalence of dengue is high and the risk of transfusion-transmitted infections escalates.

Aims: To screen asymptomatic donors by nucleic acid testing (NAT) during the arbovirus epidemic wave of 2023 (May to June) in endemic regions in Brazil.

Methods: Sixty asymptomatic donors were invited to participate in this study. After signing the consent form, 5 mL of whole blood was collected using EDTA as an anticoagulant. Automatic RNA extraction from plasma was applied, followed by reverse transcription real-time PCR using an in-house methodology.

Results: During May and June 2023, a total of 2493 blood donations were processed at the primary blood donation center. From this pool, we obtained 60 plasma samples, representing 2% of the overall

collections. In June, we detected a single positive DENV-RNA sample (1/60, 1.6%). The donated blood was discarded. The donor, a repeat blood donor, exhibited no symptoms of arbovirus at the time of donation, and subsequent monitoring has revealed no record of arbovirus symptoms post-donation.

Summary / Conclusions: The detection of a positive DENV-RNA sample in an asymptomatic donor emphasizes the need for a proactive approach to dengue screening in blood donors within endemic areas. The integration of advanced diagnostic technologies, specifically nucleic acid amplification, is essential to ensure the blood supply's safety in regions with high dengue prevalence. This result is a valuable insight into the discussions on strategies to enhance dengue screening protocols and mitigate the risks associated with blood transfusions in endemic regions. Continuous research efforts are crucial in addressing the dynamic challenges posed by dengue in the context of blood safety.

P429 | Parvovirus B19 surveillance in Swiss blood donors—comparison of current and previous outbreaks

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Background: Human Parvovirus B19 (B19V) is a common human pathogen causing rash-like symptoms mostly in children but also in the adult population. The small non-enveloped DNA virus can be transmitted by respiratory droplets and blood products. Three distinct genotypes exist with different geographical distribution. Although B19V diagnostics is not a mandatory release-relevant test for blood products since 2013, testing is still performed to release products for the plasma fractionation industry.

Aims: Since 2013, plasma samples of donated blood is tested mandatorily for the presence of B19V DNA. All positive samples were further characterized for B19V IgG and IgM serology, viral load determination and viral DNA sequencing in conjunction with our surveillance program. Epidemiological monitoring of occurring outbreaks allows for close examination of B19V viruses present in the general population.

Methods: From 2013 to 2023, blood donations from six regional Swiss blood donation services were tested in pools of 480 samples for the presence of B19V DNA with an in-house PCR test. Since 2023, pools of 96 samples are being analysed with the Cobas DPx test (Roche Diagnostics). Positive pools were resolved by pool deconstruction and single sample testing. Single samples are considered positive if the titre exceeds 10⁴ IU/mL. The presence of B19V IgG/IgM in plasma was detected in immunoassays. For genotype identification, an 880 bp DNA sequence encompassing regions of VP1 and NS1 was amplified by an in-house heminested PCR and analysed by Sanger sequencing.

Results: From 2013 until 2023, a total of 1.678 Mio blood donations were tested for the presence of B19V DNA, of which 119 samples

tested positive. In this 10-year timespan, the incidence rate fluctuated between 0 and 17 cases per 100,000 donations, with minor B19V outbreaks in the years 2013 and 2017. A steep increase in B19V incidence was recorded in late 2023, representing an ongoing B19V outbreak. Serological analyses showed that most B19V-positive donors were IgG/IgM negative, indicative of an early stage of infection. The age of B19V-positive donors peaked around the age of 40. B19V DNA sequence analyses unveiled that genotype 1a1 and 1a2 are the most prevalent in Switzerland, with rare appearances of genotype 2.

Summary / Conclusions: Surveillance of blood donors detected a strong B19V outbreak at the end of 2023, which has continued in 2024. Further data of this ongoing outbreak will be presented. The absence of cases in the preceding years (2021/2022) was most probably due to COVID-19 protective measures (e.g. masks, hand sanitation, social distancing) and may have led to a stronger incidence in the current outbreak.

P430 | Hepatitis E virus seroprevalence in Dutch blood donors, 1988-2023

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Background: In the Netherlands, since 2013, all donations collected for the manufacture of solvent detergent (S/D) plasma are screened for the presence of hepatitis E virus (HEV) RNA. In addition, since 2017, all donations for the manufacture of labile blood products are also screened. The incidence of HEV infection over the years is unstable. To study the infection pressure of HEV over the years we determined the presence of HEV antibodies in Dutch blood donor samples collected in 1988, 2000, 2011, and 2023.

Aims: We studied historical and recent trends of HEV seroprevalence in Dutch blood donors, thus providing a more complete context for policy decisions regarding transfusion safety and the screening of donors for HEV.

Methods: Samples were selected to include an equal number of donors per 10-year age cohort and sex. Samples were tested using an anti-HEV IgG assay (Beijing Wantai Biological Pharmacy Enterprise Co., Ltd) according to the manufacturer's instructions. Samples with a signal-to-cutoff >0.5 were retested. We compared our 2023 findings with previous serosurveillance studies performed on donor samples collected in 1988, 2000, and 2011.

Results: Of 2181 tested samples from 2023, 534 (24.5%) tested positive for anti-HEV. Seroprevalence increased with age and was higher in men. Compared to our previous serosurveillance studies, seroprevalence increased in donors aged ≤40 since 2000 (Table 1). In donors aged >50 a decreasing seroprevalence occurs since 1988. Table 2 presents seroprevalences by year of birth, and shows a decreasing

P430 - Table 1. Anti-HEV seroprevalence (%) by age group.

Age	1988	2000	2011	2023
18-30	20.9	6.3	12.8	15.3
31-40	34.8	11.9	13.0	21.0
41-50	54.9	33.3	20.3	20.8
51-60	71.1	50.9	32.9	20.5
61-70	76.1	59.4	42.6	31.2
<70	-	-	-	38.6
Total	46.3	29.5	26.8	24.5

P430 - Table 2. Anti-HEV seroprevalence (%) by year of birth

Birth cohort	1988	2000	2011	2023
1921-1930	70.9	44.4	-	-
1931-1940	65.5	63.4	-	-
1941-1950	52.3	44.1	42.1	37.3
1951-1960	33.6	29.9	31.9	33.9
1961-1970	16.1	7.4	20.3	23.4
1971-1980	-	7.3	12.5	22.6
1981-1990	-	9.1	12.8	20.6
1991-2000	-	-	13.4	18.2
2001-2010	-	-	-	9.1

seroprevalence over time in older cohorts. In the 1951-1960 and younger cohorts, seroprevalence increased in the samples collected since 2000.

Summary / Conclusions: An age-cohort effect in donors over 50 years could explain their continued decreasing HEV-seroprevalence, which initially may have been caused by a very high infection pressure from HEV or an HEV-like agent several decades ago. Subsequently, HEV may have re-appeared more recently, causing an increasing seroprevalence in non-immune younger donors.

P431 | WNV infection in Croatian blood donors—results of the six years of ID-NAT screening

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Background: West Nile Virus (WNV) is a neurotropic human pathogen that can cause West Nile fever (WNF) and West Nile neuroinvasive disease (WNND) in immunodeficient and elderly patients. Since the WNV infection is mainly asymptomatic, infected blood donors could donate blood which could cause severe disease in recipients. Nucleic acid testing (NAT) of blood donors for WNV was implemented in Croatia in the summer of 2018, by the decree of the Croatian Ministry of Health due to the WNV outbreak. Since then, individual donation

P431 - Table 1 Results of WNV RNA screening and confirmatory tests for WNV RNA-positive blood donors and donor-related data.

Blood donor	Age	Procleix WNV test s/co	WNV RNA Ct ^a	Post-donation symptoms
1	53	12.11	31.07	rash, fever, weakness
2	50	13.20	29.20	fever
3	20	9.36	NEG	headache
4	36	15.48	28.00	headache, muscle pain, fever
5	25	13.25	30.50	none
6	24	14.62	29.00	none
7	45	13.77	28.62	none
8	43	14.16	22.50	fatigue, headache, muscle pain, diarrhoea
9	61	12.94	26.05	somnolence

^a Ct values were obtained with Real Star WNV RT-PCR Kit 2.0 Altona Diagnostics.

nucleic acid testing (ID-NAT) for WNV RNA has been conducted seasonally for all blood donations in Croatia.

Aims: We aimed to present the results of WNV NAT screening in Croatia for the period from 2018 to 2023.

Methods: A total of 482,390 blood donations were screened for WNV RNA during mosquito seasons, in the period between 2018 to 2023. Blood donation samples were tested on Panther instruments with Procleix WNV assay (Grifols, Spain) in ID-NAT format. This assay detects WNV L1 and L2, and the Usutu (USUV) virus, which cross-reacts with WNV. All initially reactive (IR) NAT samples were retested in duplicate and IR donations were rejected. If the donation was found repeatedly reactive (RR), the blood donor (BD) was deferred from donating blood for 120 days and all his accessible donations in the past period of 120 days were retracted. For WNV RNA confirmation the Real Star WNV RT-PCR Kit 2.0 assay (Altona Diagnostics, Germany) was used. This test detects WNV L1 and L2 and does not cross-react with USUV.

Results: Out of 482,390 tested blood donations, nine were WNV NAT-positive (1:53,599 donations; 95% CI – 1:117,217 to 1:28,235, 0.0019%). The overall specificity of testing was 100%. Eight RR ID-NAT WNV RNA positive donations were confirmed with RT-PCR, while one sample was negative possibly due to low viral titre or the USUV infection which we were not able to confirm. All WNV-infected blood donors were asymptomatic at the time of the donation, but six developed symptoms subsequently. All the infections were autochthonous. Table 1 sums screening and confirmation test results and donor-related data.

Summary / Conclusions: The results of testing for WNV RNA in Croatian blood donors prove the presence of the WNV in the healthy population in Croatia. Persistent WNV circulation suggests that WNV is endemic in Croatia. The implementation of seasonal ID-NAT screening for the WNV RNA has additionally improved blood safety in Croatia.

P432 | Travel and sexual risk of Zika for cord blood donor—a risk-based approach

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Background: Zika virus can be transmitted through blood transfusions, organ or hematopoietic cells transplantation. Infected pregnant women may represent a risk for cord blood transplantation. Zika virus through mosquito bites in endemic areas or through sexual contact with an infected partner. They may subsequently donate a Zika-positive cord blood donation upon the birth of their child, and risk transmission to an immunocompromised recipient. To mitigate the risk of transmission, cord blood banks typically screen potential donors for risk factors, such as recent travel to Zika-endemic areas or potential exposure to the virus, and at-risk donors may be deferred. At our blood service, cord blood units are discarded if the donor's mother or father has traveled to a Zika-endemic area during the pregnancy. However, the discard rate has strongly increased, reaching up to 24% since the end of the COVID-19 pandemic and the return of international travels.

Aims: Given the waning Zika epidemic and the increasing discard rate for cord blood donations, we aimed to estimate the risk of collecting a cord blood unit infected with Zika virus among female donors (and their partner) who travelled to an endemic area while pregnant.

Methods: The model considered the following parameters (each with specified values and ranges): risk of travel exposure in a Zika-endemic country, duration of travel, daily risk of acquiring Zika in an endemic region, probability of Zika transmission to the fetus, probability of asymptomatic viremia in the fetus and probability of transmission by sexual contact. One million Monte Carlo simulations were run, and the mean and 95% confidence intervals (CIs) were evaluated.

Results: In the most-likely scenario (probability of traveling to a Zika-endemic area while pregnant = 0.089), the risk was estimated at 1.8 Zika-positive donations per 1M donations (95% = 0.5–3.6) – or one every 389.8 years at our blood service. In the pessimistic model (which considered that 17.8% of donors travelled outside of Canada, the United States, and Europe), the risk was estimated at 3.6 Zika-positive donations per 1M donations (95% CI 1.3–6.2) – or one every 194.9 years.

Summary / Conclusions: The current criteria for discarding cord blood donations due to travel in Zika-endemic areas may be reconsidered without compromising safety, thus potentially expanding the cord blood supply.

P433 | WNV and blood donors of the province of Brescia during the summer-autumn season 2019–2023

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Background: West Nile Virus (WNV) is a RNA Flavivirus transmitted to humans by Culex mosquito. The interhuman transmission can occur through placenta or blood and HSCs transfusion. Usually the infection is asymptomatic but the virus can cause severe neurological symptoms in immunocompromised patients. Since 2019, during the summer-autumn season, after a positive trigger event, all the whole blood, blood components and HSCs donations collected in Brescia and in its province are tested for WNV-RNA at the NAT Laboratory of the ASST Spedali Civili, Brescia using an ID (individual donor) real-time PCR screening test.

Aims: Evaluating the role of WNV-NAT screening in the detection of asymptomatic infections in blood donors.

Methods: 107,769 whole blood, blood components and HSCs units were screened by an ID qualitative test for WNV-RNA using the Cobas 6800. According to the Italian law, all initially reactive samples (IR) were retested in duplicate on the same tube within 24 hours. Given a repeat reactive (RR) result, new samples were tested by ID NAT and both RT-PCR WNV-Panflavivirus and IgG and IgM test were performed at Fondazione IRCCS Policlinico San Matteo, Pavia. Confirmed reactive donors were notified to the Regional SRC and to the local ASST; new samples were collected after one month as follow-up.

Results: Out of 107.769 donations tested, 29 were IR: 27 were confirmed RR when retested in duplicate. Confirmatory/follow up test: 25 repeated donors were WNV positive: 19 NAT + (5 also RT-PCR+; 5 also IgM+; 1 RT-PCR+ and IgG+), 5 only IgM+; in 1 donor IgM were detectable after one month. All donors were IgG positive after one month. USUTU Virus (USUV) was detected in a male donor (RT-PCR USUTU RNA+); after one month both IgM and IgG were also detectable. A coinfection WNV-USUV was detected in a female donor, RT-PCR was negative, while WNV and USUV IgM were positive; after one month WNV IgG and IgM and USUV IgG were detected. Most donors (age range 28–66 years old) were asymptomatic; a male donor, aged 58, referred headache and fever ten days after the donation and he went to the ER, while another male donor, aged 42, reported a febrile episode. The female donor (64 years old) WNV-USUV coinfecting had a diarrhea episode.

Summary / Conclusions: Our results highlight the importance of an integrated surveillance of the circulation of WNV on the Italian territory in order to reduce the transfusion transmitted risk and to avoid blood shortages. By using the WNV NAT screening, the NAT Laboratory of the ASST Spedali Civili, Brescia identified, in five years, during the summer-autumn period, 27 donations WNV and/or USUV positive. The introduction of WNV NAT screening avoided the possible transmission of WNV to frail patients: indeed all the donors were eligible to give blood and had a negative medical history. Finally, the virus circulates only in areas of the province of Brescia with a higher humidity rate

and higher temperature: no donors living in the north of our province, rich in mountains and valleys, had a positive screening test.

P434 | Prevalence of HEV in Macedonian blood donors

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Background: Our understanding of the molecular virology and pathogenesis of hepatitis E virus (HEV) is still incomplete but there is a body of evidence concerning the health consequences of HEV infection via blood transfusion in pregnant women and immunocompromised recipients.

Aims: To estimate HEV reactivity rate in blood donors and donations and to evaluate the impact of HEV screening on blood safety.

Methods: In 2023, routine ID-NAT screening of HEV was introduced in the Institute for transfusion medicine of North Macedonia using Procleix UltrioPlex E transcription-mediated amplification assay. The total of 56450 blood donations from 36670 donors were screened for the presence of HEV RNA. Initially reactive (IR) samples were retested in duplicate by the same method. Repeatedly reactive (RR) samples were subjected to anti-HEV IgM and IgG serology testing using Vidas anti-HEV IgM and IgG assays. All of the NAT HEV RR donors were recalled for a new blood sample to repeat HEV testing.

Results: Out of 56,450 blood donations, 50 (0.09%) were initially NAT HEV reactive and 30 (60%) of them were repeatedly reactive. The prevalence of HEV RNA in donors and in donations was 0.081% and 0.053% respectively. One in 2016 donations was found to be viremic. HEV prevalence in first time versus repeat donors was 0.057% and 0.090% respectively. According to the sex, there was 1 (3.33%) female and 29 (96.6%) male NAT HEV RR donors. IgG and IgM anti-HEV antibodies were present in 5 (16.6%) out of the 30 NAT HEV RR donors. We obtained a new blood sample from 12 donors in a period from 3 to 10 months after the first NAT HEV RR donation. All of them were anti-HEV IgG positive and 2 donors were also IgM positive. Only 2 (16.6%) of recalled donors were still HEV NAT positive and have IgG antibodies.

Summary / Conclusions: The estimated prevalence of HEV RNA is much higher than the prevalence of HCV and HIV RNA in Macedonian blood donors which justifies the implementation of HEV testing. So far, we consider serology testing very useful for conclusion that NAT HEV RR donations are window period donations rather than false positive/nonspecific reactions. However, further testing is necessary for the decision to be made if previously HEV RNA positive donors who become HEV RNA negative can donate blood and how long after they have no longer detectable HEV RNA.

P435 | Dynamics of SARS-CoV-2 immune response in Chinese blood donors - insights from inactivated COVID-19 booster vaccination and BA.5.2/BF.7 breakthrough infection

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Background: SARS-CoV-2 infection has imposed a global health burden, prompting widespread vaccination efforts. To combat the evolving landscape, China started booster dose vaccination in August 2021. With the emergence of the Omicron variant, China modified the dynamic clearance policy in December 2022. In this study, we evaluate the dynamic changes in immunity among Chinese blood donors during this pivotal period.

Aims: To evaluate the evolving immune responses in blood donors post-inactivated COVID-19 booster vaccination and subsequent breakthrough infections.

Methods: Blood donors were recruited from six blood stations in five geographic divisions of China, Dalian (Northeast), Kunming (Southwest), Zhengzhou (Central-South), Sanmenxia (Central-South), Nanjing (East China), and Taiyuan (North China). Peripheral blood was collected pre-booster vaccination, post-vaccination (1 and 3 months), and post-infection (1 month). SARS-CoV-2 antibody levels (S-IgG, N-IgG, total antibody, neutralizing antibody) and SARS-CoV-2 specific IFN γ were assessed at each time point using kits from Wantai Bio-Pharm (Xiamen, China). 15 samples were selected and levels of antibodies against WT, BA.5, BF.7, and XBB.1.5 strains at each time point were detected using the lentiviral based pseudovirus neutralization assay.

Results: A total of 134 blood donors were recruited, including 82 males and 52 females. The median age was 39 (33.5, 47.0) years.

The interval between the second and booster vaccinations was 223 (205.3, 253.8) days. The levels of S-IgG, total antibody, neutralizing antibody, N-IgG, SARS-CoV-2 specific IFN γ , and neutralization titer of wild type, BA.5, BF.7 and XBB.1.5 strains at the time points before booster vaccination, 1 month, 3 month after booster vaccination, and 1 month post-BA.5.2/BF.7 infection were shown in Table 1 below. Significant increases in these immune response indicators were observed post-booster vaccination and infection.

Summary / Conclusions: In Chinese blood donors, inactivated COVID-19 booster vaccinations led to a substantial increase in SARS-CoV-2 antibodies and T-cell immunity. Furthermore, breakthrough infections amplified the specific immune response. These findings provide crucial insights into the evolving dynamics of SARS-CoV-2 immunity in the context of booster vaccinations and breakthrough infections.

P435 - Table 1: Dynamics of SARS-CoV-2 immune response during inactivated COVID-19 booster vaccination and BA.5.2/BF.7 breakthrough infection.

	B-0M [#]	B-1M	B-3M	I-1M [#]
S-IgG (IU/mL)	9.65 (5.08, 20.08)	205.79 (113.77, 382.68)	97.68 (42.52, 219.79)	1488.87 (784.16, 2452.44)
TA γ (COI)	11.63 (3.92, 31.78)	659.40 (386.15, 885.29)	401.71 (127.56, 665.36)	867.07 (786.88, 907.03)
NA γ (IU/mL)	9.88 (4.59, 20.82)	594.28 (153.28, 1652.00)	193.34 (59.84, 738.27)	3396.89 (1209.34, 12765.83)
N-IgG (S/CO)	0.14 (0.05, 0.68)	6.52 (2.35, 10.32)	3.81 (1.25, 7.25)	255.11 (201.27, 302.33)
IFN γ (pg/mL)	33.82 (6.36, 95.01)	154.34 (32.56, 415.87)	112.21 (32.29, 292.64)	315.17 (133.90, 590.18)
NT50-WT	46.01 (10.00, 179.02)	213.93 (160.00, 456.98)	92.03 (38.00, 190.94)	1978.24 (1160.07, 3258.52)
NT50-BA.5	<10	55.99 (23.00, 152.96)	14.00 (10.00, 60.00)	978.2 (472.12, 2091.03)
NT50-BF.7	<10	30.00 (13.00, 42.99)	<10	1663.49 (466.91, 3590.57)
NT50-XBB.1.5	<10	16.00 (10.00, 31.00)	<10	90.02 (50.01, 250.04)

[#]"B" represents "booster vaccination", "I" represents "infection".

P436 | Recent SARS-CoV-2 infection in Dutch blood donorsF Quee^{1,2,3}, B M Hogema^{1,4}, K Van den Hurk^{1,2,3}, E Slot⁵, H L Zaaier¹¹Donor Medicine Research, Sanquin Research, ²Public and Occupational Health, Amsterdam UMC, ³Amsterdam Public Health Research Institute, Amsterdam UMC, ⁴Virology, Sanquin Diagnostic Services, ⁵Medical Affairs, Sanquin Corporate Staff, Amsterdam, Netherlands

Background: After cessation of nationwide SARS-CoV-2 PCR testing of symptomatic individuals, surveillance of SARS-CoV-2 circulation is mainly based on serosurveillance studies with a cross-sectional design. After more than three years of SARS-CoV-2 circulation, most individuals in the Netherlands acquired antibodies, and cross-sectional studies become less informative. However, longitudinal serosurveillance studies provide insight into antibody waning and can be used to estimate the incidence of new and reinfections. Blood donors are excellent study subjects for such longitudinal studies, as they donate regularly and provide consent to use serial, archived blood samples for scientific studies.

Aims: In this study we quantify the proportion of blood donors, without prior SARS-CoV-2 infection, that acquired SARS-CoV-2 infection since August 2022.

Methods: Based on data collected in previous SARS-CoV-2 serosurveillance studies, we created a cohort of donors who tested negative for anti-nucleocapsid (NC) antibodies between August 2022 and March 2023. If these donors donated blood in the data collection period, between October 1st and December 3rd 2023, their donation was again tested for anti-NC to see whether they had seroconverted since their last donation, using an assay that does not suffer from limited sensitivity regarding the persistence of NC-antibodies.

Results: Of the 1206 donors whose donation(s) tested negative for between August 2022 and March 2023, 560 (46%) donated in the data collection period. Older donors were overrepresented in the study. Of the included donors, 78% seroconverted for anti-NC between August 2022 and December 2023. Over time, SARS-CoV-2 infection pressure in the study population decreased; between March and December 2023, 38% of the studied donors seroconverted for NC-antibodies.

Summary / Conclusions: The majority of studied Dutch donors without prior SARS-CoV-2 infection before August 2022 seroconverted for SARS-CoV-2 NC-antibodies between August 2022 and October 2023. Fortunately, this was not accompanied with health care systems overflowing with severe COVID-19 patients. Hopefully the resulting high proportion of older persons with natural infection aids to vaccine-induced protection of vulnerable persons against severe COVID-19, when new strains of SARS-CoV-2 arise.

P437 | Dengue, zika and Chikungunya virus in Chinese blood donors—establishment of quality control system and two-year NAT screening resultsY Yan¹, L Chang¹, H Ji¹, H Sun¹, L Wang¹¹National Center for Clinical Laboratories, Beijing hospital, Beijing, China

Background: To assess the prevalence of transfusion-transmitted viruses, and enhance screening capabilities for emerging and re-emerging viruses, China has been implementing nucleic acid test (NAT) for Dengue, Zika and Chikungunya viruses at eight border central blood stations in Yunnan and Guangxi provinces. To ensure testing quality, the National Center for Clinical Laboratories facilitated the development of quality control materials, evaluated the performance of NAT screening reagents, conducted external quality assessment (EQA) for the eight blood stations, and confirmed screening-reactive samples.

Aims: To establish the quality control system for DENV/ZIKV/CHIKV NAT screening, and to explore the prevalence of these viruses in Chinese blood donors.

Methods: 1. Quality control system establishment: Genome sequences of DENV, ZIKV, and CHIKV that covering the amplified regions of all NAT screening reagents used in China were synthesized and inserted into PQCXIG plasmid, and Moloney Murine Leukemia Virus (MMLV) based pseudoviruses were produced. Digital PCR was conducted to quantify the pseudoviruses. Quality control materials were prepared by diluting pseudoviruses in negative plasma to certain concentrations. Using the quality control materials, LoD and precision of six reagents (KHB, Wanti, Xinbo, Sansure, Bacme and Daan) were evaluated in accordance with GLSI EP17-A2, EP15-A3 and CNAS-GL039 guidelines. EQA consisting ten samples was carried out annually for the eight border central blood stations. 2. Positive sample confirmation and sequencing: Reactive samples in screening test were confirmed using another four NAT reagents. Serological tests for antigen (Wondfo BIOTECH CO., LTD, Guangzhou, China) and antibody (Beijing Wantai Biological Pharmacy Enterprise CO.,LTD, Beijing, China) were also performed. Confirmed positive samples were further sequenced for typing.

Results: 1. Establishment of quality control system: Of the six reagents for DENV/ZIKV/CHIKV NAT screening, Xinbo achieved the lowest LoDs individual donor (ID)-NAT test, with 11.23 copies/mL for DENV, 11.79 copies/mL for ZIKV, and 12.84 copies/mL for CHIKV. In minipool (MP)-NAT test, Xinbo got the lowest LoDs for CHIKV at 26.84 copies/mL, and DENV at 98.41 copies/mL. While KHB achieved the lowest LoDs for ZIKV at 93.72 copies/mL. All kits showed good precision, with CV values less than 5% at 2LoD. EQA scores for all eight blood stations exceeded 80 over two years. 2. two-year NAT screening results: 45383 blood samples were screened in 2022, with no reactive samples were detected. 44972 samples were screened in 2023, and nine Dengue-reactive samples were identified in Xishuangbanna blood stations. Six of the samples were confirmed positive. All the nine samples were negative for DENV IgG. Two samples were positive for NS1 antigen. And two samples with high viral loads were sequenced for DENV type 1.

Summary / Conclusions: This study presents the successful establishment of a robust quality control system for the nucleic acid test (NAT)

screening of DENV/ZIKV/CHIKV viruses in Chinese blood donors. The screening identified Dengue-positive blood donor samples in Yunnan province, with a prevalence rate of 1.33‰, emphasizing the importance of ongoing surveillance and control measures in blood safety.

P438 | A negligible risk of transfusion-transmitted SARS-CoV-2 in populations with high immunization coverage during the first Omicron surge in Taiwan

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Background: SARS-CoV-2 viral RNA was detected in the blood of 1-8.7% of COVID-19 cases before late 2021, and the detection rate decreased after a mass vaccination program. No transfusion-transmitted infection was confirmed. In 2022, SARS-CoV-2 Omicron surge affected 8.8 million individuals in Taiwan, and containment measures on transfusion safety were implemented. By the end of August 2022, 87% of the general population had been fully immunized.

Aims: To evaluate the residual impact of containment measures on transfusion safety in highly immunized populations during 2022 Omicron surge, we conducted an analysis using post-donation information (PDI) data and test results of the repository specimens collected from COVID-19 donors.

Methods: Blood donations collected during 2020-2022 were included. Blood donors in Taiwan could provide PDI through callback reporting if they experienced any blood safety-related issues within two weeks of donating blood. PDI and donation details were retrieved to assess the incidence of various PDI, including COVID-19 identification, COVID-19 close contacts, cold-like symptoms, etc. The chi-square test and logistic regression were used to assess main effects of PDI reporting. During the first Omicron outbreak surge from April to August 2022, an investigation of COVID-19 donors identified through PDI was prompted. We collected these donors' disease information, and their repository specimens collected during blood donations were tested for SARS-CoV-2 RNA and antibodies against spike protein and nucleocapsid protein.

Results: A total of 9435 donations reported PDI events to blood centers in 2020-2022. The incidence increased from 10.5 per 10,000 donations in 2020-2021 to 29.9 per 10,000 in 2022. Nearly 70% of PDI reported in 2022 were related to COVID-19. The incidence of PDI due to COVID-19 identification and COVID-19 close contacts increased significantly, from 0.02 to 18.6 per 10,000 donations ($p < 0.0001$) and from 0.4 to 2.2 per 10,000 donations ($p < 0.0001$), respectively. The incidence of PDI with cold-like symptoms dropped significantly from 5.5 per 10,000 to 4.7 per 10,000. Female donors reported more PDI related to COVID-19 identification, close contact with COVID-19, and cold-like symptoms. Young donors reported more PDI in all categories, with incidence decreasing with age.

Geographic differences were observed. A total of 1148 COVID-19 donors were investigated. Approximately 80% of these cases reported to be diagnosed with COVID-19 within 3 days after their blood donations. Among them, 67.7% confirmed their COVID-19 through rapid antigen tests, and 32.3% through PCR tests. SARS-CoV-2 RNA was not detected in their repository serum specimens, and the seroprevalence of anti-nucleocapsid(N) and anti-spike(S) antibodies was 0.61% and 98.4%, respectively. This result also indicated that the majority of these COVID-19 donors developed antibodies through vaccination.

Summary / Conclusions: The risk of transfusion-transmitted SARS-CoV-2 infection could be negligible in populations with high immunization coverage during Omicron surge. Current measurements are sufficient to ensure blood safety without sacrificing blood adequacy.

P439 | Abstract withdrawn

P440 | First confirmed case of transfusion-transmitted hepatitis E in a thalassaemia patient in Pakistan

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Background: Hepatitis E Virus (HEV) is considered an emerging threat to blood safety. Since 2004, there have been 86 cases of HEV transmission by blood transfusions. Even at relatively low blood levels of the virus, HEV can be transmitted. There is ongoing discussion on the significance of universal HEV screening for blood donations.

Aims: To investigate the post-transfusion transmission of HEV in a thalassaemia patient.

Methods: On October 20, 2023, a single unit of red cell concentrate (RCC) was administered to a thalassaemia major patient (aged 14 years) admitted to the Thalassaemia Centre of the Divisional Headquarters Teaching Hospital, Mirpur, AJK, Pakistan. The transfused RCC had passed all mandatory blood screening tests at the Mirpur Regional Blood Centre. On December 5, 2023, routine blood testing of the patient revealed deranged liver function tests, i.e., alanine aminotransferase (302 U/L; reference interval, < 40 U/L), aspartate aminotransferase (79 U/L; reference interval, < 35 U/L), and bilirubin (2.4 mg/dl; reference interval, < 1.0 mg/dl). The patient was tested negative for Hepatitis B (HBsAg) and Hepatitis C (anti-HCV) through chemiluminescence immunoassay (CLIA). Follow-up of the donor and patient for HEV was carried out by means of serological and molecular assays. For RT-PCR, the RNA extraction and cDNA synthesis were done using commercial kits. The HEV ORF2 (Open Reading Frame-2, capsid protein) was amplified using nested PCR targeting a 348-bp segment. Informed consent was obtained from the patient's father concerning

the presentation of the case. The study protocol followed the ethical guidelines of the Helsinki Declaration of 2013.

Results: The transfused patient had developed typical symptoms, including abdominal pain, reduced appetite, and joint pain. HEV RNA was detected in the patient's sample, the archived sample of the donor's blood, and also in the fresh frozen plasma (FFP) present in the blood bank's inventory. The donor and patient samples were sequenced, analysed, and compared with reference sequences available in the NCBI data bank. Multiple alignment sites with gaps in any of the sequences were omitted. The sequencing data were analysed through Bio-Edit sequence alignment software. Phylogenetic trees were constructed using the neighbor-joining method. The full genome sequences from both the donor and the person who got acute hepatitis E showed that they were both genotype 1 and had a match for 326 nucleotides in ORF-2. Phylogenetic analysis indicated that HEV genotype 1 detected in both samples is closely related to genotype 1 from London, UK (MH504163). The donor, a 37-year-old male living in Mirpur, was followed up by telephone. He had remained asymptomatic but had a history of recent travel abroad (UK). Hence, it was confirmed that the patient contracted HEV by RCC transfusion, despite the fact that blood components with larger plasma quantities, primarily FFP and platelet concentrates, are believed to transmit HEV more easily.

Summary / Conclusions: This is the first report of transfusion-transmitted Hepatitis E in Pakistan. It is imperative to establish an evidence-based screening policy for asymptomatic HEV-positive blood donors to safeguard public health.

P441 | Assessing the West Nile virus transfusion-transmission risk in a blood bank located in a mediterranean archipelago—a blood donor screening study

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Background: West Nile virus (WNV) is one of the most widely distributed flaviviruses worldwide. Birds are their natural hosts and it circulates through bird-mosquito interactions. People bitten by an infected mosquito could become accidental hosts and may become sick. Infection with WNV is asymptomatic in around 80% of cases, but about 20% will develop West Nile fever and approximately 1 in 150 persons will develop neuroinvasive disease. The first known human case in Spain occurred in 2004. There are epidemiological data that supports the suspicion of a potential risk of autochthonous WNV transmission in the Balearic archipelago: autochthonous WNV transmission has recently been detected in Spain's mainland areas located close to the Balearic archipelago; WNV main vectors (*Culex spp.* mosquitoes) have been established in the archipelago since years; the presence of WNV in horses in Majorca have been isolated by seroprevalence studies.

Aims: To assess the WNV transfusion-transmission risk in the Balearic Islands by determining the presence of WNV RNA in blood donors from areas with potential risk of autochthonous transmission.

Methods: Territories from the archipelago where birds are most likely to stop during their migration to Africa were defined in collaboration with the Balearic Ornithological Group (GOB). Since birds are the main reservoir for WNV, these regions were established as the higher risk areas. Blood donors from these areas were selected to participate in the study. Samples were collected in tubes containing EDTA, and centrifuged at 1800 g for 15 min in order to obtain serum. Detection of genetic material of WNV was performed by real time PCR using the cobas WNV assay (Roche Diagnostics GmbH, Germany) in individual samples.

Results: A total of 247 samples were analysed from the previously selected population: 118 from Majorca (9.67% of total donations), 83 from Minorca (24.27% of total donations), and 46 from Ibiza and Formentera (53.49% of total donations). All samples were found to be negative for WNV RNA. Additionally, 141 samples were analysed in our laboratory from donors who have travelled to an autochthonous WNV transmission area outside the Balearic Islands within the 28 days prior to their donation. All samples were also negative for WNV RNA.

Summary / Conclusions: Areas where birds are most likely to stop during their migration and in which mosquitoes of *Culex spp* are present, represent the areas with the higher potential autochthonous WNV transmission risk. Our results show 0% of presence of WNV RNA in the studied sample which suggest a very low autochthonous WNV transmission risk in our region. However blood donors seroconversion analysis would be needed in order to complete this study.

P442 | Asymptomatic West Nile virus prevalence amongst Canadian blood donors compared to clinically reported cases across Canada

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Background: West Nile Virus (WNV) is a member of the Flaviviridae family belonging to the Japanese encephalitic antigenic complex. Although it can cause fever, headache, joint pain, rash, and uncommonly more serious illness, approximately 70%–80% of infections are thought to be asymptomatic. WNV is transmitted by *Culex* genus mosquitoes, which are native to Canada and found in southern Ontario, Quebec, the Maritimes and the Prairies.

Aims: We sought to examine the prevalence of WNV NAT positivity over time among Canadian blood donors and compare the asymptomatic infection rate in blood donors to diagnostically reported clinical cases across Canada.

Methods: All blood donors in Canada (except for Quebec) were tested in mini-pools of 6 by the Roche Cobas WNV NAT assay during summer-fall season (June–Nov) by Canadian Blood Services. Should a mini-pool flag positive, the pool was broken, and each

sample was individually re-tested to identify the positive donation. Positive results during the summer-fall months triggered individual testing of all donations collected within 100 km from site of assessed donor WNV acquisition for 7 days. Travel-related testing was performed in winter-spring season (Dec-May). All donors confirm they are asymptomatic at time of donation. WNV NAT positivity rates in asymptomatic blood donors were compared to clinically diagnosed national WNV cases reported by the Public Health Agency of Canada.

Results: Between 2018 and 2023, all summer-fall season WNV NAT positive donor results were detected Aug-Sept except in 2018 and 2023 (July-Oct and Aug-Oct respectively). No increasing trend of positivity was observed over time. All travel related winter-spring season testing was negative except 3 cases in 2019 which were shown to have had recent vaccination against Japanese Encephalitis Virus (JEV). During the study period, the highest rate of WNV in blood donors was summer-fall 2018 where 9.95 per 100,000 donations tested positive by WNV NAT. Similarly, the highest rate of clinical WNV cases was in 2018, with 437 reported across Canada (236 excluding QB). The lowest rate of WNV NAT positives was observed for blood donors in 2019 (0.25 per 100,000 donations), and the lowest rate of clinical cases in 2019 and 2021 (31 cases excluding QB; 0.1 per 100,000). While the overall trend of WNV NAT positives remains similar among blood donors and diagnostically identified cases in Canada (higher asymptomatic detection tracks with higher numbers of reported clinical cases), the rate of asymptomatic infection is higher than expected in blood donors. Using an 80% asymptomatic infection rate for WNV, we would estimate a 1.48 per 100,000 blood donor positivity rate based on the reported clinical infections for 2018, rather than the observed 9.95 per 100,000.

Summary / Conclusions: In this surveillance period, WNV positivity was sporadic, and the number of clinical cases directly tracked with the number of infections detected in the asymptomatic donor population. The asymptomatic rate was higher than expected in the blood donor population, likely suggesting that WNV clinical cases are under-reported nationally. Finally, false positives were noted from donors who recently received JEV vaccination. Canadian Blood Services has implemented a 3-week temporary donor deferral after JEV vaccination, to minimize risk of vaccine confounding WNV NAT results; blood operators may also consider additional testing following WNV NAT positive screening results to confirm WNV infection.

P443 | Trends in hepatitis E incidence among blood donors in Eastern Austria—a retrospective analysis (2015-2023)

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Background: Hepatitis E virus (HEV) infection poses an emerging threat worldwide, primarily transmitted through the consumption of contaminated food or water. In industrialized countries, including

P443 - Table 1

Year	HEV+ Donations	Total Donations Tested	Incidence (%)	p-value
2015	2	20168	0.01	0.045
2016	18	92191	0.02	0.023
2017	23	100717	0.02	0.016
2018	31	102922	0.03	0.008
2019	21	96302	0.02	0.011
2020	10	98303	0.01	0.039
2021	14	110185	0.01	0.034
2022	14	144157	0.01	0.027
2023	32	169467	0.02	0.006

Austria, zoonotic transmission from infected animals to humans has also been reported. Understanding the prevalence of HEV among blood donors is crucial for ensuring blood safety, given that transfusion transmitted HEV cases have been documented (Hewitt et al., 2014). Austria primarily encounters HEV genotype 3, which is known for its zoonotic transmission (Smith et al., 2016). This study aims to analyse the incidence trends of Hepatitis E among blood donors in Eastern Austria over a nine-year period.

Aims: This study aims to investigate the trends in Hepatitis E incidence among blood donors in Eastern Austria over a nine-year period and to assess whether there are significant associations or trends in Hepatitis E incidence year-on-year.

Methods: A retrospective analysis of data from blood donations in Vienna, Lower Austria, and Burgenland from 2015 to 2023 was conducted. Records of HEV-positive and HEV-negative donations were examined to determine the incidence rates of Hepatitis E among blood donors in the region.

Results: Hepatitis E incidence among blood donors in Eastern Austria exhibited an undulating pattern from 2015 to 2023. The table below summarizes the results, including the p-values for each year based on the Chi-square test for trend. Statistical analysis using the Chi-square test for trend indicated significant associations in Hepatitis E incidence over the study period ($p < 0.05$) for year-on-year changes.

Summary / Conclusions: The findings indicate an undulating incidence of Hepatitis E among blood donors in Eastern Austria from 2015 to 2023. This underscores the importance of continuous surveillance and implementation of appropriate measures to ensure blood safety. Despite the rise and fall of incidence, measures must be taken to ensure availability of Hepatitis E negative blood products for at-risk populations, including immune-compromised, organ transplant, and pregnant patients. Further research is warranted to explore underlying factors contributing to the modest elevation in prevalence of Hepatitis E in this population and to develop comprehensive strategies for testing, education, and prevention.

P444 | Parvovirus B19 but not hepatitis A nucleic acid test positivity rates in Canadian plasma donors are still reduced in the pandemic recovery period

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Background: Plasma sent for fractionation from Canadian Blood Services to Grifols is tested for both parvovirus B19 (B19V) and hepatitis A virus (HAV) using a nucleic acid test (NAT). A previous study suggested that the percentage of B19V and HAV positive donations were significantly reduced from the pre-COVID-19 restriction era (2015 - end of February 2020 [Q1]) to the pandemic-impact period (April 1, 2022 - March 31, 2023). Questions have arisen as to whether B19V and HAV NAT positivity rates had increased in the pandemic recovery period (April 1, 2023 - September 30, 2023).

Aims: This study compared B19V and HAV NAT positivity rates between a pre-pandemic period, a pandemic impact period, and a pandemic recovery period.

Methods: EDTA plasma specimens were collected by Canadian Blood Services from plasma donors (except Quebec/northern territories) and sent within 24 months of blood draw. Specimens were generally tested within two weeks of receipt by the fractionator. EDTA plasma specimen storage and transport temperatures were -20°C. The Procleix Panther System (Grifols Diagnostic Solutions Inc, San Diego, CA, USA), a transcription-mediated amplification/hybridization protection assay NAT platform, was used to test pooled (N = 16) EDTA plasma specimens. Pools that generated a reactive signal for either B19V or HAV were resolved by individual

specimen testing. The quantitative range of the B19V NAT is 500 to 100,000 IU/mL. The test algorithm is as follows: B19V NAT positive (reactive $\geq 2,000$ IU/mL) pools and a signal to cutoff (S/CO) ≥ 1.0 for HAV pools were resolved by individual specimen testing. For B19V, an individual sample results > 500 IU/mL was considered reactive. For HAV, a S/CO ≥ 1.0 was considered reactive. All data was stored with Excel (Microsoft, Seattle, WA, USA). For statistical analyses, the following were undertaken. Descriptive statistics (sum, mean, standard deviation [SD]), and contingency tables [e.g., Chi-square analyses] used GraphPad Prism (Version 9.5.1, GraphPad Software, Boston, MA, USA).

Results: This study analyzed 3,299,883 specimens for B19V and 3,294,218 specimens for HAV using NAT in three time periods (Table 1).

Summary / Conclusions: In the pandemic recovery period, HAV NAT-positivity rates in Canadian plasma donors are rebounding to pre-pandemic levels while B19V positive donations are still reduced. Factors driving these diverging patterns of B19V, and HAV NAT-positivity require further study.

P444 - Table 1. HAV and B19V positivity in the pre-pandemic, pandemic impact, and pandemic recovery periods.

Period	Specimens tested for B19V, n	Specimens positive for B19V, n (%)	Specimens tested for HAV, n	Specimens tested for HAV, n
Pre-pandemic (January 1, 2014 - March 31, 2020)	2,412,701	240 (0.01)	2,407,036	26 (0.001)
Pandemic impact (April 1, 2022 - March 31, 2023)	589,227	5 (0.0008)	589,227	1 (0.0002)
Pandemic recovery (April 1, 2023 - September 30, 2023)	297,955	5 (0.002)	297,955	2 (0.0007)

There was no difference in B19V positivity in the pandemic recovery vs the pandemic impact period (Fisher's exact test, $p = 0.1$). There was a difference in B19V positivity in the pandemic recovery period compared to pre-pandemic period (Chi-square, $p < 0.0001$).

There was an upward trend for HAV NAT positivity (0.0002% to 0.0007%) in the pandemic recovery period vs the pandemic impact period (Fisher's exact test, $p = 0.3$). There was no difference in HAV positivity in the pandemic recovery period compared to pre-pandemic period (Chi-square, $p = 0.7$).

P445 | The inherent correlation between false positive outcomes using different detection methods in the detection of emerging and reemerging infectious diseases within populations of low prevalence

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Background: During the onset of emerging and re-emerging infectious diseases, the incidence of confirmed infections in regions with low prevalence is typically limited. Given that reagent specificity may not achieve complete accuracy, the occurrence of false positive results is possible during extensive screening efforts. The accurate identification of true infection cases within the screened population is crucial for effective disease containment and prevention, underscoring the importance of appropriately acknowledging and managing positive test results.

Aims: To analyze the results of different methods for reactive samples screened by the enzyme linked immunosorbent assay (ELISA) against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in blood donors.

Methods: From March to April 2020, a total of 8632 blood donors in Shenzhen were screened for SARS-CoV-2 total antibodies (including IgG, IgM, IgA, Tab) in plasma using ELISA, the antibody reactivity samples and their follow up plasma samples (FC), disease control group (DC group) from January to April 2020 were detected using the following methods: 1. ELISA method for detecting IgG, IgM, and (or without detection) TAB; 2. Pseudovirus Neutralizing Antibody Test (pVNT); 3. Western Blot (WB) of SARS-CoV-2 antibody, the negative control group (NC group) from February to April 2020 performed 1. and 3. methods.

Results: Among the 34 total antibody positive samples, 2 were positive for pVNT test, and both the total antibody, IgG and WB in the initial screening and tracking samples were positive. Thereafter, it was determined to be confirmed positive. The other 2 cases were positive for pVNT test, while the samples with positive WB results were in the follow-up stage. The TAB, IgG, and pVNT results did not conform to the dynamic evolution of antibodies, and cannot be determined as confirmed positive.

Summary / Conclusions: The infection status of antibody reactivity samples screened by SARS-CoV-2 ELISA can be judged by the logic of pVNT, WB and the dynamic change of antibody.

P446 | Assessment of complement activation in SARS-CoV-2 (COVID-19) infection with D-dimer greater than 3000 ng/mL FEU

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Background: The complement and coagulation cascade share some active proteases and there is a large amount of evidence showing their interconnection. Enhanced D-dimer is a marker of severity in

COVID19 and the complement's activation has been associated to respiratory failure.

Aims: None.

Objective: To evaluate the activation of the complement system in patients with COVID19 and enhanced D-dimer.

Methods: 50 patients hospitalized with COVID19, confirmed by RT-PCR, and D-dimer greater than 3,000 ng/mL FEU were recruited between February to June 2021. After signing the informed consent form, epidemiological data and blood were collected for clinical evaluation as well as assessment of the vital outcome of hospital admission.

Results: Fifty patients were recruited. The time from the onset of symptoms to the collection of blood samples was 15 days (13.0, 19.0 days), 72% male, aged ~ 62 years (53.8, 68.0), 52% hypertensive, 46% obese, 34% diabetic, 26% heart disease, 10% asthmatic and 6% previous diagnosis of neoplasia. At the time of blood collection, 94% were admitted to the ICU, 64% were on invasive mechanical ventilation, 98% were anticoagulated, 100% were using corticosteroids, 37% were using vasoactive drugs, 6% were on dialysis, 72% had a presumed or confirmed infectious diagnosis in addition to COVID-19. It was observed that 44% of patients had common (CH50) and/or alternative pathways (AH50) out of reference value of test (VRt). Based on this information, the patients were divided into 3 groups according to the CS activation profile: group 1 (G1): normal CH50 and AH50; group 2 (G2): enhanced activation of CH50 and normal AH50; group 3 (G3): impairment of AH50 and CH50 with or without alteration. Regarding the dosage of the complement activation, a statistically significant difference was observed between the median of CH50 ($p < 0.001$), AH50 ($p < 0.001$); C3 ($p = 0.023$) and C4 ($p = 0.041$) among the 3 independent groups. In multiple comparisons test there was a significant difference in CH50 between G1vsG2 ($p < 0.001$); G1vsG3 ($p = 0.005$) and G2vsG3 ($p = 0.002$). Related to AH50 the difference was observed in G1vsG3 ($p < 0.001$), G2vsG3 ($p < 0.001$) and for C3: G2vsG3 ($p = 0.038$). There is also a statistically significant difference between the median of leukocytes ($p < 0.001$) and neutrophils ($p < 0.001$) counting among the 3 independent groups, being higher in the groups with complement activation (G2 and G3) in relation to G1. Lymphopenia was more pronounced in G3. The median level of D-dimer, ferritin and haptoglobin was lower in G2 compared to G1 and G3, while fibrinogen, IL-6 and platelets was higher. Regarding to the clinic, there was a statistically significant difference in relation to the respiratory support among the 3 independent groups ($p = 0.0109$) with 91.7% of patients in G3 requiring invasive mechanical ventilation. As the D-dimer level was increased, venous thrombosis (DVT/TEP) was investigated in all patients and confirmed, by imaging, in 55.6% of G2, 50% of G3 and 32.1% of G1. The mortality rate was highest in G1 (32%), followed by G3 (25%) and G2 (11%).

Summary / Conclusions: Patients with D-dimer higher than 3000 ng/mL FEU presented alterations complement activation (44%) that was related to an "inflammatory pattern" in G2 and a "consumption of complement" in G3. Our work suggest that monitoring the complement activation during COVID infection would contribute to guide more effective therapies.

P447 | Acute hepatitis E transmitted by transfusion of packed red blood cells

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Background: Present a case of acute hepatitis E after a transfusion of packed red blood cells, proven phylogenetically, in an immunocompromised patient.

Aims: To present the experience of the Blood, Tissues, Cells Transfusion Center

Methods: A 75-year-old patient with a bladder carcinoma history, with a partial response and multiple transfusions was diagnosed with acute hepatitis. He had received a total of 10 RBC in the 6 months prior to the date of diagnosis. Among other diagnostic studies, such as HEV IgG and IgM serology were performed, with positive results for both Igs. There are no previous studies on the HEV recipient. The patient progressed adequately during diagnosis and was discharged few weeks later. We have no record of the treatments administered. The donor involved was located and a clinical interview was carried out without reporting any symptoms. An HEV study was performed on a new donor sample, obtaining the following results: IgG positive, IgM negative, PCR-HEV negative. The plasma library sample from the donation prior to the donation in question was analyzed resulting in non-reactive results, both serological and HEV-PCR. A phylogenetic study was carried out at the Blood and Tissue Bank of Barcelona with the following results: Amplification of a region of HEV ORF2 (RT-PCR, Nested PCR, Agarose gel, Gel amplicon purification and sequencing). To perform the phylogenetic analysis, the HEV reference sequences for all viral genotypes and additional sequences from GenBank, all named as "accession number", were selected. All the analysis were performed using the Neighbor Joining method in Mega 11 Software. Phylogenetic study result: Positive.

Results: The presence and coincidence of the hepatitis E virus genome in the donation and the recipient is confirmed.

Summary / Conclusions: Transfusion transmission of HEV is described with increasing frequency. More epidemiological studies are necessary in our area, to support the need to screen for this virus in blood donations.

Immunohaematology—red cell immunohaematology: serology

P448 | Abstract withdrawn

P449 | Immunohaematology laboratory workup for non-ABO Incompatible HSCT Challenges

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Background: Immunization against red blood cell (RBC) antigens represents an important cause of hemolysis following blood group-mismatched hematopoietic stem cell transplantation (HSCT). The resultant anemia, having an auto- or alloimmune origin, demands in both scenarios prompt diagnosis and directed management. Non-ABO incompatibility is less frequently encountered but can have similar complications to ABO incompatibility, leading to clinically significant hemolysis and adverse clinical outcomes. RBC alloimmunization resulting from non-ABO mismatch could originate from the donor, recipient, or both, depending on the temporal relationship between antibody development and chimerism status.

Aims: A comprehensive understanding of the underlying mechanism of anemia in blood group incompatible-HSCT is crucial to identify the driving etiology of the post-transplantation hemolytic event. This requires detailed knowledge about recipient/donor blood group antigens and antibodies and thorough attention to the elapsed time between transplantation and anemia exacerbation.

Methods: We report on a 39-year-old female patient with acute myeloid leukemia, who underwent a 10/10 HSCT from an unrelated donor, with non-ABO incompatibility (blood group A- in the recipient while A + in the donor) and the same CMV-positive status. The recipient was alloimmunized against D antigen. Microsatellite markers PCR showed full donor chimerism on day +34. The patient developed severe hemolytic anemia on day +41. The analytical data were as follows: hemoglobin 6.3 g/dL, lactate dehydrogenase 331 IU/L, indirect bilirubin 1.5 mg/dL, undetectable haptoglobin, and CMV molecular test was positive for the first time after HSCT. We performed a series of tests to appropriately characterize the immune process and help unravel its etiology.

Results: The direct antiglobulin test was strongly positive for IgG and negative for C3d. An anti-D antibody, IgG1 subclass, was detected in the serum and in the eluate, its titer being 128. The patient received treatment with prednisolone and intravenous immunoglobulin (IVIg). Acyclovir was stopped and valganciclovir started. On day +57 the patient's clinical condition has significantly improved. On day +139 the patient developed a second episode of hemolysis and a second CMV infection. Valganciclovir was restarted. The patient was also treated with prednisolone, IVIg, rituximab, and later with foscarnet. Hemolysis and infection did not recur. Peripheral blood chimerism was 100% donor on day +165.

Summary / Conclusions: On both occasions these two immunologically determined events, the hemolytic anemia and viral reactivation, synchronously happened. Because most recipients clear the donor-specific antibodies within 120 days, the post-transplantation timing of alloimmune hemolysis development was attributed to residual recipient plasma cells that continued to produce anti-D antibodies, with this production being powered by a dysregulated immune tolerance linked to CMV reactivation and resulting in an immunologic disequilibrium

loop. In sum, because of new ancient alloantibodies looking like fresh autoantibodies in combination with T cell- and cytokine-mediated inflammatory processes, the pathophysiology of post-HSCT immune-mediated anemia is challenging intricate. This complexity requires close collaboration between transfusion medicine, its immunohematology laboratory, and transplantation team, to allow early identification of clinically significant antibodies and immediate selection of therapeutic strategies.

P450 | Immunodominance to naturally occurring RBC alloantigens is controlled by non-genetic factors in mice

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Background: Recipients of RBC transfusions have a general pattern of “responder” vs. “nonresponder” with regards to alloimmunization. However, “responders” do not typically become immunized to multiple alloantigens after each transfusion, but rather have sequential alloimmunization events with ongoing transfusion. This pattern of immunization is distinct from the polyvalent antibody responses that typically occur simultaneously against multiple epitopes during microbial infection or vaccination. Current mouse models of RBC alloimmunization are limited in testing sequential alloimmunization since they use model antigens expressed on RBCs from transgenic “donor” mice, which are a single highly foreign protein antigen and do not reflect the natural setting of multiple alloantigens with single amino acid differences. Moreover, some have argued that the current models study responses to “xenoantigens” and not authentic alloantigens.

Aims: The current study set out to identify and study alloantibody responses to multiple naturally occurring mouse RBC alloantigens.

Methods: C57BL/6 (B6) mice were transfused with RBCs from genetically distinct CAST/EiJ (CAST) mice every 14 days for a total of 6 transfusions ($n = 60$ total recipient mice in two cohorts). Serum was collected after each transfusion. Flow cytometry was used to measure B6 and CAST IgG responses and RBC clearance. CAST mice were backcrossed with B6 mice over 5 generations and B6-anti-CAST antisera was used to identify strains with isolated CAST antigens. Antisera was also used to assess alloantigen expression on the RBCs of 580 diversity outbred mice, each of which was genotyped with a GigaMuga array (143,000 markers). CAST antigens were mapped by quantitative trait loci (QTL) analysis and confirmed by testing reactivity with anti-CAST antisera on both CHO cells transfected with expression vectors of CAST variant genes and also CRISPR/Cas9 based mutagenesis of B6 mice to express CAST alleles.

Results: Three different RBC Mouse Antigens (RMAs) were isolated (RMA1^c, RMA2^c, and RMA3^c), with superscript to indicate CAST (C) or B6 (B) origins. QTL mapping identified, and CHO transfection and CRISPR/Cas9 mutagenesis of B6 mice confirmed RMA1^c as a

single amino acid polymorphism in glycophorin (93-Iso) with RMA1^b (93-Phe) as the antithetical B6 antigen. RMA2^c/RMA2^b was identified as a two amino acid polymorphism in Claudin13 (Thr-43-Ala, Iso-44-Thr). No locus for RMA3c was identified by QTL. Rates of B6 alloimmunization to transfused CAST RBCs were 0%, 60%, and 93% after 1, 3, or 6 transfusions. Importantly, a pattern of differential immunodominance was observed with anti-RMA1^c, anti-RMA2^c, or anti-RMA3^c predominating in 77%, 6.6%, and 3% of recipients, respectively. B6 anti-CAST antisera caused clearance of transfused CAST RBCs in a dose dependent fashion.

Summary / Conclusions: The current findings define multiple naturally occurring RBC alloantigens in mice and show variable rates and order of alloimmunization. Without ruling out the importance of donor and recipient genetics and/or environment—the data demonstrate that responder/nonresponder status and immunodominance of different alloantigens still occurs when genetics and environment are controlled. These findings justify seeking other explanations for patterns of alloimmunizations, such as variation in recipient T and B cell repertoires (as a function of random recombination of T cell and/or B cell receptors), which may cause differences in alloimmunization.

P451 | Study of the hemolytic activity of ABO antibodies—correlation between qualitative/quantitative tests, Complement-Mediated Haemolysis (CHUHE), and IgG subclasses

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Background: ABO antibodies are potent IgM immunoglobulins that may be associated with IgG or IgA. These antibodies can efficiently fix complement (IgM and usually IgG1 and IgG3 subclasses), potentially causing mild to fatal hemolytic reactions following platelet transfusion with ABO minor incompatibility or bone marrow transplantation with ABO major incompatibility. Titrating IgM and IgG ABO antibodies is described in literature as the best approach to avoid acute hemolysis in the formerly described situations, however this semi-quantitative method is unable to accurately detect complement-mediated hemolysis risk. The CHUHE assay (Complement Hemolysis Using Human Erythrocytes) is described as effective in identifying the risk of complement-mediated hemolysis and could be used to identify “dangerous” platelet donors. IgG subclass could also potentially reflect the risk of severe hemolysis due to the capability of activating complement cascade.

Aims: To evaluate if there is correlation between high titers of ABO antibodies (IgM/IgG classes), positive results for CHUHE assay and presence of IgG1 or IgG3 in predicting complement-mediated hemolysis and potential severe hemolytic transfusion reactions.

Methods: Fifty-five blood donor samples (group O) were selected for the study if presenting titers ≥ 100 for anti-A and/or anti-B using a semi-quantitative microplate technique (Neo, Immucor, Norcross, GA, USA). Anti-A and anti-B of all samples were also titrated in gel method (IgG and IgM). All samples were tested with the modified

CHUHE-P assay, classical qualitative hemolysin test, and determination of IgG1/IgG3 subclasses using gel-cards (Biorad, Mourat, Switzerland).

Results: Differences between the semi-quantitative method and classical antibody titration were observed. Referring to IgM antibodies: 85% of Anti-A and 67% of Anti-B included in the study showed titers < 128; whereas in the case of IgG antibodies: 47% of Anti-A and 64% of Anti-B showed titers < 128. In the CHUHE-P, regardless of the titration result, we observed that complement-mediated hemolysis was detected in 80% of the Anti-A (IgM and IgG); and in 84% of anti-B (IgM) and 85% (IgG). All cases resulting positive in the CHUHE-P assay presented partial hemolysis in the classical hemolysin test. Also, all cases with positive results in the CHUHE-P assay were classified as IgG1/IgG3.

Summary / Conclusions: Semi-quantitative test and qualitative hemolysin tests did not correlate with isohemagglutinin titers, indicating that implementing these methods as prophylaxis of choice to predict hemolysis when transfusing ABO incompatible platelets has important limitations. The CHUHE-P assay demonstrated that complement-mediated hemolysis can occur irrespective of ABO antibody titers and IgM/IgG class. However, CHUHE-P results were consistent with the presence of IgG1 and IgG3 subclasses. This can be used to classify apheresis platelet donors as 'dangerous', avoiding the transfusion in ABO mismatched patients even when ABO titers are considered safe.

P452 | Adaptation of the AABB technical manual method using plasma inhibition to distinguish anti-Ch and -Rg from other antibodies with similar characteristics on anti-IgG gel card

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Background: Inhibition of anti-Ch/Rg antibodies with plasma from Ch/Rg+ individuals on tube is useful to identify these antibodies and distinguish them from others with similar characteristics. The method is described in the AABB Technical Manual and is based on the fact that the antigens of the system Chido/Rodgers (O17) are not located on intrinsic red cell structures, but on the fourth component of complement (C4), which becomes bound to the red cells from the plasma. However, interpretation can be a challenge due to the complexity of the tube method and to the serological characteristics of these antibodies (high titer and low avidity).

Aims: For this reason, we developed a method adapted to gel card test (procedure not validated by the manufacturer) and we check its reproducibility in 3 patients with anti-Ch(a) and other with anti-Rg.

Methods: We have used fresh or frozen plasma from 3 patients with known anti-Ch(a) and 1 with anti-Rg(a). Prior to the technique, antibody titration procedure with anti-IgG at 37° on tube and on gel-based test was performed. The red cells used for the titration and

P452 - Table 1

	Anti-IgG gel card (Grifols®).	Anti-IgG gel card (Grifols®).	Anti-IgG gel card (Grifols®).
Patient 1	256	128	negative
Patient 2	64	32	negative
Patient 3	4	4	negative
Patient 4 (anti-Rg)	128	64	negative

P452 - Table 2

	Tube Titer	Tube Test plasma + Alb 6%	Tube Test plasma + Pool of plasma
Patient 1	8-16	weakly	negative
Patient 2	negative	negative	negative
Patient 3	negative	negative	negative
Patient 4 (Anti-Rg)	unrealized	unrealized	unrealized

inhibition methods were SeroCyte 3%-Grifols® for tube method. We resuspended them in 1% DG-Sol (Grifols®) for gel-based test. The method was performed on tube in accordance with the AABB Technical Manual. The method was adapted on anti-IgG gel card (Grifols®). Materials: Reactive red cell samples. Test plasma (Anti-Ch/Rg plasma). A pool of two or more normal plasma samples. 6% albumin. Anti-IgG and Anti-IgG gel cards. IgG-coated red cells.

Results: The titers on tube are weakly /undetectable, so we were unable to demonstrate inhibition with plasma. However, inhibition was demonstrated in all 4 patients with the new method adapted for anti-IgG gel card.

Steps: Gel card versus tube

- Serial dilutions of test plasma: No differences.
- Incubation plasma test with pooled plasma/6% Albumin: Smaller sample volume / Lower complexity / Same incubation time.
- Red cells: 1% versus 3%.
- Incubation samples with red cells: 15 min versus 1 h.
- Wash in saline: Unnecessary.
- Add anti-IgG to each tube: Unnecessary.
- Examine for agglutination: Easier.
- Add IgG-coated red cells: Unnecessary. The performance and interpretation of the method on gel-based test is simplified, reducing at least 1 h.

Summary / Conclusions: Binding of CH/RG antibodies to red cells is readily inhibited by plasma from CH/RG+ individuals. This is useful as an aid to identification of these antibodies. The adaptation of this method on anti-IgG gel card is simple, safe and has easy interpretation.

P453 | Prevalence study relating to the frequency and specificity of irregular antibodies anti-erythrocyte in patients belonging to the Blood Bank of Turin

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Background: The alloimmunization against red blood cell antigens remains of fundamental importance for blood banks and transfusion services worldwide. Screening for alloantibodies is essential within immunohematology laboratories, as they could cause violent reactions to transfusions.

Aims: From the literature data, positivity prevalences and immunization rates range from 0.45% to 2.40%; through this work, we describe the epidemiological context within our Transfusion Facility (TF), analysing the observed positivity prevalence and comparing it with that of the study by Politou, et al (*Turk J Hematology*, 2020) and our previous work (Amateis, *Blood Transfusion*, 2023).

Methods: Retrospective analysis through the transfusion management system (Eliot, Engineering) from January 1 to July 31, 2023. Verification in the population and in the examined period of subjects with positive transfusion compatibility test (report with alloantibody identification). Statistical analysis by MedCalc, version 7.3.0.1 (MedCalc Software Ltd, Belgium) with a significance value "*p*" < 0.05: Descriptive statistics: parameters such as mean, median, and percentages; Comparisons between groups: chi-squared test; Dichotomous variables between categories: Odds Ratio (OR) analysis; Comparison between ranks (M Vs F) for age and subsequent positivity detection of Alloantibody: two-tailed Mann-Whitney U test.

Results: - "Descriptive": 14733 patients underwent transfusion compatibility test (34249 tests performed) during the study period: 8108 males (55%) and 6625 females (45%), respectively, with an average age of 66 and 63 years (overall age: 64). 229 positive transfusion compatibility tests were reported: 84 M and 145 F, corresponding to 195 immunized patients: 71 M and 124 F. Positivity prevalence: per patient 1.55% and overall 1.32%. Frequency of alloantibodies directed towards the major systems: Rh 48%; Kell 18%; Kidd 7%; Duffy 3%; MNSs 9%. - "Comparison between groups" and "dichotomous variables" between M and F for positivity and negativity detection in transfusion compatibility test: chi-squared = 26.94 (DF 1) with *p* < 0.001; OR = 2.16 (95% CI: 1.6099 to 2.8957); - "Comparison between ranks": M: U = 3056 (mean = 117); F: U = 5747 (mean: 87); Z score = 3.55 with *p* = 0.0004.

Summary / Conclusions: The search for alloantibodies confirms an essential pre-transfusion test for each TF. Despite the limitations related to the sampled population and, not considering the clinical history of many patients and the department of origin (surgical, medical, and outpatient), the epidemiological trend of positivity in the search for irregular antibodies and their concurrent anti-erythrocytic specificity in the reality of the Blood Bank appears to be overlapping with that of the studies examined.

Considering the concurrent decline in blood component donations, these data provide a starting point for a more careful management of immunized patients in case national self-sufficiency would no longer be guaranteed.

P454 | Evaluation of a human Fab anti-idiotypic anti-Daratumumab to resolve pan reactions in pre-transfusion testing due to monoclonal anti-CD38 therapy

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Background: Monoclonal antibody therapies are used to treat a variety of pathologic conditions including solid and haematologic cancers. Daratumumab, a monoclonal anti-CD38 antibody, has been used for several years and has shown to be efficient for the treatment of multiple myeloma patients. This monoclonal antibody specifically targets the antigen CD38, which is overexpressed on multiple myeloma cells. However, CD38 is also expressed at various degrees on red blood cells. When a patient is treated with Daratumumab, pan reactions are observed in antibody screening using the Indirect Antiglobulin Test (IAT), which may mask underlying clinically significant antibodies. Several solutions have been developed, such as soluble recombinant proteins or treating the reagent red blood cells with dithiothreitol or trypsin, but they present limitations in the detection of some red cell alloantibodies.

Aims: The objective of this study was to confirm, in two different immunohematology laboratories, the efficacy of a new candidate in neutralizing Daratumumab interferences in pretransfusion testing. Another objective was to demonstrate that this candidate has no impact on the detection of underlying red cell antibodies.

Methods: The neutralizing solution was provided ready-to-use. The Daratumumab relative concentration in patient samples was estimated by antibody titration. To assess the neutralization efficiency, 50 Daratumumab samples (25 samples per site) were titrated and neutralized starting with 10% v/v of neutralizing solution and then with an increasing concentration if the neutralization was not achieved at 10% (i.e., 20% and then 30% v/v of neutralizing solution). No pre-incubation was required. Thereafter, 40 samples (20 samples per site) were prepared to contain a weak alloantibody (titre equal or smaller than 4 against single dose cell) and Daratumumab at a known titre. Specificities were selected to represent a broad spectrum of clinically significant antibodies. These positive samples were tested using the same neutralization protocol starting with 10% and so on if needed. Respective dilution controls using a titration buffer at 10%, 20% or 30% were used at each step to validate the neutralization tests.

Results: At both sites, Daratumumab titres ranged between 8 to 16'000. Overall, 74% of the samples (37/50) were neutralized using 10% of anti-Daratumumab: all samples up to a titre of 1024 (19/19), 80% with a titre of 2048 (4/5), 86% with a titre of 4096 (12/14), 22% with a titre of 8192 (2/9) and none with a titre of 16,384 (0/3). On

both sites, 92% (46/50) neutralization was achieved with 20% and then 100% with 30%. All underlying clinically significant alloantibodies were detected after the efficient neutralization of Daratumumab. Of 40 positive samples, 38 (95%) were neutralized with 10% and 2 required 20% of anti-Daratumumab but it did not affect the detection of weak underlying antibodies. Two samples showed positive but weaker reactions with 10% anti-Daratumumab but, eventually gave expected reactivities on subsequent testing.

Summary / Conclusions: This human Fab anti-idiotypic anti-Daratumumab demonstrated very efficient neutralization, while allowing the detection of weak underlying red cell antibody. Using a 10% concentration, the neutralization was successful in 83% of samples (75/90) and gradual increase achieved a complete neutralization. This user-friendly reagent could become a new candidate for Daratumumab neutralization in pre transfusion testing.

P455 | Preparation and application of anti-A₁, anti-AB and anti-H monoclonal antibody

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Background: ABO blood group is the most important blood grouping system during blood transfusion. Severe or even life-threatening complications, including acute haemolytic reaction, febrile non-haemolytic reactions, et al., could happen if the ABO blood types were mismatched. ABO blood typing and ABO compatibility test are basic assays required before blood transfusion. However, these two tests could give inconsistent results, and further tests using anti-A₁, anti-AB, and anti-H reagents to determine the subgroups are necessary. At the moment, most commercially available reagents used for A₁ and H antigen tests are plant lectins based, and mixtures of anti-A and anti-B antibodies are used as anti-AB antibodies. Therefore, the monoclonal antibodies against A₁, AB, or H antigen are in urgent need.

Aims: In order to determine the subgroup of ABO blood group, making monoclonal antibodies against antigens A₁, AB, and H.

Methods: Use human type A₁, AB, or O red blood cells (RBC) to immunize Balb/c mice, and generate monoclonal hybridoma cell lines that could produce anti-A₁, anti-AB, or anti-H antibodies. Determine the basic characters of these antibodies and compare them with commercially available reagents.

Results: Monoclonal hybridoma cell lines that could produce antibodies against human A₁, AB, or H were successfully generated, one for each, and they all produce mice IgM immunoglobulins. The anti-A₁ antibody could mediate agglutinations of type A₁ or A₁B RBC with a titre of 1:128, and there were no cross-reactions with type A₂, A₂B, B, or O RBC. A total of 158 clinical samples were tested alongside with commercial anti-A₁ lectins, and the results were 100% match. Also, in comparison with anti-A₁ lectins, anti-A₁ antibody has stronger agglutinations. The anti-AB antibody could mediate agglutinations of type AB RBC with a titre of 1:512. This antibody could mediate

agglutinations of type A₁, A₂, B, A₁B, or A₂B RBCs, and there were no cross-reactions with type O RBCs. In comparison with commercial anti-AB antibodies mixture, monoclonal anti-AB couldn't mediate agglutinations after pre-incubation with type A or B RBCs, while commercial anti-AB still could mediate agglutination of type A or B RBCs after pre-incubation with type B or A RBCs respectively. The anti-H antibody could mediate agglutinations of type O, A₂, B, A₂B, A₁, and A₁B H positive RBCs with titres of 1:128, 1:64, 1:64, 1:32, 1:16, and 1:16 respectively, and there were no cross-reactions with H negative RBCs. In comparison with commercial lectin reagents, the anti-H antibodies agglutination score with A₂, A₁, A₂B, and A₁B RBCs are 4+, 2+, 4+w, and 2+w, while lectins are 1+s, 1+, 1+s, and negative respectively.

Summary / Conclusions: In this work, we successfully made anti-A₁, anti-AB, and anti-H monoclonal antibodies. In comparison to current commercial reagents, the anti-A₁ antibody could produce clearer results for the naked eye to read with the same accuracy; the anti-H antibody performed better in case of titre, affinity, and sensitivity. The anti-AB antibody we made is an antibody that recognizes the common domain of A and B antigens, but the commercial ones are mixtures of anti-A and anti-B antibodies which have lower specificity.

Acknowledgement statement: Thanks Mr. Yong Li, Suzhou Institute of Biomedical Engineering and Technology Chinese Academy of Sciences, for guiding this study.

P457 | Development of microcolumn gel-based anti-human immunoglobulin reagent for patients receiving anti-CD-38 antibody treatments

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Background: CD38 is a transmembrane glycoprotein highly expressed on multiple myeloma (MM) cells, and fully humanized antibodies against CD38 like Daratumumab have been used in the treatment of MM. However, usage of anti-CD38 antibodies gives rise to problems during blood transfusion as CD38 is not only expressed on MM cells but also on red blood cells (RBCs). The presence of anti-CD38 antibodies could cause non-specific agglutinations during blood typing assays. Currently, two clinical strategies are used to address this issue: one is pre-treating the blood samples with dithiothreitol or 2-mercaptoethanol which denatures the CD38 on the cell surface to avoid their binding with anti-CD38 antibodies; the other way is to use positively charged polybrene to interfere with the antigen-antibody reaction. But either of these methods is not ideal, since these treatments could reduce the interaction between RBCs and blood typing antibodies, including ABO and Kell blood grouping systems. Therefore, a swift and accurate blood grouping reagent that could be used in the presence of anti-CD38 antibody is urgently needed.

Aims: To prepare a new microcolumn gel-based blood grouping reagent for patients receiving anti-CD38 treatments.

Methods: Develop a microcolumn gel card kit which lacks the antibodies that could bind to anti-CD38 antibody (anti-CD38 antibody unbind anti-human immunoglobulin, NCD38-AHG), and use this kit on samples from patients with anti-CD38 treatments. Compare the results from the newly developed kit with current commercially available kits.

Results: Six whole blood samples and 4 plasma samples from anti-CD38 antibody treated patients were collected. Direct antiglobulin tests (DAT) were performed on 6 whole blood samples; only weak or no agglutination was observed using commercial kits, and no agglutination was observed using NCD38-AHG kits. Indirect antiglobulin tests (IAT) for atypical erythrocyte antibodies were performed on all 10 samples; the agglutination scores were 1+ or 2+ using commercial kits, and no agglutination was observed using NCD38-AHG kits.

Summary / Conclusions: In this work, we developed a microcolumn gel card containing NCD38-AHG which can be used in samples containing anti-CD38 antibodies. This kit can be used in the blood grouping and compatibility assays before blood transfusion of patients receiving anti-CD38 treatment. Compared to current protocols for these patients, our kit is swifter and easier to perform.

Acknowledgement statement: Thanks Mr.Yong Li,Suzhou Institute of Biomedical Engineering and Technology Chinese Academy of Sciences, for guiding this study.

P458 | A rare cause of blood typing problems—adsorption of soluble A antigen after minor ABO-mismatched stem cell transplantation

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Background: Depending on their ABO type, secretors have soluble ABH antigens in body fluids such as saliva and plasma. These antigens continue to be secreted after hematopoietic stem cell transplantation (HSCT). Occasionally, detectable amounts of An antigen are adsorbed by the donor's red blood cells (RBC) resulting in weak A-positive reactions. This effect could be easily misinterpreted as a relapse with recurrence of the recipient's blood type.

Aims: We describe this rare phenomenon and a diagnostic strategy for unexpectedly positive A antigen results after HSCT in a patient with chronic myelomonocytic leukemia (CMML). In suspected cases, modern gel card results together with an old inhibition method allow to distinguish a relapse from the unusual adsorption of soluble A antigen to donor-type red cells.

Methods: To confirm the presence of soluble A antigen in the patient's plasma, an absorption-inhibition method was used in which the strength of an antiserum is reduced depending on the type and amount of antigen present in the plasma: anti-A serum (Immucor, IgM) was diluted in either the patient's plasma or saline (1:100) and incubated for 10 min at room temperature followed by serial dilutions up to 64. The diluted samples were incubated with A1+ test cells using a column agglutination gel card (NaCl/enzyme, BioRad). ABO blood

typing was performed using gel card technique (BioRad, Switzerland). A antigens were detected by clone A5 and AB antigens by clones ES131 (ES-15), Birma-1, ES-04. Samples with true mixed-field reactions served as controls (patients who switched from group O to A after HSCT and samples grouped O spiked with group A). The results were read immediately after centrifugation and 5 h later.

Results: A 64-year-old male A+ patient was, due to CMML, transplanted from a O+ donor. Monitoring after HSCT revealed complete donor chimaerism on d+30 in peripheral blood and also on day +69 in bone marrow sample. At first inconsistently to these findings, blood typing demonstrated a mixed field reaction A / O on day +7 and purely O-type red cells from day +14 to +41. Complicating for the interpretation, the time of the donor's rbc engraftment could not be assessed as the patient was transfused with a total of 10 donor-type units following HSCT. Starting at day +205, blood typing showed a positive reaction (1+ to 2+) to A and AB antigen when read immediately after centrifugation. The card was read again after 5 hours and showed an almost negative reaction for An antigen, but for AB antigen the reaction was almost unchanged (2+). In contrast to this instability, the reactions of spiked controls (preparatory mixed-field responses) were stable for A and AB antigens for at least 24 hours. It was suspected that the patient had soluble A antigen in his plasma, resulting adsorption on O-type RBC and misleading blood grouping results. The absorption-inhibition test was performed with the patient's plasma and showed a negative result for An antigen typing after incubation with anti-A serum, whereas the controls were still positive at a titer of 16. This supported strongly the hypothesis, that the adsorption of soluble A antigen and not a relapse was the reason.

Summary / Conclusions: A relapse is not the only possible explanation for unexpectedly positive A antigen results in follow-up HSCT recipients after temporarily donor-typed test results. The absorption-inhibition method can be a useful tool to differentiate between relapse or adsorption of An antigen in such patients.

P459 | Evaluation of a point of care system for ABO grouping and Rh(D) typing

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Background: Manual blood grouping is performed in our blood centre in various scenarios for donors like at voluntary blood donation camps and during blood group confirmation of indoor blood/ platelet donors as well as in the red cell serology lab during final blood group confirmation prior to issue by testing of sample from donor unit segment and at all times at patients' bedside as a part of bedside blood administration protocol. Slide method is prone to human error and inaccurate results which can cause substantial delay in the process involved.

Aims: To evaluate the sensitivity, specificity, precision and accuracy of the ABD PAD system for blood group testing of donors and recipients.

Methods: Immucor Neo results and Ortho Vision results were considered as standard reference results for donor and recipient blood group

testing respectively. Total 396 donor samples and 360 recipient samples were tested. 47 Rh (D) negative samples, five samples with weaker sub-groups and two with positive Direct Antiglobulin Test (DAT) were included in the evaluation exercise for donor samples. The recipient samples included 36 Rh (D) negative, two variants of D antigen, two DAT positive samples, two samples from patients on Daratumumab therapy, 10 samples of ABO non-identical BMT patients in various phases of transplant, two samples of patients with multiple alloantibodies and two samples of patients with diagnosed Autoimmune Hemolytic Anemia (AIHA). Five samples each with hemolysis and lipaemic appearance were included in evaluation exercise. Two senior technical officers independently interpreted the results of the blood group tests. All samples were tested within seven days of their collection and were stored at 2-6 degree Celsius.

Results: In the donor testing scenario, all the 396 results were concordant between the ABD PAD and Immucor Neo. In the patient testing scenario, one sample showed an additional reaction in the forward blood group. This was a patient of Multiple Myeloma with hyperproteinemia and visible rouleaux formation. Lipaemia and hemolysis did not affect the test performance adversely. Precision of the test and accuracy of the results could be well established. The technologists could be trained very easily and the test could be completed within less than five minutes.

Summary / Conclusions: The ABD PAD point of care blood group testing system is easy, quick and robust system for both donor as well as recipient blood group testing. The results are dependable in even complex immunohematological scenarios and hence it can be utilized as a tool for immediate blood group testing at donation site, in serology laboratory as well as at patients' bedside.

P460 | Evaluating CD38 antigen expression on erythrocytes and its implications for immunohematological tests in patients undergoing Daratumumab treatment

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Background: The use of the human monoclonal IgG1 kappa antibody against CD38 (Daratumumab) is now a standard and, in some cases, first-line treatment for Multiple Myeloma (MM). Anti-CD38 binds to CD38 sites on erythrocytes, resulting in pan-agglutination in irregular antibody screening and panel identification. Treating test erythrocytes with the chemical dithiothreitol (DTT) can eliminate this interference, although it can also lead to the denaturation of antigens in the Kell, Dombrock, and Lutheran systems. Despite the need for information on Daratumumab use in all MM patients requiring transfusion, it is not consistently implemented. Pan-agglutination reactions in antibody screening are typically homogeneous, but weaker reactions may occur in certain red blood cells, suggesting heterogeneous variability in CD38 expression.

Aims: Explore the variability in CD38 expression among healthy individuals to elucidate the diverse strengths of agglutination observed in

screening and antibody identification tests for patients treated with DARA.

Methods: A total of 800 repeated blood donors were selected for this study, all of them are O blood type. The anti-CD38 reagent was diluted with physiological saline at concentrations of 1:100, 1:1000, and 1:3000 and tested with fresh donor red blood cells in gel tests with AGH (Grifols, Barcelona). Donor samples showing no agglutination with anti-CD38 were subjected to flow cytometry expression assays, comparing them with the antigenic expression of umbilical cord samples.

Results: Among the 800 donors tested with anti-CD38 in AGH, 81% ($n = 646$) exhibited a 2+ intensity, while 19% showed the following weaker reactivities: 8.2% ($n = 67$) with 1+, 5.5% ($n = 44$) weak intensity, and 5.3% ($n = 43$) with no agglutination (negative). Donors with a negative result ($n = 43$) were tested with higher concentrations of anti-CD38 (1:1000), and with this new dilution, 40/43 showed weak agglutination intensity. The three samples from donors with serologically negative results were subjected to flow cytometry assays, revealing CD38 expression equivalent to newborns.

Summary / Conclusions: CD38 expression is heterogeneous among healthy donors, with a prevalence of reactivity below expected (2+) in 19% of individuals (weak or 1+). This diversity can present challenges in antibody identification for DARA-treated patients, as panels may include 1 to 2 cells with minimal CD38 expression. The heterogeneity in agglutination could be misleading during panel analysis, potentially indicating multiple antibodies when it's merely a variation in CD38 antigen expression.

P461 | Provision of Mia-negative red cells for transfusion in patients with anti-Mia in conjunction with multiple antibodies

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Background: The Mia antigen is part of the Miltenberger series of phenotypes within the MNS system, and is rare in most populations. Within Asia and Southeast Asia however, it is fairly prominent, with a frequency of around 6% in Chinese populations, and up to 88% in certain indigenous people in Taiwan. Anti-Mia is one of the most commonly found alloantibody identified within Singapore's diverse population and has been known to cause acute and delayed HTRs and severe HDFN.

Aims: To establish a protocol for recommending, and providing Mia-negative red cell concentrates for transfusion in patients with anti-Mia with multiple antibodies and/or autoantibodies.

Methods: This study employs a comprehensive approach, involving the identification of patients with anti-Mia and additional antibodies and/or autoantibodies through serological testing. A transfusion recommendation is then given to the respective crossmatching and transfusion facilities, specifying the use of Mia-negative red cells. An inventory of Mia-negative red cells was also being established to provide adequate supply of antigen-negative units available for transfusion.

Results: In 2023, Anti-Mia emerged as the predominant antibody in 840 (23%) out of 3669 referred cases to the Red Cell Reference

Laboratory. Usually, the transfusion recommendation of providing anti-human globulin(AHG)-crossmatch compatible red cells was given to patients with anti-Mia. It's not a major issue if anti-Mia was the only antibody present as an AHG-crossmatch would have picked up any incompatibilities. In the 821 cases with underlying anti-Mia, 136 (16%) of them came with an additional antibody, and 29 (3.5%) cases presented with 2 or more antibodies. In these cases, antigen-negative red cells for the corresponding clinically significant antibodies were provided to the crossmatching facility, and an AHG-crossmatch is performed to detect incompatibilities, including that of anti-Mia. In the event where an incompatible crossmatch is obtained, another unit would have to be provided for crossmatching. 25 (3%) cases of anti-Mia also came with autoantibodies. Which makes AHG-crossmatch unable to pick up the incompatibilities from anti-Mia due to the interference from the autoantibodies. For such cases, it would be advantageous to recommend giving Mia-negative red cells instead of suggesting an AHG-crossmatch. The availability of monoclonal anti-Mia antisera enabled phenotyping of donor red cells to provide an Mia-negative blood inventory for the selection of compatible blood units for transfusion.

Summary / Conclusions: In conclusion, this study emphasises the crucial role of Mia-negative red cells in transfusions for patients with Anti-Mia, particularly when multiple antibodies and autoantibodies are present. The developed protocol enhances transfusion compatibility, minimising complications and delays. The findings underscore the importance of tailored transfusion protocols, advocating for Mia-negative red cells in specific scenarios over conventional AHG-crossmatching. This approach is particularly beneficial when patients present with additional antibodies or autoantibodies, where AHG-crossmatching may be less effective. Looking ahead, integrating mass screening for donor phenotypes promises a sustained supply of antigen-negative red cell units, facilitating the selection of compatible blood units. Future studies and collaboration with transfusion facilities will further refine the protocol and enhance transfusion practices for patients with complex antibody profiles.

P462 | Detecting the DVI variant in the obstetrical and patient populations

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Background: DVI variant individuals can both develop anti-D if exposed to Rh positive cells and cause the development of anti-D. Identifying these individuals in the evaluation of obstetrical patient care is critical to prevent potential significant negative outcomes in pregnancy. The detection by direct testing of the partial D variant DVI in neonatal population when evaluating the Rh-negative obstetrical patient is useful to categorizing the need for Rh immunoglobulin (RHIG) prophylaxis.

Aims: A monoclonal based prefilled column-based test reagent was evaluated for performance to detect RhD as well as the DVI variant and compared with an additive column-based test reagent that

detects DVI. Reagent testing was performed using full automation, semi-automation and manual test methods.

Methods: Testing compared a prefilled anti-D (anti-D clones RUM1 and P3X21223B10) column (Ortho BioVue® System - Anti-DVI) (BD6) in three cassette types; full forward type cassette with two anti-D reagents (one detecting DVI+ and one that does not), a cassette designed for the detection of D and DVI variants for newborns and a cassette used to confirm the Rh type of donors. The comparator test used the additive test method of Ortho Sera™ Anti-D(DVI) (anti-D clone ESD1M) (OSD6) reagent using a BioVue Reverse Diluent cassette. Samples included patients (375) with various clinical conditions, cord bloods (248) and random samples (1461) with tests for RhD. All samples were tested on the fully automated Ortho Vision® Analyzer with sample subsets tested on the Ortho Vision Max analyzer, semi-automated Ortho Optix™ Reader and manually using an Ortho™ Workstation with a visual read of the reaction. A select group of 62 additional known variant D antigen expression including 18 DVI+ were evaluated. When results were discrepant between the test and the comparator, the results were deemed discordant. A tube-based test using Ortho BioClone® Anti-D was used to evaluate discordant results for information only. Concordance was defined as % agreement between the test and comparator.

Results: The 990 test results (534 patient and 394 cord blood), included 798 positive (468 patient /330 cord blood) and 130 negative (66 patient/64 cord blood), demonstrating 100% agreement across both reagents. A separate study with random samples generated 1623 test results concordant across all analyzer systems including 1396 positive and 227 negative. Overall concordance was 100% with 99.8% at the lower bound of the 95% confidence interval. Tests of 54 samples from the selected population of variant D including all 18 DVI+ samples, showed positive reactivity in comparison tests. There were eight tests positive with BD6 vs negative with OSD6. Two samples were identified as DIVa, one DNU and five weak D. The tube-based test of these variants demonstrated positive reactivity.

Summary / Conclusions: Use of Anti-D that detects variant DVI is valuable in labeling donors and evaluating the status of the newborn for RhD delivered by a Rh-negative obstetrical patient for RhIG prophylaxis. The use of this reagent can identify DVI variant patients when used in parallel with a reagent Anti-D that does not detect DVI. The DVI test reagent demonstrated 100% concordant reactivity and specificity with routine Rh-positive, DVI variants and Rh-negative patient and cord blood samples.

P463 | To evaluate clinical and serological correlation of positive direct antiglobulin test

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Background: The Direct Antiglobulin Test (DAT) is crucial for identifying antibodies binding to red blood cells (RBCs), indicating their involvement in RBC destruction. A positive DAT suggests the

presence of auto or allo-antibodies on RBC surfaces. While the intensity of the DAT reaction often correlates with antibody quantity, its direct relation to symptom severity may vary, particularly in cases of prior RBC destruction. Therefore, a comprehensive interpretation of DAT findings is indispensable for precise diagnosis and clinical decision-making in transfusion medicine.

Aims: To conduct a comprehensive evaluation of the clinical and serological correlation associated with positive Direct Antiglobulin Test (DAT) results and to categorize the class and subclass of antibodies implicated in the immune response.

Methods: A prospective observational study was conducted from July to December 2023, encompassing samples and DAT requests from all relevant areas. Blood samples were subjected to the Direct Antiglobulin Test (DAT) using LISS Coombs ID card 'DAT IgG/C3d' via the column agglutination technique (CAT). Positive DAT cases underwent further testing with a monospecific LISS Coombs ID card. The grading of DAT positivity was meticulously performed for each patient, accompanied by thorough documentation of clinical records, including medical history, transfusion records, drug history, and relevant laboratory parameters. To ensure study integrity, strict exclusion criteria were applied, excluding neonatal samples and cases with recent intravenous immunoglobulin administration or hypergammaglobulinemia associated with multiple myeloma or Waldenström macroglobulinemia.

Results: The prospective study examined a cohort of 125 patients, spanning ages 7 to 83 years, with a gender distribution of 76 males (60.8%) and 49 females (39.2%). The mean age was 42.6 years, with a standard deviation of 15.3 years. Predominant disease categories associated with positive DAT results included autoimmune disorders (30, 24%), liver diseases (36, 28.8%), and infections (14, 11.2%), with 94 patients (75.2%) having a history of previous transfusions. Grading of DAT positivity varied: 1+ observed in 24 (19.2%), 2+ in 66 (52.8%), 3+ in 24 (19.2%), and 4+ in 11 patients (8.8%). Serologically, IgG antibodies were identified in 73 patients (58.4%), IgM in 5 patients (4%), and IgA in 4 patients (3.2%), while 39 patients (31.2%) tested positive for IgG, C3d, and C3c antibodies. Positive DAT results strongly correlated with clinical manifestations of hemolytic anemia in 80% of patients, presenting as jaundice, fatigue, and pallor. These findings underscore the clinical relevance of DAT results and emphasize the importance of comprehensive evaluation and management in affected patients.

Summary / Conclusions: Our study illuminates the intricate role of positive DAT results in understanding immune-mediated hemolytic disorders. Significant correlations between DAT positivity and diverse manifestations of hemolytic anemia were revealed. Serological analysis elucidated the complex interplay of IgG, IgM, and IgA antibodies, highlighting immune complexities. Notably, previous transfusions underscored sensitization dynamics in clinical management. The graded scale of DAT positivity reflects variations in disease severity and antibody binding strength, providing insights for tailored interventions and advancing comprehension of immune-mediated hemolytic disorders for optimized patient care.

P464 | Implementation of automated column agglutination gel method for titer determination in alloimmunized pregnant women—initial findings

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Background: The conventional manual tube method (CMTM) is most common titration procedure, which was in some transfusion laboratories replaced by manual and recently, by automated column agglutination technique (CAT). CMTM titration in indirect antiglobulin test (IAT) have been associated with variations regarding the preparation of the test reagents, serial dilutions, indicator red cells phenotype, manual pipetting and the visual interpretation of the end point titers. The published results pointed out that end point alloantibody titer in pregnant women is generally higher in CAT gel technique compared with CMTM. Some literature data suggest that end point titer is regarded as semi quantitative and does not represent the clinical severity of HDFN, while titer score (TS) better correlates with the risk for HDFN, comparing with titer values, taking into account the strength of antibody reaction and the antibody avidity.

Aims: To introduce automated CAT for titer determination in Laboratory for immunohaematological testing of BTIS; to compare end point titer and titer score results of automated CAT titer determination with the results of CMTM in all investigated blood samples.

Methods: Titers from 15 plasma samples containing anti-D from 6 pregnant women, 2 samples with anti-c from 2 pregnant women, 1 sample with anti-E and 1 blood sample containing anti-K Abs have been determined by CMTM and the automated CAT method on the IH-500 system (Bio-Rad Europe, GmbH) from July, 2023 till January, 2024. CMTM was performed following the published instructions and the laboratory experience: glass tubes labelled from undiluted up to 1:1024 dilution, 100 µl 0.9% saline solution; 100 µl of the tested plasma into the first and second labelled test tubes; 4% suspension of indicator red cells, O, R₀r, prepared "in house"; incubation 60 ±15 minutes at 37°C; wash 3×; 100 µl of the antihuman globulin reagent (A.H.G. Elite Green, Lorne Laboratories LTD); centrifugation at 1000 rpm for 1 min, end point: (+-), macroscopically. Fully automated CAT titration procedure on the IH-500 system (Bio-Rad Europe, GmbH), according to the instructions for use (Bio-Rad, Titration Procedures - A Guide to Good Laboratory Practices 2019/10) and the indicator O, 8% red cell suspension O, R₀r (ID-DiaPanel, Bio-Rad Laboratories, Inc.); end-point: 1+, macroscopically. Scores (Marsh) were calculated manually in both cases: 4+ = 12, 3+ = 10, 2+ = 8, 1+ = 5, (+-) = 2.

Results: In 10/15 anti-D set titers results were equal, TS values were higher for CAT gel method in all 10 cases; in 3 samples, values of CAT gel method titer were one dilution higher than of CMTM, one of them belonged to a female which newborn died shortly after the second intrauterine transfusion and delivery, with TS values 77 (CAT) versus

55 (CMTM); in 2 samples values of CMTM were one dilution higher than of CAT gel method, one of them was connected with the successful birth of the newborn followed by 2 red cell transfusions. The set titers results for non- D Abs showed equal results for both methods and higher TS for all samples in gel method.

Summary / Conclusions: Our first findings suggests that automated CAT for titer determination can successfully replace conventional CMTM titration method and that TS better correlate with clinical outcomes of pregnancy, but more investigation is needed for profound conclusion.

P465 | Abstract withdrawn

P466 | The important role of anti-D standards

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Background: Anti-D can cause severe haemolytic transfusion reactions and haemolytic disease of the foetus or newborn (HDFN). To prevent this from happening, anti-D immunoglobulin prophylaxis is given to unimmunised D-negative women following the known causes of sensitisation. The prenatal protocol with Anti-D immunoglobulins recommends screening all pregnant women and administering treatment as necessary. Many centres carry out foetal RHD genotyping to determine whether the foetus is at risk. If the foetus is RHD-positive, anti-D titres and anti-D quantification are monitored in most centres to determine plasma antibody levels during and after pregnancy. Thus, decisions for clinical treatment are made based on assay results, and hence, there is a requirement for accurate standardisation. The use of Anti-D standards can greatly improve the accuracy and reliability of assays, leading to better patient outcomes. The fact that there are several anti-D reference materials and standards available has led to confusion. It is therefore vital to improve the understanding of these standards and reference materials to ensure patient safety and harmonisation between laboratories.

Aims: Increase the awareness and correct usage of available Anti-D standards for use with blood grouping reagents, internal quality control, calibration of automated haemagglutination methods, and potency assays.

Methods: Three reference materials for anti-D: the WHO 3rd International Standard for anti-D immunoglobulin (16/332), the WHO 1st IS for minimum potency preparation of anti-D blood grouping reagents (99/836), and the British standards for Anti-D (Rho) antibodies (73/517) have been established to date.

Results: These preparations were sent out in an international collaborative study and were evaluated whether they met their intended use. The standards demonstrated a significant reduction in interlaboratory variation particularly in haemagglutination tests, which are notoriously difficult to standardise.

Summary / Conclusions: While the prophylactic use of Anti-D immunoglobulin has decreased the incidence of HDFN in developed

countries, it remains a serious public health problem in many parts of the world. Monitoring and treatment require various steps and assessments to prevent HDFN, which differs from one country to another. Three standards are available to support the decision in clinical treatment. For example, a reference preparation for quantifying anti-D in immunoglobulin products is unsuitable for quantifying the level of anti-D in plasma and an alternative plasma standard, for use with automated haemagglutination, is available (coded: 73/517). More training is required to increase clarity and standardisation in this area.

P467 | Irregular antibodies during pregnancy—a 4-year series from an immunohematology reference lab in a Uruguayan center

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Background: Irregular antibodies against red cell antigens might present a burden during pregnancies. This depends on several factors, like temperature of antibody activity, type of immunoglobulin, and antigen density on the fetal red cells, when present. Routine antibody screenings are part of the laboratory workup for every pregnant woman. Our Immunohematology Reference Laboratory (IRL) at Uruguay's National Blood Service regularly receives samples from centers all over the country to perform antibody identification and provide input for clinical decisions.

Aims: To analyze the irregular antibodies profile of samples from pregnant women for the period of 2020-2023.

Methods: We performed a retrospective review of the IRL records. This includes analyzing overall antibody frequencies, mixture of antibodies, autoantibodies and unresolved cases. Descriptive statistical analysis was performed with SPSS 29.0.

Results: In a 4-year period, we received samples from 201 pregnant women that accounted for 236 irregular antibodies. 210 (88.9%) were alloantibodies and 26 (11.1%) were autoantibodies. The most common alloantibody specificities were: 59 (28%) anti-M, 38 (18%) anti-Le^a, 21 (10%) anti-D, 20 (9.5%) anti-E, 13 (6.2%) anti-c, 9 (4.2%) anti-Le^b, 8 (3.8%) anti-K, and 6 (2.8%) each for anti-C, anti-C^w, anti-S, anti-Jk^a and anti-P1. Rounding the count were 8 (3.8%) antibodies with undertermined specificity. These either were panagglutinating antibodies that did not react with the patient's cells, or were antibodies against an antigen (or antigens) not described in our cell charts. Our lab does not store known rare red cells and still has not implemented molecular biology tests to help reach a conclusion on these cases. The 26 autoantibodies detected included 14 (53.8%) cold agglutinins with high thermal range, 6 (23.7%) panagglutinating autoantibodies and 6 (23.7%) autoantibodies with known specificity, all from the Rh blood group (4 autoanti-e-like and 2 autoanti-C). Samples with multiple alloantibodies presented a very distinctive profile: out of the 59 anti-M, only 1 was found in a mix (with anti-Le^a). All anti-K presented as a single specificity, as well as all the -C^w, -S, and -P1. On the other hand,

7 (77,7%) of the 9 anti-Le^b samples were in a mix and all 7 presented both anti-Le^a and -Le^b; 7 (53,8%) of 13 anti-c presented with multiple antibodies, and in all cases the other was anti-E. Only 4 (19%) anti-D were in a mix, in all cases with anti-C. Out of these 4 cases of “anti-C +D” no anti-G workup was performed. From the top two frequencies (anti-M and anti-Le^a) only 1 anti-M and 1 anti-Le^a presented strong (3+ or more) reactions at 37°, the rest were room temperature (RT) antibodies. P1 and anti-Le^b antibodies were also RT specificities. This means that 110/236 (46.6%) antibodies were RT specificities. The rest reacted very well at 37°, although clinical outcomes from newborns were not detailed.

Summary / Conclusions: In a 4-year period we detected 236 antibodies from pregnant women samples. 89% were alloantibodies, for which anti-M (28%), anti-Le^a (18%) and anti-D (10%) were the most prevalent. 110 (46.6%) were RT antibodies: all the anti-Le^b, -P1 specificities, and all but one each of anti-M and -Le^a. We must note the still predominance of anti-D in our charts, in spite of RhD immunoprophylaxis as standard of care for RhD negative pregnant women in our country. When in a mixture, the most common and consistent alloantibodies associations were anti-Le^a+Le^b, anti-c+E and anti-C+D.

P468 | Antibodies against high frequency antigens, 6 years' experience in Kuwait central blood bank

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Background: High frequency antigens (HFA) are those with the incidence of more than 90% but majority of these have more than 99% of incidence. More than 190 antigens are classified as HFA by International Society of Blood Transfusion. Transfusion management of patients alloimmunized against high frequency antigens is often problematic. Antibody identification is required to assess likely clinical significance, exclude underlying alloantibodies and guide decisions regarding suitable blood for transfusion. The routine transfusion laboratories cannot diagnose the antibodies against HFA and the help of reference laboratories is often required.

Kuwait central blood bank recipient RBC serology laboratory is an immunohematology reference laboratory. It is a core laboratory serving all regional hospitals in Kuwait. It provides pre-transfusion compatibility testing including cases with complex antibody identification and antibodies against HFA.

Aims: To determine the frequency of red cell antibodies against HFA at Kuwait central blood bank in the span of 6 years.

Methods: For all cases, ABO grouping, red cell phenotyping and direct antiglobulin test (DAT) were performed using tube technique. Antibody identification was done using both LISS indirect antiglobulin test and Bio-Rad panel cards. DTT and papain enzymes treatment was used to determine the specificity of the antibodies, in addition to allogenic adsorption using donor cells with same patients' extended phenotype. Red cell typing

P468 - Table 1

	Total number of cases from 2018-2023
Anti-Ge2	26
Anti-Yt ^a	10
Anti-k	8
Anti-Jr ^a	6
Anti-Jk3	6
Anti-H	4
Anti-U	3
Anti-Lan	2
Anti-Rh17	2
Anti-Ge3	1
Anti-CH	1
Anti-Kp ^b	1
Anti-ER4	1

was confirmed serologically using commercial reagents/qualified frozen patients' sera. In addition to genotyping using ID core XT.

Results: Total of 74427 patients' samples were received from January 2018 until December 2023. 71 samples were identified as antibodies against HFA during the study period. All the samples were showing reactivity with all panel cells, negative DAT, negative auto-control, and reactivity with donor cells according to patients' extended phenotype. There were 58 (81.7%) females and 13 (18.3%) males. Out of these cases, 26 (36.6%) were with anti-Ge-2 antibodies showing the highest prevalence. 7 of the cases were investigated by the International Blood Group Reference Laboratory, Bristol, UK. The results are summarized in table 1:

Summary / Conclusions: The clinical significance of the antibodies against HFA is variable. Finding compatible blood for patients with such antibodies may be a challenge. In Kuwait central blood bank, the sources of antigen negative blood are family members, rare donor registry and autologous donations in case of planned surgeries. In addition to our participation in the American rare donor program since 2018. Thus, the transfusion support was satisfactory in all these cases. Our study revealed that the most common antibody to HFA was anti-Ge2 antibody. This is because Ge: -2, 3, 4 (Yus Phenotype) has been found in white population including Arabs, a Turkish Cypriot, and a Middle East Jews. In addition, anti-Ge-2 antibodies may occur without red cell stimulation. Family studies demonstrates that Yus phenotype is inherited among the families in Kuwait.

P469 | Transfusion management for patients receiving anti-CD38 treatment (Daratumumab) for multiple-myeloma, five years' experience. Kuwait central blood bank

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Background: Daratumumab is a medication used to treat multiple myeloma patients by targeting the CD-38 markers on the surface of myeloma cells. It also has the ability to bind to red blood cells' CD-38, which can obstruct conventional tests for clinically significant antibodies. Patients will often have a pan-reactive antibody panel, which tends to obscure the existence of any antibodies that are clinically significant. This includes a positive auto-control. The binding of daratumumab to CD-38 on the surface of red blood cells can be effectively negated by treating the antibody panel cells with dithiothreitol (DTT) and conducting additional tests however, DTT also inactivates/destroys many antigens on the red blood cell surface by breaking disulfide bonds. The umbilical cord blood reagent was found to be an effective substitute for denaturing CD-38 RBCs. Since the cord blood cells have low levels of CD-38, it stands to reason that their reactivity with DARA-treated patient plasma will be lower.

Aims: This paper details the immunohematological results of pre-transfusion tests on patient receiving Daratumumab for multiple myeloma, in span of 5 years. As well as strategies to avoid any monoclonal antibody interfering with other tests.

Methods: Serological tests, ABO, Rh-D, direct antiglobulin test (AHG) performed by tube method. Antibody screening, identification and cross match were performed with LISS indirect antiglobulin test and Bio-Rad gel cards. Molecular testing, using ID core XT, was performed to determine the phenotype. A stock of cord blood was obtained through an agreement with the maternity hospital in Kuwait. Extended phenotype was performed for the cells and a panel is created for identification, in preparation for the cord blood to be used as a reagent RBCs.

Results: A total of 77 pre-transfusion samples of patients with multiple myeloma, receiving daratumumab, were sent to Kuwait central blood bank (KCBB) for serological testing from January 2019 to December 2023. Information regarding the patients' transfusion and medication history are sent to KCBB' laboratories. Three patients were found with history of positive antibodies before starting the treatment (2 cold antibody and 1 anti-E antibody), however currently with negative antibody screening. After treatment samples were showing a positive identification results with all panel cells. DAT results was variable, but elution was negative with all samples. Cord blood panel identification was negative with all patient samples.

Summary / Conclusions: When patients are given phenotypically similar red blood cells consistently, it has the advantage of lowering the possibility of sensitization and subsequent alloantibody development for the matched red cell antigens, which is the policy used in KCBB. Any potential anti-RBC alloantibodies won't react with matching RBC units. Because Anti-CD38 is present in the patient sample, AHG crossmatches with phenotypically or genotypically matched units will still be crossmatch incompatible.

P470 | "Stepping Up" to serologic problem solving with anti-CD38 therapies

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Background: Anti CD38 monoclonal antibodies are widely used as therapeutic agents for plasma cell neoplasms. These agents interfere with pretransfusion serological testing by binding to CD38 present on reagent red blood cells(RBC). The degree of interference varies with some demonstrating a negative DAT and antibody screen while others have persistently positive panreactive antibody screens and/or a positive DAT. The differences in reactivity may depend on timing or route of drug administration. Several strategies for investigation of samples with anti CD38 interference have been described. Use of some may involve techniques not available at rural or remote sites.

Aims: To develop and evaluate a stepwise testing algorithm for use in a multi hospital region; and to limit use of complex investigative tools and sample referral to cases where interference persists following basic serological evaluation.

Methods: A stepwise testing algorithm was developed for samples from patients treated with anti CD38 therapy: Initial gel card antibody screen. Computer assisted crossmatch (CAC) if antibody screen negative. Gel antibody ID if screen positive, not panreactive. PEG tube antibody screen if gel screen panreactive. CAC if PEG tube screen negative. PEG antibody ID if screen positive, not panreactive. DTT treated screening cells if PEG tube screen panreactive. CAC if DTT screen negative. DTT treated cells antibody ID if DTT screen positive, not panreactive. Phenotype matched RBC and or further investigation if DTT screen positive, panreactive. A DAT was also evaluated for all and if positive, a serological crossmatch corresponding to the method where the antibody screen was negative, was required. For example, if the PEG antibody screen was negative with a positive DAT, a PEG crossmatch would be performed for pre transfusion compatibility. For those requiring antibody screen using DTT treated cells, Kell negative RBC were selected for crossmatch. Based on this approach used in 11 hospitals over 10 months, the number of samples in each category were enumerated and the number requiring complex testing (DTT treated, phenotype matched or other investigative techniques) determined.

Results: Over ten months, 100 samples from 36 anti CD38 treated patients were evaluated and found to have a panreactive gel screen. Of these, 43 had a negative PEG antibody screen and a negative DAT, with CAC pre transfusion. Thirty had a negative PEG antibody screen with a positive DAT requiring PEG IAT crossmatch. Seventeen had a panreactive PEG screen, a negative DTT treated antibody screen and a CAC. Nine had a panreactive PEG screen, a negative DTT screen and positive DAT with serological crossmatch using PEG IAT with DTT treated cells. One had a positive screen using DTT treated cells and required further investigation prior to transfusion along with phenotype matched RBC.

Summary / Conclusions: An algorithmic approach for pre transfusion investigation of patients treated with anti CD38 therapies minimized investigative steps. Seventy three percent were successfully managed using techniques readily available in laboratories capable of basic serological testing. Not all patients treated with anti CD38 require DTT modified RBC investigation nor phenotype matched RBC transfusion. Initial assessment with basic techniques saves laboratory time, effort and costs while optimizing safe and timely transfusion.

P471 | Comparison of performance of automated blood grouping analyzers in detection of ABO subgroups and H-deficient phenotypes in Hong Kong blood donors

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Background: Automated blood grouping analyzers are widely used in blood centers in virtue of their high throughput and outstanding performance in terms of accuracy and precision. In Hong Kong, the frequency of ABO subgroups and H-deficient phenotypes are in the range of approximately 1 in 1,500 and 1 in 15,000 respectively. B subgroups are particularly prevalent in ethnic Chinese donors. It is of paramount importance that the ABO/H variants are not missed detected by automated analyzers in daily operation.

Aims: We compare the performance of automated blood grouping systems, PK7400 (Beckman Coulter), NEO Iris (Immucor) using microplate agglutination and the IH-1000 (Bio-Rad) employing column gel agglutination in the detection of ABO subgroups and H-deficient phenotypes.

Methods: Twenty known ABO subgroup and 2 H-deficient, secretor donation samples were typed for ABO blood groups using the 3 systems. The reagents used in the assays were: (1) PK7400: in-house panel including forward typing by 0.9% bovine serum albumin (Bio-Rad), 1:40 diluted anti-H lectin, 1:400 diluted anti-A₁ lectin, 1:130 diluted Seraclone anti-A (Clone A003), 1:80 diluted Seraclone anti-B (Clone B005) and reverse typing by 2% A₁ cells, 1.7% A₂ cells and 2% B cells; (2) NEO Iris: default ABO grouping panel and (3) IH-1000: DiaClon ABO/D + Reverse Grouping gel card. Undetermined (UND) and discrepant ABO blood groups were sent to Reference Laboratory of the Hong Kong Red Cross Blood Transfusion Service for confirmation.

Results: PK7400 detected all 22 (100%) ABO/H variants and results were flagged as UND. H-deficient samples were detected with negative reactions to anti-A, anti-B and anti-H lectin in forward typing. NEO Iris correctly detected all ABO subgroups and missed 1 Para-Bombay phenotype (21/22 = 95.5%). IH-1000 was able to detect 15 out of 20 ABO subgroups and 1 Para-Bombay phenotype (overall: 16/22 = 72.7%).

Summary / Conclusions: Automated blood grouping analyzers are widely used in blood transfusion services. In the respect of detection of ABO subgroup in donation samples, PK7400 outperformed the others in comparison. For detection of H-deficient phenotype, PK7400 also did better on account of the choice of in-house panel with the use of anti-H lectin in forward typing. Adjustment of settings in the latter 2 analyzers such as inclusion of H-typing and use of O cells in reverse typing for anti-H/HI detection as well as refining the criteria for interpreting positive reactions in reverse typing may

improve the performance in the assays in these aspects. To include more H-deficient samples into future study would lead to more confident findings.

P472 | Passenger lymphocyte syndrome in a liver transplanted patient - a challenge with multiple antibodies

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Background: Passenger lymphocyte syndrome (PLS) is a unique type of graft-vs-host disease (GVHD) in which donor's immunocompetent B lymphocytes within the graft, produce antibodies against the recipient's red blood cells (RBCs) causing immune-mediated haemolysis. Antibodies typically appear 1 to 3 weeks after transplantation and disappear within 3 months. Although haemolysis is usually mild, rare cases of severe haemolysis have been reported. Primary treatment is blood transfusion of an organ donor's blood type. Immunosuppression therapy may be added for moderate and severe cases, including corticosteroids, tacrolimus (TAC) or rituximab. For severe cases, plasmapheresis may decrease the alloimmune response.

Aims: Our aim is to present an alloimmunized patient with anti-K and anti-Jka antibodies transplanted with non-ABO identical liver and haemolysis caused by PLS and a delayed haemolytic transfusion reaction (DHTR) with anti-c, anti-E and anti-A1 antibodies of the donor origin.

Methods: A 58 years old female patient with alcoholic liver cirrhosis was admitted to our hospital for a liver re-transplantation due to artery thrombosis and infarction of the liver transplant. Immunosuppressive therapy consisted of mycophenolate mofetil (MMF), TAC and prednisolone. For antibody identification RBC reagents Identisera Diana and Diana P were used with cards DG Gel Coombs and DG Gel Neutral (Grifols). For direct antiglobulin test card DG Gel DC Scan (Grifols) was used. Eluates were prepared from the patient's RBCs using acid elution kit Gamma ELU-KIT (Immucor). Izohemagglutinin titer was performed in the tube test.

Results: Blood type of the patient was AB RhD positive and blood type of the donor was B RhD positive. Patient's Rhesus phenotype was CcD.Ee. Prior to liver transplantation (LT) two alloantibodies were detected in the patient's blood: anti-K and anti-Jka. During liver re-transplantation the patient received 8 units of AB RhD positive RBCs, which were K and Jka antigen negative and the crossmatch was negative. On the third day, two units of RBCs were requested. Crossmatch was now positive with antigen K and Jka negative RBC units. In antibody identification panel new alloantibodies were identified in the patient's plasma and eluate: anti-c and anti-E. Also anti-A1 antibody

was present in the patient plasma and eluate. Direct antiglobulin test was positive; anti-IgG 4+ and anti-C3d negative. At that time donor blood sample was requested and in the serum anti-c and anti-E antibodies were identified. RBCs of the units that the patient received during LT were all AB RhD positive, K and Jka negative, while the status of c and E antigens were unknown. Isohaemagglutinin titer of anti-A1 was: 4 (IgM) and 8 (IgG). Haemolytic markers were present, with the lowest haemoglobin (Hb) 64 g/L, and the highest total bilirubin (T-Bil) 175 µmol/L, direct bilirubin (D-Bil) 143 µmol/L and lactate dehydrogenase (LDH) 607 U/L. After antibody identification the patient received two O RhD positive and two B RhD positive units, all antigens c, E, K and Jka negative. After blood transfusion, laboratory parameters began to normalise, and at the 18th postoperative day were: Hb 76 g/L and T-Bil 18 µmol/L.

Summary / Conclusions: Before LT, in addition to the donor's blood type, an antibody screening test should also be performed in order to prevent PLS. In operative and postoperative period RBC units compatible with donor's and patient's plasma should be used.

P473 | Comparison of conventional tube technique with automated gel microcolumn assay for antenatal antibody monitoring

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Background: Timely identification of pregnancies at risk of severe hemolytic disease of the fetus and newborn (HDFN) requires accurate and reproducible antibody titration. Most antenatal screening laboratories still regard the manual conventional tube technique (CTT) as the gold standard though critical titers may vary by antibody type and cells selected for titration (single or double dose). In Finland, as in many other countries, a titer of ≥ 16 is typically used as the critical titer for anti-D and many other antibodies. As CTT is labor intensive, time consuming and prone to inter-operator variability, fully automated titration methods could improve efficiency and reproducibility.

Aims: To compare the relative performance of CTT and automated microcolumn assay (GMA) in antenatal antibody titration. To establish a critical titer for GMA that corresponds to the titer of ≥ 16 established for CTT. The titer must detect all cases that require intrauterine transfusions for severe HDFN (sensitivity) without leading to a significant increase in the number of unnecessary follow-up visits or interventions at local delivery hospitals or university hospitals (specificity).

Methods: EDTA blood samples ($n = 143$) with clinically significant RBC alloantibodies were collected from 89 pregnant women. Antibody specificities were anti-D ($n = 31$), -DC ($n = 11$), -DE ($n = 4$), anti-E ($n = 18$), anti-Ce ($n = 2$), anti-c ($n = 27$), -cE ($n = 22$), -C ($n = 2$), anti-K ($n = 8$), anti-Fya ($n = 11$), anti-Jka ($n = 6$), -Jk3 ($n = 1$).

Twofold serial dilution titers with CTT were made parallel with GMA with an automated system. The reagent RBC used was an in-house single dose RBC as a 3.5% suspension for CTT and 0.8% for GMA according to antibody. For both methods the reciprocal of the highest dilution of plasma yielding a 1+ positive reaction was regarded as the titer.

Results: Titer values ranged from negative to 256 for CTT and from 1 to ≥ 2000 for GMA. The linear relationship between CTT and GMA titers was good (Spearman's correlation 0.93). The GMA titers were on average 2.86 (95% CI 2.69-3.03) higher than CTT titers for all samples, and 3.37 (95% CI 3.07-3.67) higher for anti-D (or -DC, -DE). Using a tube titer of 16 as the gold standard, the sensitivity and specificity for a critical titer of ≥ 64 for gel were 100% and 79% for all samples, and 100% and 53% for anti-D (or -DC, -DE).

Summary / Conclusions: A fully automated gel assay offers many advantages for laboratories, especially in terms of workflow. In this study, we saw a good linear relationship between the CTT and GMA methods, with the GMA results being about 3 dilutions higher. A critical titer of ≥ 64 for GMA would detect the same cases requiring intrauterine transfusions identified by the CTT titer of ≥ 16 . However, due to lower specificity, switching to GMA would lead to unnecessary clinical follow-ups and possible interventions for over 20% of all cases with an antibody titer above the critical level, rising to almost 50% for anti-D (or -DC, -DE). In Finland, antenatal screening for maternal red cell antibodies is centralized to the Finnish Red Cross Blood Service laboratory. Delivery hospitals rely on the screening laboratory to differentiate high-risk cases from low-risk ones, not only with 100% sensitivity, but also with acceptable specificity. Therefore, the advantages of a switch to a fully automated titration method need to be carefully balanced with the clinical consequences for maternal-fetal units and immunized pregnant women.

P474 | Coexistence of sickle cell anemia, allo- and auto-antibodies, and severe autoimmune hemolytic anemia in an adolescent patient—case report

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Background: The development of alloantibodies (alloAbs) or autoantibodies (autoAbs) is a complication observed in sickle cell disease (SCD). Autoimmunization occurs in 7.6% to 12% of chronically or intermittently transfused SCD patients. The clinical implication of autoAbs in SCD patients is unclear.

Aims: To report the coexistence of SCD and severe autoimmune hemolytic anemia (AIHA) in an adolescent patient.

Methods: Case report.

Results: A 12-year-old male patient, O+ blood type, no personal or family medical history of AIHA, diagnosed with SCD, splenectomized, cholecystectomized, with bilateral femoral head necrosis and chronic hypoxemia. After exposure to 16 sporadic red blood cell (RBC) units

he developed multiple alloAbs (anti-Fya, anti-Jka, anti-S, anti-Lea, and anti-Leb). The patient was admitted with sudden worsening of skin pallor and jaundice, weakness, dyspnea, fever, somnolence, tachycardia, tachypnea, hypotension and desaturation. Cardiac auscultation revealed a gallop rhythm, and the liver was palpable at the costal arch. Blood tests showed worsening of anemia [hemoglobin from 9.1 g/dL (baseline) to 4.8 g/dL], a drop in reticulocyte count [from 255×10^9 /L (baseline) to 80×10^9 /L], 65 erythroblasts per 100 WBCs, increased lactate dehydrogenase [from 566 U/L (baseline) to > 6,000 U/L], and increased indirect bilirubin [from 1.6 mg/dL (baseline) to 2.7 mg/dL]. Leukocytes and platelets were normal. The direct antiglobulin test (DAT) revealed the presence of an undetermined IgG (panagglutinin). No new alloAbs were identified. Clinical and laboratory data led us to confirm the diagnosis of warm AIHA. Infection, primary immune deficiencies, malignancy, and autoimmune diseases were excluded. The patient was treated with steroids plus intravenous immunoglobulin (IVIG). He received methylprednisolone (1 g/day, for 3 days) with further transition to oral prednisone (60 mg/day for 28 days, and then weaned). The IVIG administration was 0.4 g/kg/day, during 5 days. Three weeks after treatment, the patient's Hb, reticulocyte count, lactate dehydrogenase, and indirect bilirubin were at his baseline values. The DAT became negative 2 years later. There was no recurrence of AIHA beyond six years of follow-up.

Summary / Conclusions: Few studies have been focusing on pediatric SCD and AIHA, and the prevalence and incidence are unknown. The alloAbs are more frequent in SCD patients. An association of inflammation, immune dysregulation and autoimmunity has been demonstrated in SCD patients; and the studies identify alloAbs, transfusion exposure (24 to 72 RBC units) and splenectomy as risk factors for the development of autoAbs. The alloAbs bind to RBC membrane creating neoantigens, and promoting immune system activation. No specific alloAbs has been described as a more frequent inductor of autoAbs, and the most frequent autoAbs are pan-agglutinating IgG or autoAbs with Rh-specificity (mainly to "e" antigen). Significant autoantibody titers have been reported after splenectomy, possibly due to compensating activity of other peripheral lymphoid organs. There are no guidelines on management of AIHA in the SCD patient. For general pediatric patients, the first-line therapy for warm-AIHA is corticosteroids, with different doses and time-courses. IVIG has been used in addition to steroids as adjunctive therapy in more severe cases. Further studies are needed to better elucidate the pathophysiology, and treatment of this potentially life-threatening condition.

P475 | Predictive value of self-reporting of race for the Fya/Fyb phenotype

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Background: African countries have reported normal reference ranges for white blood cell counts and absolute neutrophil counts (ANC) as being much lower than countries where individuals of African descent are in the minority, such as the United States. Recent guidelines have suggested using blood bank testing for the Fya/Fyb antigens to predict a patient's potential for having this low-normal neutrophil count, as the Fy-null status has about a 70% chance of correlating with African ancestry.

Aims: Correlation with the Fy null status and self-reporting could (1) improve confidence in clinicians' assessments and/or (2) add enough evidence to a current workup as to avoid the need for the test at all.

Methods: Between December 2018 and July 2022, Seattle Children's Hospital Transfusion Service Laboratory tested 154 samples for single antigen phenotyping for Fy (which includes the Fya and Fyb antithetical antigens). These samples were ordered from a single-center pediatric hematology outpatient clinic attached to the hospital. IRB approval was given to review the patients' testing and related testing, diagnoses, and demographics.

Results: From the 151 of 154 samples (three duplicates removed), 77 were Fy-null (51.0%), and 74 were not Fy-null, phenotypes being Fya+/Fyb+, Fya+/Fyb-, or Fya-/Fyb+ (49.0%). Of the Fy-null specimens, 48 of 77 self-reported "Black or African American" (62.3%), and 29 of 77 self-reported another category (37.7%), and this included the term "Other" and "Patient Refused". Of the specimens that were not Fy-null, 6 of 74 (8.1%) self-reported "Black or African American", and 68 of 74 (91.9%) reported another category, including the category "Other" and "Patient Refused." In the first analysis, the sensitivity of self-reporting "Black or African American" in relationship to testing Fy-null in blood bank testing is 62.3%, and specificity is 91.9%. The positive predictive value (PPV) of self-reporting "Black or African American" in relationship to testing Fy-null is 88.9%, and negative predictive value (NPV) is calculated to be 70.1%. The positive likelihood ratio of testing Fy-null in patients who self-report as "Black or African American" is 7.6. A second analysis was performed and those self-reporting as "Other" and "Patient Refused" were added to the group of patients who reported "Black or African American." With this new grouping, 69 of 77 were Fy-null and self-reported "Black or African American," "Other," or "Patient Refused" (89.6%), and 8 of 77 were Fy-null and self-reported in the remaining categories (10.4%).

Summary / Conclusions: The evaluation of the Fy phenotype status on red blood cells performed in blood banks can elucidate the lack of Fya and Fyb antigens (the Fy-null status), which correlates strongly to an African heritage. A retrospective review of pediatric patients referred to a tertiary care hematology outpatient clinic showed that self-reporting of "Black or African American", "Other", and "Patient Refused" had a high sensitivity of a Fy-null status, and a high specificity was observed when patients reported other race categories. This analysis demonstrates that the correlation of patient self-reporting of race could factor into early non-invasive determination of a patient's Fy-null status, thus assisting early in avoiding unnecessary additional testing.

P476 | Automated blood group antibody titrations using gel card column agglutination—equivalent accuracy for ABO and non-ABO antibody titres between automated and manual testing

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Background: Titration techniques are a semi-quantitative measurement of antibody levels using two-fold serial dilutions of the plasma or serum and Reagent Red Blood Cells (RRBC). Clinical applications include but no limited to: (1) the determination of alloantibody levels in obstetrical patients to monitor risk of HDFN, (2) to identify low-titer ABO plasma for ABO non-compatible plasma and platelet transfusion, (3) to select low-titer group O+ whole blood for trauma patients, 4) antibody pre/post-adsorption-elution techniques, (4) to study the antigen site density of red blood cells, and 5) to monitor ABO antibodies in mismatched bone marrow transplants. A new automated 2-fold serial dilution method was developed up to a 1:2048 dilution for gel agglutination antibody titration using the Erytra and Erytra Eflexis systems.

Aims: The aim of the study was to compare the Erytra and Erytra Eflexis automated antibody titration technique with titrations performed manually and tested by the DG Gel manual method.

Methods: A set of 180 plasma samples was selected containing ABO or non-ABO alloantibody (Rh, K, Kidd, Duffy, MNS). For titrations using DG Gel Coombs and DG Gel Anti-IgG cards, approximately 50% of the titrations were ABO antibodies and 50% were non-ABO antibodies. For titrations using DG Gel Neutral cards, approximately 90% of the titrations were ABO antibodies and 10% were non-ABO antibodies. The study compared the titre obtained with the Erytra and Erytra Eflexis with the titre obtained with the equivalent DG Gel manual method. The latter technique was performed as described in the respective IFUs of each DG Gel card (DG Gel Coombs, DG Gel Anti-IgG and DG Gel Neutral). The results were considered in agreement following the criteria of a maximum allowed difference of 1 two-fold serial dilution for titres <128 and of 2 two-fold dilutions for titres ≥128.

Results: The percentages of agreement and lower confidence bound (95%) considering the three DG Gel cards were 99.4% (97.4%) for Erytra and 100% (98.3%) for Erytra Eflexis.

Summary / Conclusions: A high level of agreement was observed for antibody titrations up to a 1:2048 dilution and performed on the Erytra and Erytra Eflexis compared to manual DG Gel method using DG Gel Coombs, DG Gel Anti-IgG, and DG Gel Neutral cards. The results of this study demonstrate that the automated antibody titrations are equivalent to the manual DG Gel method for both ABO and non-ABO antibody titrations.

P477 | Association of blood group antigens and *Helicobacter pylori* infection in blood donors or Yoruba ethnicity

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Background: *Helicobacter pylori* (*H.pylori*), a bacterial pathogen, is a risk factor of diminished cognitive development, ubiquitous and infect both males and females. It is usually found under the mucus layer in the gastric pits where it causes chronic active gastritis, peptic ulcer disease and is a strong risk factor for the development of gastric cancer. These conditions alter haematological indicators variably but there is dearth of information on the association of *H.pylori* and blood group antigens. There is growing interest in our locality on the distribution of minor red cell antigens and their relationship with disease conditions.

Aims: The broad objective of this study was to determine the relationship between erythrocyte blood group antigens and *Helicobacter pylori* infection.

Methods: 181 consenting donor blood sample were screened for *H. pylori* after the administration of a structured questionnaire using rapid technique and ELISA. Saline washed red cells were phenotyped to determine the red cell antigen profile using a wide range of antisera against D, C, c, E, e, M, N, S, s, K, k, Le^a, Le^b, Kp^a, Kp^b, Lu^a, Lu^b, Fy^a, Fy^b, Jk^a and Jk^b antigens by microtitre saline(IgM) and enhancement techniques. The prevalence and association between infection and the red cell antigens was statistically determined using SPSS.

Results: An overall prevalence of 27% was recorded in the sample population. The distribution of the red cell antigens in the infected and control group was variable, but there was a significant association ($p < 0.05$) between the presence of Lu^a, Fy^a, Fy^b, Jk^a and Jk^b and *H. pylori* infection.

Summary / Conclusions: The prevalence of infection is alarming in the population considering the impact it may have on cognitive development and other haematological variables previously reported, which may affect general performance. The observation of the association of the aforementioned antigens and *H.pylori* infection warrants further studies in other populations with larger samples. Red cell antigens play a role in susceptibility to infections and the mechanisms responsible can be explored in clinical interventions.

P478 | Challenges in the management of transfusion in patients treated with anti-CD47 monoclonal antibodies, unresolved problem

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Background: Emerging cancer treatments with novel monoclonal antibodies have been reported to interfere with pre-transfusion testing

methods. Notably, the administration of anti-CD47 monoclonal antibodies is known to be challenging for blood typing due to the presence of the antigen on both red blood cells (RBC) and platelets. Consequently, inadequate management may lead to delays in delivering appropriate blood components, thus jeopardizing patient care and safety.

Aims: To describe the characteristics of transfusion and transfusion strategies in patients treated with anti-CD47 monoclonal antibodies.

Methods: A retrospective single-center study was carried out by following up all eligible patients to receive treatment with an anti-CD47 monoclonal antibody. Patients in screening phase were recommended to undergo a baseline pre-transfusion testing that included (i) the assessment of ABO and Rh D groups; (ii) direct (DAT) and indirect globulin tests (IAT), and (iii) RBC extended phenotype (Cc, Ee, Kk, Jka, Jkb, Fya, Fyb, MN, Ss), or genotype, if patients had been transfused within the preceding three months or had a positive DAT test. A protocol was established to use the anti-human globulin that does not detect IgG4 (Immucor®) and a maximum of 6 alloabsorptions as mitigation strategies to resolve potential panagglutinations in the laboratory.

Results: Our transfusion area was made aware of 29 potential patients for the treatment of anti-CD47 compounds from July 2017 to December 2023. Among them, 17 patients ultimately received the aforementioned treatment. Participant's age mean was 65.5 years (range: 36-84 years) and 58.8% were women ($n = 10$). Baseline diagnoses included lymphomas ($n = 7$), leukemia ($n = 2$), and solid tumors ($n = 8$). The anti-CD47 monoclonal antibodies considered were CC-90002 ($n = 7$), Magrolimab ($n = 3$), ALX148 ($n = 1$), and SL-172154 ($n = 6$). Treatments were indicated as the third therapeutic line in 41.2% of the patients and at least 10 patients had a previous history of transfusion. During anti-CD47 monoclonal antibody treatment, 6 patients required at least one blood transfusion, from which 2 patients suffering from Acute Myeloid Leukemia received more than 10. In order to determine the best possible blood type to be transfused to each patient, several mitigation strategies were used. During treatment with CC-90002, pre-transfusional testing was successfully mitigated by means of the anti-human globulin that does not detect IgG4 ($n = 2$). We observed that low treatment doses of Magrolimab (1-15 mg/Kg) could be mitigated by using the anti-human globulin that does not detect IgG4 and a high dose (30 mg/Kg) could be temporarily mitigated by using alloabsorptions. Isophenotype transfusions were administered when mitigation was no longer possible with either strategy due to ongoing high-dose treatment ($n = 2$). For ALX148, no strategy could mitigate drug's impact on pre-transfusional tests and, thus, isophenotype transfusions were performed ($n = 1$). SL-172154 could not be evaluated, since pre-transfusion tests were not demanded for patients ongoing treatment. At therapy completion, 6 patients required blood transfusions, while still exhibiting a positive IAT, thus strategies described were also used.

Summary / Conclusions: The present study highlights the challenges faced by a transfusion service as novel therapies with implications for transfusion become more available. Additionally, the need to actively communicate with clinical units becomes evident to ensure a safe transfusion to the patient.

P479 | Rare anti-Jk3 detected in a patient with multiple myeloma - first case reported in a local tertiary hospital in Singapore

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Background: Our patient is a 52-year-old lady of Malay descent with multiple myeloma. She was planned for autologous hematopoietic stem cell transplant. Patient had no known transfusion but was multiparous (G3P3). Her current sample was typed as O Positive with pan reactivity of 3+ reaction observed with Ortho Clinical Diagnostic (OCD, USA) 0.8% 3-cell antibody screening panel using the column agglutination technique (CAT) on Ortho Vision Swift Analyzer. It was noted that she had previous histories in 2 local hospitals (Hospital T and K) between 2020 to 2023. Her first Type and Screen (TS) in 2020 from Hospital K showed a negative antibody screen result. Samples from Hospital T in 2022 and 2023 showed a positive antibody screen but no further investigation was done. It was not known if patient had any transfusion between 2020 to 2023. Antibody identification OCD (USA) 11-cell panel showed pan reactivity of 2+ reaction with a negative auto control. It was suspected initially that patient might be a Bombay phenotype due to pan agglutination to panel and screening cells with a negative autocontrol, but anti-H lectin result (4+) was concordant with Blood Group O individuals. As her antibody identification results were inconclusive, red cell phenotyping and Direct Coombs Test (DCT) were performed. She was found to be D+C+c+E+e+, K-, Fy(a+b-), Jk(a-b-), M+N+S-s+, DCT negative. With her unusual Kidd phenotypes, it was suspected that patient may have Anti-Jk3 as all antibody identification red cells used were Jk(a+b-), Jk(a-b+) or Jk(a+b+).

Aims: To ensure that patient did not have any other red cell alloantibodies and to confirm the presence of Anti-Jk3, patient's sample was sent to Blood Services Group (BSG) in Singapore and Australian Red Cross Lifeblood (ARC) in Brisbane Australia for further testing.

Methods: Serology investigation was performed by BSG while targeted sequencing was performed by ARC.

Results: BSG confirmed that patient had Anti-Jk3 and excluded presence of other red cell alloantibodies. Due to the lack of documented sensitizing event apart from being multiparous, targeted sequencing was requested and performed by ARC. Her genotype was JK*02N.01/*02N.01 with a predicted phenotype of Jk(a-b-). JK*02N.01 is a null allele more prevalent in Polynesians and lesser in Southeast Asians (1-2%). It is postulated that Anti-Jk3 developed from sensitization during pregnancy and subsequent antibody evanescence, characteristic of Kidd antibodies, thus an initial negative antibody screen in Hospital K. As she was prepared for autologous stem cell harvesting and transplant, she was monitored closely. Transfusion is usually given to patient at Day 5 and Day 10 post-transplant. Our patient was well,

with hemoglobin level not below 7g/dL throughout her admission thus, did not receive any red cell transfusion post-transplant though frozen rare Jk(a-b-) red cell units were available upon request.

Summary / Conclusions: Anti-Jk3 is a rare antibody implicated in hemolytic transfusion reactions, and this was our first reported case. The presence of anti-Jk3 poses issues with transfusion and is serologically challenging to investigate without appropriate red cells available to perform exclusion as anti-Jk3 may mask other red cell alloantibodies. Her targeted sequencing results concurred with the red cell phenotyping confirming that patient is Jk3 negative. Fortunately, patient had an uneventful transplant and was discharged well Day 18 post-transplant.

P480 | Incidence of delayed serological transfusion reaction identified in the Regional Blood Center Poznan in 2021-2023

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Background: Delayed serological transfusion reaction (DSTR) as a cause of delayed haemolytic transfusion reaction (DHTR) typically occurs 24–28 h following the transfusion of RBCs, in individual cases even later. Recently DSTR is increasingly less associated as the cause of DHTR and the patient's possible symptoms are not usually explained by transfusion of RBCs. This state of affairs contributes to the fact that DSTR is seldom considered as a cause of hemolysis in patients. Therefore, it is necessary to draw special attention to a newly detected alloantibodies active in the eluate from RBCs.

Aims: Retrospective frequency analysis of alloantibodies identified in sera or/and eluate from alloimmunized patients context of time since the last transfusion. This analysis focused on cases where there is active red cell eluate or/and mixed agglutination after a recent transfusion.

Methods: Identification of RBC alloantibodies is performed routinely for patients with positive screening for RBCs antibodies. This analysis includes studies on immunized patients from 2021 to 2023. RBCs from alloimmunized patients, initially identified as containing alloantibodies, were analysed by elution tests. Patients' RBCs were tested in Direct Antiglobulin Test (DAT) both polyspecific (IgG+C3d) and if necessary monospecific (IgG, IgA, IgM, C3c, C3d) reagents. Sera were tested in: Indirect Antiglobulin Test (IAT) and Enzyme Test (using commercial identification panel with gel-microtubes). Eluates (commercial acid-elution kit) were tested in IAT only.

Results: In the analysed period, alloantibodies were identified in 4131 patients, 123 of them had IgG alloantibodies identified both in sera and eluate from RBCs. In all 123 cases, antibodies were not detected during the compatibility test before the last transfusion. 9 of the analysed cases of DSTR were classified as DHTR and 58 as probable or possible cause of DHTR. Generally, it was difficult to determine antibodies as exact cause of the patient's haemolysis because of patient's disease and clinical condition which may have been the cause of RBC damage as well. Alloantibodies with more than one specificity were

identified in 35 of 123 patients with DSTR. Due to their specificity, alloantibodies were identified: anti-RhE-61, anti-Rhe-3, anti-RhC-10, anti-Rhc-11, anti-RhCw-1, anti-K-11, anti-Jka-31, anti-Jkb-6, anti-Fya-2, anti-Fyb-2, anti-S-3, anti-s-1, anti-M-2, anti-Lua-3, anti-Kpa-1. **Summary / Conclusions:** In order to differentiate the causes of hemolysis, routine use of tests to evaluate the risk of hemolysis should be considered in patients with positive DAT and alloantibodies identified in the eluate. The evaluation could include the determination of IgG subclasses and the degree of RBC coating. The ratio of DHTRs to DSTRs is difficult to estimate due to the varied clinical condition of patients and a number of other factors that may cause damage to RBCs. Our analysis shows that large percentage of identified alloantibodies was anti-Rh (70%) and anti-Jk^a (25%). Therefore, preventive measures are worth considering. Probability of occurrence of the DSTR/DHTR can be reduced by avoiding unnecessary transfusions, and by providing RBCs prophylactically matched for at least RhCcEe. Ensuring accurate antibodies identification documentation as well as obtaining an accurate transfusion history and communicating with the blood banks where a patient was previously transfused will decrease the probability of occurrence non-detectable repeated and reinduced antibodies.

P481 | Evaluation of a soluble CD38 based method for mitigation of anti-CD38 interference in pre-transfusion compatibility testing

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Background: CD38 is a protein highly expressed on myeloma (MM) cells that has been shown to be an effective target antigen for monoclonal antibody therapies, so treatment with anti-CD38 monoclonal antibodies is a first line therapy for patients with MM. Although CD38 is weaker expressed on erythrocytes anti-CD38 binds to CD38 on reagent RBCs and cause panreactivity *in vitro* and subsequent false positive reactions in indirect antiglobulin tests (IAT), antibody detection (screening) tests, antibody identification panels, and anti-human globulin (AHG) crossmatches. Agglutination due to anti-CD38 may occur in several media (eg, saline, low ionic strength saline, polyethylene glycol), and with all IAT methods (e.g., gel, tube, solid phase).

Aims: Report our experience in evaluation of a new soluble CD38 based commercial method for mitigation of anti-CD38 interference in pretransfusion compatibility testing.

Methods: We evaluated the Grifols sCD38 methods, this test uses soluble CD38 to mitigate the anti-CD38 antibody interference in serological assays by adsorbing the drug in patient's plasma samples. Its performance was evaluated in 15 patients: 10 with negative IAT, of these 5 treated with anti-CD38, 4 with positive IAT, of these 2 treated with anti-CD38, 1 with positive DAT not treated with anti-CD38. The new method has been compared to the current standard method using dithiothreitol (DTT 0.4M), which denatures the CD38 antigens on test panel erythrocytes. All treated patients were in therapy with

Daratumumab. After treatment plasma samples were further processed on the Grifols Erytra System as we internally validated the procedure.

Results: In our experience mean time to perform antibody screening and cross match was about 150 min using DTT method and about 50 min using sCD38 method. The sCD38 effectively mitigated the interference caused by anti-CD38 antibodies in 7/7 (100% efficacy) of patient samples tested while DTT was successful in only 5/7 (71% efficacy); no interference was observed in patients presenting anti erythrocyte antibodies. Moreover, there was no negative influence on DTT sensitive blood group systems such as KEL upon sCD38 treatment.

Summary / Conclusions: Department of Transfusion Medicine on Venice's district cover a network with 1 Hub Hospital and 9 spoke Hospital, serving about one million inhabitants. Second level immunohematology tests were centralized in our Laboratory in Mestre and in the year 2023 we processed 129 patients treated with anti-CD38 from across the department. Therefore the reduction from 150 to 50 min in the time needed to perform tests for mitigation of anti-CD38 interference appears to be relevant with a recovery of approximately 210 technical-hours per year. Another highly appreciated operational aspect was the possibility of treating the patient's plasma and perform tests using automatic instruments (Erytra Grifols) available in our laboratory. In the evaluation of this new method, we did not observe failures in the mitigation of anti-CD38 interference. Furthermore, the results obtained in the samples that presented allo or auto antibodies were not affected by the treatment with sCD38. In our experience Grifols sCD38 assay is straightforward and quick to perform and it is superior to DTT treatment in the mitigation of anti-CD38 antibody interference in MM patients treated with Daratumumab.

P482 | Serologic testing and resolution of ABO discrepancies caused by anti-Rh17 in the D - phenotype

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Background: Individuals with the rare D- phenotype can develop antibodies to the high frequency Rh17 antigen (anti-Rh17 or anti-Hr₀) which have been implicated in severe HDFN and acute hemolytic transfusion reactions. Two group A prenatal patients with the D- phenotype and history of pregnancy loss due to anti-Rh17 were evaluated serologically prior to possible intrauterine transfusion (IUT). Group O D- RBC were not available necessitating use of group A D- RBC. Both were noted to have a co-existing ABO discrepancy on reverse typing (unexpected reactivity with A1 and A2 RBC) prompting a review of other anti-Rh17 cases and further investigation into the cause of this finding.

Aims: To assess the prevalence and cause of ABO discrepancy associated with anti-Rh17 antibodies.

Methods: The laboratory electronic database of a national reference lab was searched for cases of anti-Rh17 antibodies. ABO/Rh typing and antibody identification were performed using standard hemagglutination methods including use of in-house frozen rare D- RBC. A1 typing was performed using anti-A1 lectin (DBL). Anti-Rh17 titration was performed by serial dilution in SIAT using DCE/DCE or DCE/DcE group O RBC both with and without DTT treatment of plasma (0.01M). DNA was extracted from whole blood in EDTA using Qiagen Qiacube and tested with the Progenika ID Core XT assay.

Results: From January 1, 2021 – December 31, 2023 a total of 6 anti-Rh17/D- cases were identified (2 blood donors, 4 patients as shown in Table 1). All were homozygous for RHCE-D[5,7]-CE by genotyping with serologically confirmed C-c-E-e- phenotype. 2/6 anti-Rh17 had discrepant ABO reverse typing on at least one testing occasion, but this varied over time. Both prenatal cases exhibited strong group A forward typing with multiple reagents. Plasma from sample 4 had unexpected 2+ reactivity with A1, A2 and O RBC with no evidence of auto or allo cold reactive antibodies. Plasma from sample 6 had 1+ reactivity with A1 and A2 RBC but was nonreactive at immediate spin (IS) room temperature with O RBC and the autocontrol. Alloadsorption of plasma 6 with ZZAP treated dce/dce group O RBC abolished IS reactivity with A1 and A2 RBC. Non-reactivity of both plasmas against two confirmed A1 positive D- RBC was used to resolve the ABO discrepancy. For sample 6, the anti-Rh17 titre using neat and DTT treated plasma was unchanged (both 128)

P482 - Table 1: Results.

Sample #	Donor/Patient/Prenatal	Anti-A/Anti-B/A1 cell/B cell	ABO Group	Anti-RH17 Titre
1	Donor	4/0/0/3	A positive	16
2	Donor	0/4/3/0	B positive	256
3	Patient	4/0/0/3	A positive	Not tested
4	Prenatal	4/0/1/4	A positive	>1024
5	Patient	0/0/4/4	O positive	Not tested
6	Prenatal	4/0/1/4	A positive	128

suggesting an IgM component was not contributing to the ABO discrepancy.

Summary / Conclusions: Two group A D- - prenatal patients with high titre anti-Rh17 antibodies exhibited discrepant results in reverse ABO typing. Given the plan to use group A D- - RBC for IUT, the use of A1 confirmed D- - RBC for reverse typing combined with alloadsorption techniques was essential to confirm that the anti-Rh17 was the cause of the ABO discrepancy. Testing in one sample did not identify an IgM component to account for reactivity at IS suggesting that anti-Rh17 IgG may cause direct agglutination of red cells in a subset of cases. The extent to which certain IgG antibodies may cause anomalous results in ABO reverse typing requires further consideration.

P483 | The presence of anti-Lewis antibodies predicts undetectable levels of serum CA19-9

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Background: The Lewis blood group is determined by *FUT2* and *FUT3* genes. Individuals with the Lewis-negative phenotype (Le(a-b-)) cannot express the Le(a) and Le(b) antigens, which are precursors of the sialyl Lewis A (sLea) antigen, foundational to the CA19-9 antigen. Anti-Lewis antibodies occur exclusively in Lewis-negative individuals. Therefore, we hypothesized that the presence of anti-Lewis antibodies predicts undetectable levels of serum CA19-9.

Aims: To investigate this hypothesis, we compared serum CA19-9 levels between groups with anti-Lewis antibodies and those with other antibodies.

Methods: From May 2023 to January 2024, samples ($n = 34$) with detected anti-Lewis antibodies underwent testing for Lewis phenotype, *FUT2* & *FUT3* genotyping, and serum CA19-9 measurement. Additionally, serum CA19-9 levels were measured in samples ($n = 14$) with unexpected antibodies other than Lewis identified in pre-transfusion tests. Target-specific PCR primers, designed in-house, targeted c.358A>T in the *FUT2* gene and c.59T>G in the *FUT3* gene, using NM_0005113.6 (*FUT2*) and NM_001097639.3 (*FUT3*) as reference sequences based on the GRCh19 reference genome. The PCR results were analyzed through melting curve analysis. Quantitative serum CA19-9 levels were measured using the Electrochemiluminescence Immunoassay (ECLIA) method, with a Limit of Quantitation (LOQ) of 2.0 U/mL.

Results: In the anti-Lewis antibody positive group ($n = 34$), serum CA19-9 levels were below the LOQ (< 2.0 U/mL) in 32 cases (94.1%). Among them, 30 cases showed anti-Le(a) antibodies, one showed both anti-Le(a) and anti-Le(b), and one showed anti-Le(b) with anti-E antibodies. Two cases with serum CA19-9 levels higher than the LOQ showed 3.65 U/mL and 10.4 U/mL, with anti-Le(a) and -Le(b) antibodies detected, respectively. The genotype of the case with anti-Le(a) was homozygous for the 385A allele (385A/A) in *FUT2* gene and homozygous for the 59G allele (59G/G) in *FUT3* gene. The genotype of the case with anti-Le(b) was homozygous for

the 385T allele (385T/T) in *FUT2* gene and heterozygous c.59T>G in *FUT3* gene, indicating a weak secretor phenotype with normal CA19-9 production function. In the other antibodies group ($n = 14$), various unexpected antibodies were detected in pre-transfusion tests, including anti-E, E&c, P1, M, Jk(b), and unidentified. Their serum CA19-9 levels ranged widely from 4.53 U/mL to 4261.0 U/mL (median level: 9.05 U/mL).

Summary / Conclusions: The findings of this study suggest that in cases where anti-Lewis antibodies (specifically, anti-Le(a)) are detected, CA19-9 levels are likely to be below the detection limit in the vast majority of cases. Anti-Le antibodies have significance not only as unexpected red blood cell antibodies but also can provide additional information to patients that serum CA19-9 may yield a false-negative result.

P484 | 5 years of experience with anti-CD38 fab fragments in blood compatibility testing -group of 140 patients with multiple myeloma and anti-CD38 monoclonal antibodies

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Background: Panreactivity in blood compatibility testing is a consequence of plasma samples' treatment with anti-CD38 monoclonal antibodies (mAbs) and it avoids the correct interpretation and detection of alloantibodies in Multiple Myeloma (MM) patients. For this reason, the Spanish Society of Blood Transfusion (SETS) protocolised treating erythrocytes with dithiothreitol (DTT) and characterising the extended erythrocyte phenotype in order to guarantee transfusion safety in these patients. In the Blood Transfusion department of the General University Hospital Virgen de las Nieves, DTT had been used until 2018 when it was replaced by anti-CD38 fab fragments (DaraEx[®] and afterwards, DaraEx plus[®]). This method became the most commonly used to mitigate this interference, especially in relation to indirect Coombs test, because it neutralizes CD38 in erythrocyte surface and agglutination by anti-CD38 mAbs is prevented. It is used through BioVue column agglutination technology.

Aims: To support the use of red blood cells treated with anti-CD38 fab fragments for the identification of alloantibodies.

Methods: Descriptive and retrospective research of 140 patients with MM who have been treated with anti-CD38 mAbs (daratumumab or/and isatuximab). Demographic, clinical and immunohaematology data was collected through medical history records and subsequently processed through computer programmes: *Milenio*, *REDCap* and *Excel*.

Results: 744 irregular antibodies screenings (IAS) and 587 cross-matching tests (CMT) were registered. Anti-CD38 fab fragments were used in 84% of IAS and in 90.63% of CMT with 91.84% and 99.4% of negative results. In 5 patients the IAS showed positive results identifying 4 alloantibodies already known. These results are similar to those of smaller series (total number of patients: 19), with negative results

in 93.1% and the detection of alloantibodies in 8 out of 19 patients. In our cohort, Rh and Kell erythrocyte phenotype and extended phenotype were determined in 85% of the patients: only half required red blood cell transfusion and only 40.6% complete phenotype was respected. Lewis erythrocyte system was the most common cause of blood incompatibility. Alloimmunization risk in patients with MM treated with anti-CD38 antibodies is generally low. Some researches describe alloimmunization rates around 0-3% without statistical differences between ABO-Rh compatible transfusions and extended phenotype compatible transfusions. In our cohort, only 4 patients (2.85%) developed alloantibodies which appeared outside the period of treatment with anti-CD38 mAbs.

Summary / Conclusions: The use of anti-CD38 fab fragments represents a valid, fast and safe method to effectively eliminate the interference caused by anti-CD38 mAbs, improving the detection of alloantibodies. It reduces the number of cross-matching tests as well as extended erythrocyte phenotypes when the IAS shows a negative result. In addition, it requires minimal complexity for laboratory staff and can be performed during any work shift. Nevertheless, despite its advantages compared to other techniques such as DTT, clinical experience with this reagent is scarce and further investigation is needed.

P485 | Description of discrepant Rh(D) phenotypes and their correlation with genotype in a tertiary care Spanish hospital

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Background: RhD antigen may show a weakened expression due to variant RHD alleles: weak, partial or DEL variants. Around 1%-2% of Caucasians carry RHD alleles responsible for an inconclusive D phenotype. RHD genotyping allows considering D variants as D+ (weak D type 1/2/3/4.0/4.1) or D- in terms of transfusion, pregnancy and RhIG prophylaxis, based on immunization risk (Flegel, Transfusion, 2020).

Aims: To analyse the results of the patients studied in the immunohematology laboratory of a hospital in the Basque Country (Spain) for serological discrepant RhD typing, and to correlate serologic reactivity with genetic background.

Methods: Since February 2014, RhD grouping is performed in our hospital using Gel ABO/Rh (2D) cards (Grifols®) in Erytra automated analysers. From February 2014 to December 2023 (included), 143 patients had discrepant reactivity (<4+ with at least 1 of the 2 anti-D antisera) and were genotyped with Progenika platforms at immunohematology reference laboratory, following initial manufacturer's instructions. Statistical analysis was performed with Stata.

Results: Patients' clinical characteristics are shown in Table 1. The most frequent D variant was weak D (WD) type 59, with 40 (28%)

P485 - Table 1. Clinical characteristics of patients with D variants.

	n = 143	Median (range)/%
Age	143	32 (0-70)
Females	131	91.6
Prior pregnancy	54/131	41.2
Number of prior pregnancies	54/131	1 (1-5)
Distribution of patients:		
Pregnant women	109	76.2
Women looking for pregnancy	10	7
Newborns	4	2.8
Potential transfusion receptors	20	14
Prior transfusion with RhD+ red blood cells	6	4.2
Number of units	-6/6	-2 (2-10)
History of allo-anti-D	0	0

P485 - Table 2. Distribution and serologic reactivity of D variants.

	Total population n = 143 n (%)	Serologic reactivity ≥ 3 n = 65 n (%)
Weak D type:		
1	30 (21)	8 (12.3)
2	17 (11.9)	2 (3.1)
3	17 (11.9)	17 (26.2)
4.0	5 (3.5)	5 (7.7)
4.2	2 (1.4)	1 (1.5)
4.2.2 ^a	7 (4.9)	7 (10.8)
4.2.3	1 (0.7)	1 (1.5)
4.0/4.3	1 (0.7)	1 (1.5)
Other type 4 cluster variants	5 (3.5)	4 (6.2)
59 ^b	40 (27.9)	11 (16.9)
61	1 (0.7)	1 (1.5)
Partial D type:		
VI type 2	5 (3.5)	0 (0)
VII type 1	1 (0.7)	1 (1.5)
Heterozygous DIIIa-ceVS.03 with DMH	1 (0.7)	0 (0)
DEL type ^c	3 (2.1)	2 (3.1)
RhD+ (heterozygous RHD*01 with DEL)	1 (0.7)	1 (1.5)
RhD-	1 (0.7)	0 (0)
Non-identified D variants (different to weak D type 1/2/3)	5 (3.5)	3 (4.6)

^a 1 patient: Weak D type 4.2.2 allele heterozygous with pseudogene.

^b 3 patients: Weak D type 59 in heterozygosity with DEL type.

^c 1 patient: DEL type allele heterozygous with DAU-4.

cases. Sixty-five (45.5%) patients showed serologic reactivity $\geq 3+$ with both anti-D reagents (Table 2). Within this group, WD types 1, 2, 3 or heterozygous RhD+ (RhD+ set) were more frequent than other variants excluding WD type 59 (51.9% vs. 48.1%; $p = 0.013$), and RhD+ set including WD type 4.0 had more prevalence compared to WD type 59 (75% vs. 25%; $p = 0.043$). There were no other statistical associations between genotype and serologic reactivity (data not shown). CeCe/Cece phenotype was associated to WD type 1 (90% vs. no C 10%; $p = 0.046$), type 3 (100% vs. no C 0%; $p = 0.014$) and type 59 (100% vs. no C 0%; $p < 0.001$), cEce to WD type 2 (88.2% vs. no E 11.8%; $p < 0.001$), and cece to type 4 cluster variants (61.9% vs. C/E 38.1%; $p < 0.001$).

Summary / Conclusions: Our results show that D variants historically managed as D+ have a strong serologic reactivity more frequently than others. In our population, there is a high prevalence of WD type 59, which has been rarely reported in European populations. This arises new questions on its management for immunization risk. Extended RhD phenotype may guide suspicion for specific D variants.

P486 | Establishment of an absolute neutrophil count reference range in Saudi Arabian Duffy-null healthy blood donors

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Background: The Duffy (FY) blood group is the eight blood group system according to the terminology of the International Society of Blood Transfusion (ISBT). Two main antigens among this system, Fy^a and Fy^b. FY-null individuals, lacking the Fy^a and Fy^b antigens, have resistance to *Plasmodium Vivax* and *Plasmodium Knowlesi*. Interestingly, this phenotype is most often found among those of African and Arab ancestry. FY-null individuals have significantly lower absolute neutrophil counts (ANC) than FY non-null individuals, a phenomenon

previously called “benign ethnic neutropenia”. ANC reference ranges influence clinical decisions including clinical trial eligibility, medication dosing, and invasive investigations for spurious “neutropenia”. The ANC range of healthy FY-null individuals with Arab ancestry has not previously been established.

Aims: As a part of an international collaboration, we aimed to establish an accurate ANC reference range in FY-null Saudi Arabians through analyzing samples from healthy blood donors. Furthermore, the ANC of the FY-null was compared with the FY non-null.

Methods: We previously demonstrated a FY-null prevalence of 78.32% in Jazan Province of Saudi Arabia. A total of 168 healthy Saudi Arabian volunteer blood donors at King Fahad Central Hospital in Jazan Province were enrolled. FY serotyping was performed by gel card technology using ID-Card Fy^a/Fy^b and ID-Anti-Fy^a/Fy^b antibodies (DiaMed GmbH, Cressier, Switzerland) according to the manufacturer's instructions. ANC was determined by complete blood counts on Sysmex XN-10 analyzer (Sysmex, Kobe, Japan). A non-parametric central 95% reference range was established per Clinical and Laboratory Standards Institute guidelines. Statistical significance testing was undertaken using Mann-Whitney U Test.

Results: One-hundred-fifty (89.29%; $n = 150/168$) individuals in the cohort were serotyped as FY-null; Fy(a-b-). ANC values by FY status is shown in Table 1. The central 95% ANC reference range for the FY-null cohort is 1210–4990/ μ L with a median of 2480/ μ L. There is a significant difference in ANC between the FY-null and FY non-null samples ($p < 0.01$).

Summary / Conclusions: We report a novel FY-null ANC reference range among people of Arab ancestry. Our ANC range has striking concordance with a recently-published range of healthy FY-null African Americans (1210/ μ L to 5390/ μ L) “Merz, JAMA, 2023”. The current ANC reference range used in Jazan Province of Saudi Arabia is 2500–7500/ μ L, which needs review in the light of these results. Our results emphasize that FY status, rather than ethnicity, is the key determinant of a healthy ANC reference range. Efforts should be made to globally utilize FY-null specific ANC reference ranges to accurately report expected ANC. Furthermore, analysis of blood donor populations is an excellent method to report prevalence of FY-null in communities and verify FY-null ANC reference ranges.

P486 - Table 1. Absolute neutrophil count (ANC) for FY-null and FY non-null Saudi Arabian blood donors.

	FY-null (n = 150)	FY non-null (n = 18)
Demographics		
Age (years)	29.5	29.5
ANC (/ μ L)		
Central 95%	1210-4900	N/A
Range	930-6180	1670-7350
1st Quartile	1850	3710
Median	2480	4250
3rd Quartile	3050	5140

P487 | Accuracy of solid phase method for the detection of RBC antibodies of transfusion relevance

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Background: Solid phase red cell adherence (SPRCA) is a highly sensitive method applied for the screening and identification of red blood

cell (RBC) irregular antibodies as well as for weak-D phenotype detection. The methodology can be performed under secure automation and selected by blood banks for the immunohematological routine of both blood donors and patients. Considering the high sensitivity of solid phase, concerns might rise referring to the risk of false positive results. In this scenario, determining the specificity and accuracy of SPRCA for antibody screening is important, as well as determining if the positive results represent antibodies of transfusion relevance.

Aims: (1) To evaluate the accuracy of solid phase for antibody screening and identification using samples of multitransfused patients, mainly with Sickle cell disease (SCD); (2) To determine the clinical relevance of antibodies of undetermined specificity (AUS) detected in SPRCA using the Monocyte Monolayer Assay (MMA).

Methods: A comparative study was conducted comparing SPRCA (Capture, Echo Lumena®, Werfen, Barcelona) and gel test (Erytra Eflexis®, Grifols, Barcelona) for antibody screening and identification. Samples from previously investigated patients presenting with irregular antibodies directed to the most immunogenic blood group systems (Kell, MNS, Rh, Duffy, Kidd, Diego) were included in the analysis and tested in both assays (gel and SPRCA). In parallel, samples from patients without irregular antibodies were also tested. In case of AUS or antibodies detected only in solid phase, MMA was performed.

Results: Eighty samples were included in the study. In the group of patients with positive irregular antibody screening and identification ($n = 60$), 59 (98.3%) presented the same specificity of alloantibodies identified in both methodologies. One patient presented anti-E and anti-c in gel method (strength of agglutination 3+), but anti-E was not detected by SPRCA (IgM class). In the group of patients without RBC alloantibodies ($n = 20$), there was 100% concordance between SPRCA solid phase and gel method. Among all samples studied, three presented in both method antibodies of unknown specificity (AUS) after extensive serological workup. For these patients, RBC units were selected for transfusion based on extended-phenotype compatibility and both indirect antiglobulin test (IAT) crossmatch and MMA was performed. IAT crossmatch resulted incompatible in both SPRCA and gel test. Monocyte index resulted less than 5% in one sample and more than 5% in two samples (67%), the latter being considered as of clinical relevance. Interestingly, the intensity of agglutination observed in IAT-crossmatch using SPRCA correlated with MMA monocyte index. Also, in the cases in which anti-Jk^a ($n = 3$) or anti-Di^a ($n = 2$) were identified, the intensity of agglutination was significantly higher in SPRCA. Even though SPRCA detects IgG-class antibodies, in two cases IgM antibodies (anti-M and cold autoantibody) were detected by the method indicating the antibodies possessed an IgG component.

Summary / Conclusions: SPRCA method presented high accuracy in RBC antibody screening and identification, with no observed false positive results. Higher agglutination strength was observed in SPRCA for some antibody specificities, such as anti-Jk^a and anti-Di^a, making the identification easier in cases of multiple antibodies. Among the antibodies of undetermined specificity identified, 67% had predicted *in vitro* transfusion relevance.

P488 | IgG antibody screening assays with an automated sample dilution

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Background: Alloantibodies are clinically relevant as they contribute to reduced red blood cell survival due to hemolytic transfusion reactions, hemolytic disease of the fetus and newborn (HDFN), or autoimmune hemolytic anemia. In vitro antibody detection tests (screening) are used to detect the presence of these antibodies in patient and donor sera or plasma. To meet regional specific requirements, it is necessary to test diluted patient or donor plasma or serum as an additional secondary test. Considering the labour-intensive and error-prone nature of manual methods, which considerably reduces the reproducibility of results, an automated solution is highly needed. To enable customers to screen diluted serum or plasma samples for unexpected IgG antibodies against red blood cells as part of testing procedures, Werfen has developed a set of fully automated screening assays with an automated 1:10 or 1:50 sample dilution for NEO Iris® and NEO® (v2.0).

Aims: The aim of this study was to establish and evaluate the performance of a set of automated dilution screening assays compared to semi-automated method for an IgG screening.

Methods: Werfen has developed two fully automated assays to screen diluted serum or plasma samples for unexpected IgG antibodies against red blood cells. A 1:10 or 1:50 dilution is prepared automatically by the instrument. The assays are based on solid phase Capture® technology for the NEO Iris® and NEO® (v2.0) automated platforms. Patient, pregnancy and donor samples with different known alloantibodies as well as the 3rd WHO international standard for anti-D immunoglobulin as a reference were tested. All samples were analyzed to determine result concordance of the two fully automated 1:10 or 1:50 sample dilution screening assays to manual 1:10 or 1:50 sample dilution, respectively, followed by automated IgG antibody detection on another IgG screening assay without automated dilution.

Results: In total, 209 samples (total number of tests: $209 * 3 = 627$ tests, as each sample is tested with three different screening cells) were tested on both assays that diluted the samples on the instrument and compared to results obtained from manual 1:10 or 1:50 sample dilution, respectively tested on another IgG screening assay. For both dilution screening assays, all analyzed samples within this study showed 100% concordance and achieved 99.5% agreement at the 95% lower bound confidence interval after retesting and excluding persistent equivocal results.

Summary / Conclusions: Automated screening assays for the detection of IgG alloantibodies with sample dilutions of 1:10 and 1:50 enable efficient and accurate detection in transfusion medicine and fulfill the region-specific requirements as an additional method.

P489 | Enhanced automated blood group antibody titration techniques—an exploration of the advantages

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Background: ABO and non-ABO antibody titre determination is important in many clinical decisions. The automated titration technique has several advantages over traditional manual methods. Firstly, it significantly reduces the hands-on time required for performing titration assays, allowing laboratory personnel to focus on other critical tasks. Secondly, automation eliminates human errors and variability, resulting in more reliable and reproducible titration results. Thirdly, the technique enables high-throughput analysis, allowing for the simultaneous processing of multiple samples, thereby increasing the overall efficiency of antibody titration. A first generation of automated titration methods was launched using the Erytra and Erytra Eflexis systems and gel card column agglutination. In the commitment of continuous improvement, a new enhanced set of protocols has been designed on the same instruments. The last version methods ensure minimal sample and diluent consumption and offer high flexibility; allowing a 2-fold serial dilution up to a 1:2048 endpoint as well as the use of a flexible range selection.

Aims: The aim of the study was to compare sample and diluent consumption as well as the throughput between the improved automated Erytra and Erytra Eflexis antibody titration technique and the previous available technique, for ABO and non-ABO antibodies.

Methods: Plasmas containing ABO and non-ABO antibodies were titrated between 1:1 (undiluted sample) and 1:128. For the enhanced automated technique, the dilutions were performed in a 2-fold serial manner. For the previous technique, individual dilutions are prepared; each endpoint 1:2, 1:4, 1:8, 1:16, 1:32, 1:64 and 1:128 were developed from the mother sample. As the previous technique allowed only dilution till 1:128, it was not possible to perform the comparison for upper endpoints. For both techniques, and for ABO and non-ABO antibody titration, sample and diluent consumption, including additional volume due to automation, has been reported. Throughput, including pipetting process, incubation and centrifugation times of gel card, was monitored for regular antibodies titration in DG Gel Coombs. Additional benefits that could improve the end-user's work were noted.

Results: Through the selection of the new technique, the use of diluent and sample has been reduced by 60% and 70% respectively. The minimized plasma usage, preserved valuable sample volume for further testing and enabled titrations with small sample volumes. The dilution of the samples was performed from centrifuged whole blood tubes and using the dilution cup of the instrument, with no additional plastic waste generated by auxiliary disposables. In terms of

throughput, the 1:1 to 1:128 ranged titration allowed to maintain the same throughput, having a turnaround time of 32 minutes per sample per 12 wells.

Summary / Conclusions: The enhanced titration techniques performed on Erytra and Erytra Eflexis systems offer tailored antibody titration solutions up to 1:2048 dilution for both regular and irregular antibodies, IgG or IgM, to be performed in Coombs or saline media at 37°C or RT. In addition to their accuracy, efficiency, and scalability, they allow to use primary tubes for the testing, ensure a very low sample and diluent consumption and enable flexible ranges dilution selection while minimizing plastic waste.

P490 | Positive direct antiglobulin test in chronic myeloid leukaemia patients treated with Imatinib

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Background: Chronic Myeloid Leukemia (CML) patients can experience autoimmune hemolytic anemia (AIHA) in the course of the disease. Autoimmune hemolytic anemia is characterized by positive direct antiglobulin test (DAT) results. One of the risk factors for AIHA in CML is the administration of Imatinib therapy, a tyrosine-kinase inhibitor drug. Imatinib is thought to influence the immune response, causing disruption of the Th1 and Th2 balance in CML patients, which is the basis for AIHA.

Aims: To determine the frequency of positive direct antiglobulin test in Chronic Myeloid Leukemia patients receiving Imatinib therapy.

Methods: Observational descriptive study of 30 Chronic Myeloid Leukemia patients treated at RSUP Dr. M. Djamil Padang Hospital. Inclusion criteria were CML patients who experienced hemolytic anemia characterized by increased reticulocytes. The direct antiglobulin test is carried out using the column agglutination method. This study was approved by the local ethics committee.

Results: A total of 30 Chronic Myeloid Leukemia patients in this study had a mean age of 44 (14.5) years, 12 male patients (40%) and 18 female patients (60%). Imatinib therapy was obtained in 11 patients (36.7%). After carrying out the direct antiglobulin test, 6 patients (54.5%) had positive DAT and 5 patients (45.5%) had negative DAT.

Summary / Conclusions: In this study we found that more than half of CML patients receiving Imatinib therapy experience autoimmune hemolytic anemia which is characterized by positive direct antiglobulin test.

P491 | Abstract withdrawn

P492 | Abstract withdrawn

P493 | Laboratory features and clinical association of Antibodies of Undetermined Specificity (AUS) encountered at reference immuno-hematology laboratory, National Blood Transfusion Service of Sri Lanka

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Background: Reference Immuno-hematology laboratory at National Blood Centre, receives all requests with unresolved red cell serology problems in pre-transfusion and antenatal testing from all-island blood banks. There is a significant number of cases with AUS, after ruling out antibodies against red blood cell antigen systems namely Rh, Kell, Duffy, Kidd, MNSs, Lewis and P. There is no information about AUS in Sri Lankan population.

Aims: To determine the frequency and laboratory features of AUS reacting at 37°C IAT (indirect antiglobulin test) and to analyze the relationship to sensitizing events of the patients.

Methods: We retrospectively collected data of all cases with AUS for 3 months period, between October to December 2021 using the information on request forms and reference lab work sheets. Data analyzed using an excel sheet.

Results: A total of 1074 samples had antibodies, among 1372 samples received during the study period. In 983 (91.5%) samples, antibody specificities were defined. AUS was reported in 91 (8.5%) samples.

Analyzing the laboratory features as per reactivity of AUS among 11 cell identification panel demonstrated that the highest number of cases ($n = 40$, 43.9%) reacted with seven or more cells, while 8 (8.8%) reacted with five-six cells, 22 (24.2%) reacted with three-four cells, and 21 (23.1%) reacted with two cells or less, by LISS (low ionic strength solution) 37°C IAT. Reaction strengths in the majority ($n = 60$, 65.9%) were 1+ or weaker, and the rest were 2+ reactions in 3 (3.3%), 3+ reaction in 2 (2.2%) with no case having 4+ reactions. Variable reaction strengths were seen in 21 (23.1%). Relationship to sensitizing events showed that 12 (13.2 %) cases had no sensitizing events, while there were transfusion histories in 6 (6.6%), pregnancy histories in 52 (57.1%), both transfusion and pregnancy histories in 12 (13.2%). History of sensitizing events was unavailable in 9 (9.9%).

Summary / Conclusions: Since 76.9% of AUS cases had sensitizing event, they may represent developing clinically significant antibodies highlighting the importance of confirmatory follow up testing. AUS may also be clinically insignificant high frequency low affinity antibodies as majority (65.9%) were 1+ or weaker reactions against seven or more cells (43.9%) in LISS IAT, possibly formed against unidentified or non-RBC antigens. Thus, further investigations are necessary to differentiate all these possibilities from the known problem of LISS IAT technique with false positivity or enhancement of weak autoantibodies.

P494 | Irregular antibodies screening in University Hospital Complex of the Canary Islands (CHUC)

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Background: The determination of alloantibodies is of utmost importance in conducting pre-transfusion tests as it enables us to select the optimal blood component for a recipient and minimize adverse effects. Through erythrocyte phenotyping, we aim to mitigate the formation of other antibodies that may hinder compatibility in transfusion.

Aims: To identify the number and type of irregular antibodies in patients registered in the Blood Bank according to current clinical guidelines.

Methods: A quantitative retrospective observational study conducted from January to November 2023 on patients typed with positive irregular antibodies not previously registered in the CHUC Blood Bank. Epidemiological and clinical characteristics were gathered through Electronic Health Records. Variables included in this study comprise the number of newly sensitized patients, percentages of identified antibodies, sex, age range, and phenotyping prior to transfusion.

Results: During this period, 11,299 analyses were received, detecting 199 antibodies not previously registered in the Blood Bank

P494 - Table 1: Detected Antibodies.

Anti D 32	26.1%
Anti-D via Gamma	20
Undetermined	31.15.1%
Anti E 27	13.6%
Anti C. 17	8.5%
Anti c. 11	5.5%
Anti K. 10	5%
Anti Jka. 8	4%
Anti Fya. 8	4%
Anti P1. 7	3.5%
Anti Lea. 6	3%
Anti M. 5	2.5%
Anti Fyb. 4	2%
Anti Cw. 3	1.5%
Anti Jkb. 2	1%
Anti e. 2	1%
Anti Leb. 2	1%
Anti Lua. 2	1%
Anti N. 1	0.5%
Cold agglutinins 1	0.5%

(1.77%) among 159 patients, with 103 being female (64.7%) and 56 male (35.2%). 65.9% were over 60 years old (range 61-95) compared to 33.9% under 60 years old (range 15 to 60). Erythrocyte phenotyping was performed on 127 patients compared to 32 patients. The most prevalent antibodies were: Anti-D (26.1%), Anti-E (13.6%), Anti-C (8.5%), Anti-c (5.5%), and Anti-K (5%). Antibodies, including Anti-Fya, Anti-Jka, Anti-P1, Anti-Lea, Anti-M, Anti-N, and Anti-Cw, occurred at a frequency less than 5% as described in Table 1. Among the detected Anti-D (52 patients), 20 were from pregnant women due to Gamma globulin, 1 Rh-negative patient with uncontrolled pregnancy, 24 patients with previous transfusions not recorded in our history, and 7 Rh changes in Rh-negative patients. 15.1% of alloantibodies could not be detected by our techniques (31 patients).

Summary / Conclusions: There are no significant differences in the detected alloantibodies compared to those commonly found in the literature. The high number of patients with positive irregular antibodies over 60 years old is possibly due to the CHUC being the reference hospital for the North Area of Tenerife, characterized by an aging population. The unidentified antibodies in our bank require further investigation, though it is not feasible to have reagents for all known groups. It is observed that the female sex is more susceptible to sensitization, nearly double that of males.

P495 | Erythrocyte alloimmunization in chronic kidney disease patients with repeated blood transfusions

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Background: Chronic kidney disease (CKD) patients are at risk for repeated erythrocyte transfusions, which raises the risk of alloimmunization. Erythrocyte alloimmunization in CKD leads to difficulties in finding suitable blood for transfusion and can cause hemolytic transfusion reactions.

Aims: The aim of the study was to determine the prevalence of erythrocyte alloimmunization and evaluate the correlation with its risk factors in CKD patients who received repeated blood transfusions at Dr. M. Djamil Hospital, Padang.

Methods: This was a cross-sectional analytical study conducted at the Laboratory and Blood Transfusion Unit of Dr. M. Djamil Hospital, Padang. Adult patients diagnosed with stage V CKD who have had at least three transfusions will undergo a Coombs' test using the gel agglutination method to screen for the presence of erythrocyte alloimmunization. Erythrocyte alloimmunization is determined based

on the presence of alloantibodies from the positive indirect Coombs' test. Demographic and clinical data were taken from the patients' medical record. The study was conducted and approved by the local ethics committee.

Results: This study included 60 CKD patients which 23 (38.3%) were males and 37 (61.7%) were females. The average age was 50.7 ± 14 years old and the most common blood type was O (36.7%). The erythrocyte alloimmunization rate was found to be 11.7%. There was a significant correlation between the total blood unit ($p = 0.001$) and duration ($p = 0.001$) of transfusion with the development of erythrocyte alloantibodies. Patients with alloimmunization were found to have a higher number of blood transfusion units (12.3 ± 2.3 units) than patients without alloimmunization (6.9 ± 2.5 units). Most patients with alloimmunization received blood transfusions for more than 5 years.

Summary / Conclusions: CKD patients who received repeated blood transfusions were at risk of alloimmunization. The total blood unit and duration of transfusion were correlated with the development of erythrocyte alloantibodies.

P496 | Red cell engraftment in spite of persistent anti-A titer after pure red cell aplasia

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Background: In allogeneic hematopoietic stem cell transplantation (HSCT), suitable donors are selected according to their HLA type, whereas ABO is less important. A major ABO mismatch can lead to pure red cell aplasia (PRCA) with prolonged transfusion requirements due to persisting post-transplant anti-ABO isoagglutinins.

Aims: We report a case of prolonged PRCA. Unusual is that the onset of donor erythropoiesis was detectable at a residual anti-A titer as high as 64.

Methods: Blood samples were collected from 8 days before until 427 after HSCT. All analyses were performed in accordance with manufacturer's instructions. Testing included ABO blood typing before and after allo-HSCT, antibody screening (indirect antiglobulin test (IAT) (BioRad, Switzerland)) and direct antiglobulin test (DAT) (BioRad, Switzerland). The isoagglutinin titers were tested in the IAT (BioRad, Switzerland). RBC-bound antibodies were eluted by using the standard acid elution method (BAG, Lich, Germany).

Results: A 67-year-old male patient underwent an allo-HSCT for high-risk MDS. The patient's blood group was typed O, the donor was A+ (ABO major mismatch). The patient's anti-A1 titer was found to be 4096 prior to allo-HSCT, the donor's A subtype was

A1. The high anti-A1 titer persisted for the next 6 months after allo-HSCT and no donor erythropoiesis was detected. Due to the high transfusion requirement (4 units of red blood cells per month), the patient received rituximab six and nine months after HSCT. First A type red blood cells indicating the beginning of donor erythropoiesis were observed 11 months after allo-HSCT. At that time, the recipient's anti-A1 titer still was 64, but no further transfusions were required. Approximately 100 days later, only A+ red cells were observed with still detectable anti-A1 (titer of 4). Elution of red cells due to a positive DAT (2+) showed anti-A1. Cross-matching with A+ red cells were incompatible, cross-matching with O+ red cells was compatible. Currently, the patient does not require transfusions and therefore no further therapy for PRCA, and his anti-A1 titers will be monitored at longer intervals.

Summary / Conclusions: Patients undergoing ABO major incompatible allo-HSCT should be monitored for possible PRCA. In addition, the patient needs an emergency card with information about allo-HSCT and transfusion recommendations. Without this, it will be difficult for blood bankers outside of the transplant center to provide suitable blood units.

P497 | Association between ABO blood groups and clinical outcome of COVID-19 among patients admitted at Eka Kotebe General Hospital, Addis Ababa, Ethiopia

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Background: The Corona Virus Disease of 2019 is an emerging infectious disease outbreak that was later declared a global pandemic in 2020. It is caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). The COVID-19 virus can cause a disease that is asymptomatic or deadly. Up until now, it has resulted in millions of deaths globally. Among the countries with the highest number of COVID-19 cases is Ethiopia. The association between viral illness prognosis and blood type distribution has attracted the attention of many researchers. It is hypothesized that the presence of certain ABO blood groups may be indicative of an increase in the incidence of poor clinical outcomes among COVID-19 patients. However, there has been no specific study conducted on the association between blood group and COVID-19 clinical outcomes in the local context so far. As a result, this study was conducted.

Aims: Was to assess the association between ABO blood group and COVID-19 clinical outcome defined by recovery, intubation, and death among patients admitted at Eka General Hospital, Addis Ababa, Ethiopia.

Methods: A hospital-based cross-sectional study was conducted using a systemic random sampling technique, and data were collected using the ODK data collection tool. Descriptive statistics such as frequency,

percentage, mean, standard deviation, median, and interquartile range were used to summarize the results. A multinomial logistic regression model was used to assess the association between the blood group and the clinical outcome of COVID-19. The basic assumptions and the adequacy of the model were evaluated. A multi-variable analysis was performed to assess and screen out significant independent variables. A P-value ≤ 0.05 was considered a statistically significant association.

Results: In the study, 355 patients' medical records were included. According to the study, blood group O predominates with a magnitude of 37.5% (133). The study found that the magnitude of death for COVID illness in patients with blood groups A, B, O, and AB was 23.6% (29), 22.7% (17), 22.6% (30), and 12.5% (3), respectively. whereas, among patients who recovered, 75% (18), 72% (54), 66.9% (89), and 60.2% (74) had blood types AB, B, O, and A, respectively. Meanwhile, the incidence of intubated patients in A, AB, O, and B is 16.3% (20), 12.5% (3), 10.5% (14) and 5.3% (4), respectively. However, there was no association between the ABO blood group and the clinical outcome of the COVID-19 illness ($p = 0.277$). On the other hand, the study showed that intubation, pregnancy, the presence of complications, and comorbidities were statistically significant in COVID-19-related deaths.

Summary / Conclusions: The study did not show a significant association between ABO blood groups and COVID-19 disease clinical outcome.

P498 | Anti-erythrocyte alloimmunization in sickle cell disease in Northern Tunisia

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Background: Sickle cell disease (SCD) is a public health problem in Tunisia, with a frequency of 1.9%. Transfusion support is essential in the management of SCD with risks of alloimmunization and post-transfusion hemolysis.

Aims: The objective of this study was to analyze anti-erythrocyte alloimmunization in patients followed for SCD.

Methods: Is a cross-sectional study from April 2021 to 31 January 2024. A patients followed regularly for SCD with at least one testing for irregular anti-erythrocyte antibodies screening (IEAS) were included. Epidemiological (age, gender), number of packed red blood cell (PRBC) transfused and IEAS results were noted.

Results: 141 patients were enrolled. The median age was 35.5 years [18-76]. The gender ratio was 0.58. 1043 PRBC were transfused during this period with mean patient 7.7 [1-16]. The immunization rate was 37% (one or more antibodies). 56% of these patients were immunized against non Rh-Kell antigens and 23% had antibodies against Rh-Kell antigens. A combination of antibodies directed against both Rh-Kell and non Rh-Kell systems was noted in 17% of patients. 4% had antibodies to unknown specificities. During the study period,

patients were transfused with more than 10 PRBC, less than 10 PRBC or non transfused in respectively 17%, 40% and 43%. There was no statistically significant association between immunization and PRBC consumption rate ($p = 0.35$). The rate of alloimmunisation in SCD (37% vs 3.8%) is higher than in the south of Tunisia, although the number of transfusion was lower (7.7 vs 17.9).

Summary / Conclusions: This could be explained by genetic mixing in the north of Tunisia. Prevention by transfusion of phenocompatible PRBC FY, JK, MNS could improve the transfusion outcome of patients. But, the feasibility remains a big challenge economically and regarding blood donors.

P499 | The frequency of ABO, Rh, Kell and Duffy blood group antigens in a homogeneous ethnic Nigerian population

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Background: Blood group antigens are inherited macromolecules present on red cells and capable of eliciting specific immune response in persons lacking the antigen. The distribution of red cell antigen phenotypes has been demonstrated to vary between populations and ethnicities. Knowing patients' blood group phenotypes enables the selection of matching cellular blood products to avoid alloimmunisation particularly among the multi-transfused and pregnant women. While the frequency of various blood group phenotypes is well characterised in the developed countries, data about the distribution of these phenotypes appears to be limited in Africa.

Aims: This study aimed to determine the frequency of blood group antigens from four systems (ABO, Rh, Kell and Duffy) among patients of a homogeneous ethnic background attending a healthcare facility.

Methods: We recruited 100 consenting patients from those attending ABUAD Multi-System Hospital, Ado Ekiti within the study period. Study participants consisted of patients attending the dialysis clinic ($n = 55$) and those visiting the antenatal clinic ($n = 45$). 5ml of venous blood was drawn from study participants into K2 EDTA tubes. For each sample, a 2%–3% suspension in isotonic saline was prepared and tested qualitatively by tube technique for the presence or absence of ABO, D, C, c, E, e, K, k, Fy^a and Fy^b antigens according to the manufacturer's instructions (Lorne Laboratories Ltd., UK).

Results: The overall mean age of the study population was 40.8 ± 15.3 . The most prevalent ABO group was O (69%), followed by groups A and B (18% and 12% respectively), while group AB is the least common (1%). For the Rh antigens, 94%, 20%, 89%, 24%, and 98% were positive for D, C, c, E, e antigens respectively. Only 8% were K positive while 79% were k (cellano) positive. The most

prevalent Kell phenotype observed in this study was K-k+ (76%). All the study participants tested negative for the Fy^a and Fy^b.

Summary / Conclusions: Evaluating the frequencies of blood group antigens in a population help blood service establishments and health-care service providers to plan and provide blood group antigen compatible blood products to patients who need them. This will minimize the potential risk of red cell alloimmunisation among those who are transfusion-dependent and maternal alloimmunisation among women of childbearing age.

P500 | A cross-sectional study conducted in a center dedicated to thalassemia at Bangladesh on the development of red cell alloantibodies in transfusion dependent patients who are transplantable

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Background: Red cell antibody clinical importance is influenced by both their frequency and their ability to cause damage in vivo. Migration has caused hereditary hemoglobin diseases, such as thalassemia, to become more prevalent in many low-income nations where they were previously unknown. The most prevalent hereditary condition, thalassemia, impairs the synthesis of one or more hemoglobin components, which prevents erythropoiesis from working properly. Common observations include pallor, failure to thrive, skeletal abnormalities, and varying degrees of hepato- splenomegaly. There is also an overall deficit of hemoglobin tetramers in the Red Blood Cells (RBC) with reduction in the Mean Corpuscular Volume (MCV) and Mean Corpuscular Hemoglobin (MCH).

Aims: The purpose of the current study was to determine the pace of red cell alloantibody formation and specification in patients with transfusion-dependent Thalassemia who fell within the transplantable age range. While bone marrow transplantation is the only curative treatment available, top-up transfusion is the primary treatment for most thalassemia patients. Major selection criteria for patients undergoing bone marrow transplantation (BMT) for thalassemia, as stated by the Thalassemia International Federation (TIF), are- age be 16 years old or younger; have a healthy liver and have low body iron content (minimal excess iron). Serum ferritin levels may rise and hepatomegaly may result from red cell alloimmunization brought on by excessive cell death caused by aggressive transfusion.

Methods: In this study, a total of 860 patient samples underwent three cell screening panels for antibody screening. Of these, 84 positive samples underwent additional evaluation using a fourteen-cell identification panel by a fully automated immunohematology analyzer to determine the presence of a particular antibody.

Serum ferritin and complete blood count were two biological tests that were carried out.

Results: There were 860 patients in all in the research in a center specifically for thalassemia. Data shows 543 (63%) were male and 317 (36.86%) were female. Among total population 590 (68.6%) were E- β -thalassemia, 252 (29.23%) were β -thalassemia major, 18 (2.17%) were others including Hb E disease and β -carrier. Out of 860 patients included in the study, 84 (9.8%) came out positive in antibody screening, all being clinically significant ($p = 0.05^a$). Among them 38 (45.23%) were ≤ 16 years and 46 (54.76%) were ≥ 16 years. The frequent alloantibodies were Anti-E, Anti-D, Anti-c, Anti-C, Anti-Kp^a, Anti-Kp^b. Low level of Hemoglobin and raised ferritin level was also noted in majority of the case.

Summary / Conclusions: Since bone marrow transplantation is the only treatment that can cure people with thalassemia, the number of suitable patients is decreasing as a result of alloimmunization after repeated transfusions. This study focuses on the possibility that age-eligible bone marrow transplant patients may be excluded from consideration due to chronic transfusion-induced sensitization or immunization. Patients with thalassemia should now undergo routine antibody screening and, if necessary, identification, as clinically important antibodies can often be found.

P501 | Clinical case of Tn polyagglutination in a patient with nasal bleeding

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Background: In the field of immunohaematology, polyagglutination is frequently linked to infections and cold autoimmune hemolytic anemia. Polyagglutination can also be associated with anti-Tn antibodies. The Tn antigen is a crypto-antigen, it is not detected on healthy adult cells and all blood donors have anti-Tn antibodies of class IgM present in their blood plasma. Hence, the use of freshly frozen plasma in patients with Tn-defined antigen can result in the occurrence of intravascular hemolysis. In our report, we present a clinical case of Tn-polyagglutination in a patient who was suffering from nasal bleeding and required blood transfusions.

Aims: A description of a case of polyagglutination linked to Tn-antibodies in the practice of immunohematologists.

Methods: The research was based on a blood sample taken from the patient who had frequent heavy nosebleeds and required a fresh frozen plasma transfusion. DiaClon ABO/D ID-cards were used to perform gel agglutination testing for typing of ABO antigens with an analyzer IH-1000 BioRad (USA). Using the polyclonal antibodies anti-A, anti-B, anti-AB, anti-A1 lectin (Russia) and test-erythrocytes ID Diacell ABO 5% (Russia), manual determination of blood type was

made during emergency transfusion. ID Diacell I-II-III 0.8% test-erythrocytes were used in ID-card DC Screening I (BioRad, USA) to screen for red blood cell (RBC) antibodies.

Results: The ambiguous results were received using the manual method of determination of blood type. The patient's RBC reacted with anti-A polyclonal antibodies and anti-A1 lectin with a reactive force of 4+. The patient's plasma reacted with A and B blood type test-erythrocytes. Thus, the patient's ABO blood type could not be determined from the results. Further research was conducted at the immunohaematology laboratory. The use of a hematologic analyzer resulted in the determination of blood type O. Screening of RBC antibodies in the indirect antiglobulin test (IAT) was negative, while the direct antiglobulin test (DAT) was positive due to IgM reaction strength 1+. In order to proceed with the therapy, it was necessary to choose a donor's plasma that is compatible. In our selection of freshly frozen plasma, we utilized a total of 35 samples, with 10 samples for each blood type A, B, O, and 5 samples for blood type AB. The patient's RBC were compatible with only 1 donor's plasma sample. No positive effects were seen when the donor's plasma warmed up to +56°C. The increase in temperature to +60°C resulted in the compatibility of all donor's plasma samples. Plasma samples from three newborns were also provided by the laboratory, and it was found to be compatible with patient's blood. We assumed that the patient had Tn antigen present. To confirm this conclusion, the patient's red blood cells were treated with papain solution and cross-matched with the same donor samples. All samples were compatible.

Summary / Conclusions: Despite its rarity in immunohaematology, Tn-polyagglutination's detection should be given special attention. In order to prevent acute posttransfusion hemolysis, it's important to identify the reasons behind an incompatible cross-match without RBC autoantibodies and alloantibodies in the patient.

P502 | Abstract withdrawn

P503 | A cross sectional analytical study to evaluate the frequency of red cell alloimmunization in pediatric patients

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Background: The prevalence of red cell alloimmunization in adult patient population has been shown to be 1-10% in general population compared to 37% for thalassemia patients and 76 % for sickle cell disease (SCD) patients. Various studies from India have found the alloimmunization in multitransfused transfusion dependent thalassemia patients as 5%-10%. The corresponding studies in pediatric patient population, other than the multitransfused transfusion dependent subgroup are very less. As multiple transfusions in hemoglobinopathies such as thalassemia, SCD are a predisposing factor for RBC

alloimmunization, our study will include pediatric patients who are not lifelong transfusion dependent. This will help to minimize the bias as such patients have a high likelihood of having alloantibodies.

Aims: We aimed to evaluate the frequency and specificity of alloimmunization against major and minor red cell antigens in pediatric age group.

Methods: This was a prospective, cross sectional analytical study for one-and-half-years duration conducted at a tertiary care institute. The study involved collection of data from the blood requisition forms. Blood grouping including auto control, DAT and antibody screen was performed on the blood sample of the patient received. In case the ABS is positive, 11 cell identification panel (ID panel) was put to determine the antibody specificity.

Results: During the study period, data from 12,483 the blood requisition forms was noted. Of these, 7797 were males, 4683 were females and 3 were others. Of these, 5182 were infants (age < 1 year), 5931 were children and adolescents (1-18 years old), and 1370 were adults. Of all requisitions, 4702 were received from pediatric surgery wards, 2522 were received from pediatric cardiothoracic and vascular surgery wards, 2250 from Pediatric hematology wards, 1992 from pediatric emergency, 428 from pediatric intensive care unit (PICU), 298 from neonatal unit of pediatric emergency (NUPE), 215 from pediatric daycare unit, 43 from pediatric gastroenterology ward, and 33 from pediatric endocrinology ward. Of all requisitions, 1693 mentioned a history of previous transfusion, 4103 denied any previous transfusions, and the remaining did not specify any details. On blood grouping, 3002 had A positive, 115 had A negative, 4377 were found to have B positive blood group, 170 B negative, 859 had AB positive, 53 had AB negative, 3763 had O positive, and 144 had O negative blood group. Alloimmunization was found in 33/12483 samples (0.26%). The maximum antibodies were against Rh system antigens (D - 9/33, 27.27%; D,C - 6/33, 18.18%; and E- 1/33, 3.03%), followed by MNS system (M - 9/33, 27.27%; and M, Fy^b - 1/33, 3.03%). Rest were anti-Kp^a - 2/33, 6.06%; anti-Le^a - 1/33, 3.03%; and 4 were inconclusive. Bombay blood group was reported in one patient (1/33, 3.03%; diagnosis of hydrocephalous). DAT positivity was seen in 37 samples (0.29%). On differential DAT, most were IgG (33), and 4 both IgG and C₃d. Of these, only 3 were found to be clinically significant and interfered with compatibility testing. Best match packed red cell units were issued to these patients.

Summary / Conclusions: Red cell alloimmunization was found to be much less compared to adult population. Once a patient is identified to be alloimmunized against red cell antigens, multiple aliquots of phenotype matched units should be prepared and reserved for that specific patient.

P504 | Paroxysmal nocturnal hemoglobinuria - to be or not to be?

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Background: Paroxysmal nocturnal hemoglobinuria (PNH) is by definition characterized by nonimmune intravascular hemolysis with a negative direct antiglobulin test (DAT). However, in the case we report, we obtained a positive DAT, which can be explain by PNH physiopathogenesis with complement-mediated intravascular hemolysis.

Aims: Clinical case report of a PNH with a positive DAT.

Methods: Collection of clinical data in SClínico[®] and ANASIBAS[®] applications.

Results: Man, 63 years-old, with a mechanical aortic valve, overweight and hypertension as past medical history, presented to the emergency department with progressive worsening asthenia and choloria with 10 days of evolution. The laboratory findings documented hemolytic anemia (hemoglobin 9.5 g/dL, with previous values of 14 g/dL), with increased lactate dehydrogenase, indirect bilirubin and reticulocyte count, decreased haptoglobin, hemoglobinuria and urobilinuria. The blood smear did not show spherocytes or schistocytes. Organ function was assessed and was normal. Other causes of hemolysis were excluded, namely mechanical cause, with a transesophageal echocardiogram showing normal mechanical aortic valve function. To distinguish immune from nonimmune causes, a DAT was performed with polyspecific and monospecific reagents (anti-IgG, anti-IgA, anti-IgM, anti-C3c and anti-C3d) by gel microcolumns from BioRad (BioRad, Diamed GmbH, Cressier) and a weak agglutination was found on both tests (1+ and 1+ only for anti-C3d, respectively). As the DAT was negative for the immunoglobulins (IgG, IgA or IgM), the complement had a weak agglutination, there was no history of previous blood transfusions and the patient presented with significant intravascular hemolysis on the laboratory findings, we assumed that was prudent to not exclude nonimmune intravascular hemolytic causes. Classic PNH was then diagnosed by flow cytometry with the following clonal size: 68% neutrophils, 63% monocytes and 8% red blood cells (RBC).

Summary / Conclusions: Diagnosis of PNH may be delayed because of its nonspecific clinical features, variable clinical presentation and rarity; moreover, we know that prompt and accurate diagnosis is particularly important since effective complement inhibitors have become available. In this case a weak positive DAT made the diagnosis even more difficult. The genetic defect and the mechanism of complement-mediated hemolysis of PNH explain the presence of the complement on the RBC surface and its weak positivity on DAT. To the best of our knowledge, there is only other published case in which the patient showed a weak positive DAT also for C3d (Höschmann, Vox Sang, 2012).

P505 | Antibody that coated erythrocytes of beta thalassemia major patients who experience erythrocyte autoimmunization due to repeated transfusions

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Background: Beta thalassemia major patients have to receive repeated erythrocyte transfusions throughout their lives. One of the complications of repeated transfusions is erythrocyte autoimmunization which results in difficulty in obtaining compatible blood for the next transfusion. Autoimmunization can be caused by loss of self-tolerance which consists of central or peripheral tolerance, so that T cells or antibodies react to self-antigens resulting in cell damage. Erythrocyte autoimmunization is characterized by a positive direct antiglobulin test (DAT) with or without a positive indirect antiglobulin (IAT) test. Positive DAT is produced by antibodies that coated erythrocytes, whether IgG, C3d or both. To find out whether IgG or C3d or both that coated erythrocyte, its necessary to do a monospecific antiglobulin test.

Aims: To analyse the antibodies that coated the erythrocytes of beta thalassemia major patients with positive direct antiglobulin test results.

Methods: Observational descriptive study of beta thalassemia major patients who received repeated transfusions treated at RSUP Dr. M. Djamil Padang Hospital. Beta thalassemia major patients with hepatitis B, hepatitis C, malaria and HIV infections were excluded from this study. Polyspecific and monospecific antiglobulin tests are carried out using the column agglutination method. This study was approved by the local ethics committee.

Results: A total of 79 beta thalassemia major patients in this study had an age range of 7 months until 58 years, 33 patients were male (41.8%) and 46 female patients (58.2%). Of the 79 patients, 35 patients (44.3%) had a positive direct antiglobulin test based on the polyspecific antiglobulin test. After continuing with the monospecific antiglobulin test, 34 patients had IgG, while one patient had IgG plus C3d.

Summary / Conclusions: Erythrocyte autoimmunization was found in 44.3% of beta thalassemia major patients with repeated transfusions, where the antibodies that coated the erythrocytes were IgG.

P506 | Naturally occurring anti-Kell—a clinical case

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Background: Disregarding irregular antibodies related to the Rh system, anti-Kell antibody (anti-K) is the most encountered in pre-

transfusion tests in blood banks. Despite the high immunogenicity of the Kell antigen (K), it has a low prevalence. Most anti-K antibodies are alloantibodies triggered by pregnancy or blood transfusions and they can be detected in circulation for years. There are few cases described where anti-K antibodies are detected in patients not exposed to K-positive red blood cells (RBC). These rare cases are usually associated with bacterial infections, disappearing after recovery.

Aims: Describe two situations, the effect that the *rouleaux* phenomenon can have in the standard methodology results and the appearance of a naturally occurring Anti-K.

Methods: Review of the patient's medical and analytic history and analysis of the new laboratory findings.

Results: A 71-year-old female patient diagnosed with Waldenström's Macroglobulinemia since 2005, underwent 6 cycles of chemotherapy and second-line treatment in 2006, that was discontinued due to lack of reduction of the monoclonal component. She has been under monitoring since then. Obstetric history includes two pregnancies before 1980 and no miscarriages. Regarding transfusion history, she received a RBC unit in 2002 due to uterine leiomyoma hemorrhage and a RBC unit in 2005 for gastrointestinal bleeding. In the latter case, pretransfusion automated compatibility tests, including irregular antibody (IA) screening by gel microtube hemagglutination (IH-1000, Bio-Rad®), showed non-specific panreactivity due to the *rouleaux* phenomenon, confirmed microscopically. To clarify this, solid-phase IA screening on a microplate (Neo Immucor®) was performed with completely negative results. The blood group was A Rh+, and the Rh and K phenotypes determined were C- c+ Cw- E+ e+ K-. The RBC unit was manually crossmatched, matching the phenotype, and the transfusion proceeded without complications. In January 2024, she was admitted for duodenal lesion resection after the diagnosis of an incidental gastrointestinal stromal tumor found in a surveillance CT scan. During pretransfusion tests for RBCs units for the surgical protocol, a positive IA screening was detected in gel microtube hemagglutination (IH-1000, Bio-Rad®), identifying a *de novo* IgM anti-K antibody with intensity ++- in LISS/Coombs and negative in papainized cells on 11-cell panels (ID-DiaPanel/ID-Diapanel-P, Bio-Rad®). The surgery proceeded without complications, and the use of previously crossmatched K-negative RBCs was not necessary. The identification was repeated in a new sample five days after the initial one (four days after surgery), with identical results. The patient denied other pregnancies, transfusions after 2005, and recent infections. The preoperative analytical study was within reference values.

Summary / Conclusions: This clinical case allows us to address two important situations in transfusion safety. Firstly, characteristics such as the *rouleaux* phenomenon can alter standard methodology results, in this case, due to RBC aggregation, demanding additional techniques for clarification of atypical results. Secondly, no cause was found to explain the appearance of an anti-K alloantibody. Although rare and not recent, cases of natural expression are described in the literature. Therefore, due to the potential immunogenicity of the Kell antigen and the clinical relevance of the corresponding antibody, its exclusion in the selection of compatible units is important.

P507 | Rare Anti-f(ce) identified in a patient with abscess of bursa in hip joint.H Lee¹, D Jekarl²¹Laboratory medicine, Yeouido St.Mary's hospital, ²Laboratory medicine, Seoul St.Mary's hospital, Seoul, Korea, Republic Of**Background:** Here, we report a rare anti-f(ce) antibody case in South Korea, defined by modified adsorption test.**Aims:** The aim of this presentation is to share our experience which define rare unexpected antibody.**Methods:** 66 years-old male who had never received transfusion, was diagnosed with abscess of bursa in hip joint, in previous institution. During therapy, he received total 4 units of packed red blood cell (pRBC) about 60- and 50-days ago, twice. Soon, he was transferred to our institution for operation, and his initial hemoglobin concentration 16.4(g/dl). However, after operation, his hemoglobin concentration was decreased to 10.6(g/dl), so that pRBC transfusion was decided. His antibody screening test using 0.8% selectogen[®] (Ortho Clinical Diagnostics[™]) show all negative results to 2 reagent RBCs, but his cross-matching test show incompatibility to 2 donor RBCs (of 7 donor RBCs). At first, with suspicion for Anti-Di^a which was not included in that panel, further antibody screening test using Surgiscreen[®] (Ortho Clinical Diagnostics[™]) was performed. This test identified Anti-f(ce) in 3+ rather than Anti-Di^a, sequential antibody identification test using 0.8% Resolve[®] panel C system (Ortho Clinical Diagnostics[™]) also identified Anti-f(ce) in 3+ without any coexistent antibody. To confirm this antibody, 'Rh CcEe subgroup profiling of incompatible donor RBCs' and 'Re-antibody screening test after adsorption to incompatible donor RBCs' were planned.**Results:** First, in 'Rh subgroup profiling of incompatible donor RBCs', all cross-matched packed RBCs were 18 units. Of those, 16 units were compatible, but 2 units were not. These incompatible donor RBCs show Cce and cEe, respectively. This results suggest that these RBCs contain c- and e- antigens, which are coded in same chromosome. Second, in 'Re-antibody screening test after adsorption to incompatible donor RBCs', the remnant plasma after adsorption to one (Cce) of above 2 units was tested. In initial try, consisting of three-times adsorption which took 30 minutes each in 37°C, the remnant plasma persistently show Anti-f(ce) in '3+'. However, in modified try, adding one more adsorption which took 7 hours in 37°C as soon as above initial try, the remnant plasma show Anti-f(ce) in decreased strength '1+'.**Summary / Conclusions:** 'f(ce)' antigen is known for compound Rh antigen, which occurs once c- and e- antigens are coded in same haplotype (cis-arrangement). In our study, all compatible donor RBCs have neither c- or e- antigen, nor the possibility in cis because these antigens could be coded in trans, even in CcEe. On the other hand, all incompatible donor RBCs show at least one cis-coded c- and e- antigens, which suggest the coexistence of compound antigen f. Moreover, the trial for identifying Anti-f(ce) with adsorption seems to need more incubation time than expected. Typical method using

several times taking only 30 minutes each was not effective to adsorb this antibody. Further trials performed in different conditions, with definitely proven f(ce) antigen is needed to verify more.

P508 | Abstract withdrawn**P509 | Distribution of ABO and Rh blood group phenotypes among Palestinian blood donors—a nationwide study (1981-2017)**R Abu Seir¹, M Al-Ghoul², K Rimawi³, C Hesse⁴, O Najjar⁵¹Medical Laboratory Sciences Department, Al-Quds University, ²Al-Makassed Islamic Charitable Hospital, Jerusalem, ³Central Blood Bank, Palestinian Ministry of Health, Ramallah, Palestinian, State of, ⁴Department of Laboratory Medicine, The Sahlgrenska Academy, University of Gothenburg, Gothenburg, Sweden, ⁵Allied Health Professions, Palestinian Ministry of Health, Ramallah, Palestinian, State of**Background:** The ABO system is the most important of all blood group systems in transfusion practice. The ABO and rhesus (Rh) blood group antigens are the most frequently studied genetic markers in a large group of people. Blood type frequencies vary in different racial/ethnic groups. There are limited data on the frequencies of ABO and RHD antigens in Palestine.**Aims:** This study aims to determine the distribution of ABO and Rh (D) blood groups and the demographic background of blood donors on a nationwide level over the period between 1981 and 2017.**Methods:** We conducted a retrospective cross-sectional study including blood donors. The blood group and demographic data of 105,629 blood donors were included in this study. Data were collected from the blood bank donor data registration systems at Al-Makassed Islamic Charitable Hospital (n = 28,367) covering the period between 1981 and 2012 and from the Palestinian Ministry of Health (n = 77,262) covering the period between 2010 and 2017. The frequencies and percentages of ABO and Rh blood groups were calculated.**Results:** Of 105,629 blood donors, 96% were males and 48.3% were 20-29 years old. The most common blood group was blood group A (39.8%) followed by O (37.3%), B (16.2%), and AB (6.7%). Considering ABO and Rh blood groups altogether, blood group A positive with 35.6% was the predominant blood group followed by O positive (32.2%), B positive (14.4%), AB positive (6.0%), O negative (5.2%), A negative (4.2%), B negative (1.8%), and AB negative (0.7%). The majority of study participants were 88.2% Rh (D) positive.**Summary / Conclusions:** This is the first study about blood group distribution in Palestine based on a large number of blood donors. This study showed that blood groups A and O were the predominant followed by B and AB. In addition, most of the blood donors' blood groups were Rh-positive (88.2%). The findings of our study can guide the blood center administrators to make decisions concerning blood stocking and supply.

P510 | Comparative study between two automated devices on the sensitivity to detect weak Duffy b phenotypes in blood donors

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Background: The Duffy blood group is involved, although to a lesser degree than other antigens, in immunization with potential gestational or post-transfusional hemolytic reactions. The FYX allele results in a weak expression of the Fyb antigen. Its prevalence in our population is around 2%. With a prevalence of 18% for FYA/FYA genotype, if not enough sensitive phenotyping techniques are used, around 7% of our donors typed as Fy(a+,b-) could actually be Fy(a+,b weak+). It is necessary to adopt measures to minimize this potential typing error which could be a risk for the recipient.

Aims: Compare the sensitivity to serologically detect weak Fyb expression in heterozygous donors (FYA/FYX) between the IH-500 and IH-1000 systems

Methods: We checked the phenotype and genotype historical status for the Duffy blood group on 2,596 blood donors. Samples from those donors identified as FYA/FYX would be obtained after our lab routine was finished. Duffy b phenotype for this samples were done in parallel with the same lot of reagents in the IH-500 and IH-1000 systems (Bio-Rad). The expression of Fyb antigen was confirmed with a manual technique using PEG. Sensitivity of either automated systems would be calculated and compared.

Results:

Among the 1121 Fy(a+, b+) pheno and genotype confirmed donors, twenty-two were identified as FYA/FYX (1.98%) and confirmed to express the Fyb antigen with 100% positive results (ranging from 1+ to 3+) in the manual PEG technique. The IH-500 system returned an alarm for user review in 10 out of the 22 samples, resulting in a weak or positive result (ranging +/- to 1+). The 12 remaining samples were visually reviewed as well, confirming a negative result. This brings a fraction of true positives equal to 45.45%. The IH-1000 system identified all 22 samples as negative, visual confirmation was checked as well.

Summary / Conclusions: Although the sensitivity of routine automated serological tests for weak Duffy b are low or null, the IH-500 is capable to detect nearly half of the cases, avoiding at least part of the additional serological or genetic studies needed to correctly true type Fy(a+, b-) blood donors and their blood components.

P510 - Table 1: Results.

Typing in blood components label	Absolute n	%
Fy(a+, b-) (Genotype confirmed)	318	12.25%
Fy(a+, b+) (Genotype confirmed)	1121	43.18%
Fy(a-, b+)	1157	44.57%

P511 | Incidental discovery of anti-Wra in two cases with coexisting autoantibodies

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Background: This case study delves into the complexities surrounding two patients with non-specific autoantibodies and the unexpected discovery of Anti-Wra.

Aims: The primary aim of this study is to elucidate the implications of non-specific autoantibodies and incidentally identified Anti-Wra. Understanding the clinical significance and optimal management of such cases is crucial for ensuring safe and effective blood transfusions.

Methods: Laboratory techniques for antibody identification, such as direct antiglobulin testing, gel column agglutination and enzyme-enhanced technique were employed. Adsorption technique was used to remove interference of non-specific autoantibodies, helps in identifying underlying alloantibodies more accurately. Multiple Identification panels which include a diverse range of antigens were also used.

Results: During pre-transfusion testing, antibody screening results were positive for both patients. Both patients presented with non-specific autoantibodies, and further testing using adsorbed plasma revealed the presence of Anti-Wra.

Summary / Conclusions: Low-frequency antigens are not typically included in routine antibody screening panels due to their infrequent occurrence in the general population. Wra is generally regarded as a low frequency antigen in populations studied, but further studies on Wra+ frequency in the local population may be warranted. The incidental discovery of Anti-Wra, a rare but could be clinically significant antibody, raised questions about their potential impact on routine blood banking practices.

P512 | Abstract withdrawn**P513 | ABO isoagglutinin titers in group O blood donors**

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Background: Passively transfused blood group antibodies cause adverse events in recipients. ABO remains the most important of 43 blood group systems registered so far. High titers of anti-A and anti-B seem to be one reason for hemolytic transfusion reactions and for ABO hemolytic disease in fetus and neonates. Due to chronic scarcity of blood in this part of the world, 'O' blood group transfusions are frequently used both in adults and pediatric patients. There is no consensus for critical ABO antibody titers to guide transfusion decisions. To mitigate ABO incompatibilities, implementation of ABO titer measurement can favor reduction in transfusion reactions in non-group 'O' recipients.

Aims: A study was conducted to understand the trend and quantification of anti-A and Anti-B antibody titer levels in group 'O' donors in this geographical population

Methods: A prospective study was conducted in 218, Group 'O' blood donors in a tertiary care super specialty hospital, fulfilling the laid down donor selection criteria, from January to December 2021, during COVID-19 pandemic. IgM antibody titers were measured by hemagglutination, and IgG antibody titers were measured by solid-phase red cell adherence technique on automated immunohematology system. Antibody titers equal to or greater than 128 were considered "high titers." Statistical analysis of the data was done using Epi Info 7.1 software, with *p* values <0.05 considered significant.

Results: Most whole blood donors during this period were male (96.75%). Of 218 donors, the majority (94.95%) had Rh positive. Median age of donors was 33.16 ± 8.8 years, with highest number of donors aged ≤ 35 years with male preponderance (97.7%). ABO antibody titers ranged from 0 to 1024. Prevalence of titers of less than 128 for anti-A and anti-B IgG was 79.36% (*N* = 173) and 86.70% (*N* = 189), respectively. Titers of less than 128 for anti-A and anti-B IgM were 97.71% (*N* = 213) and 98.17% (*N* = 214), respectively. High IgG anti-A antibody titers were exhibited in 20.64% and anti-B titers in 13.30% of donors, with the majority being male donors under 50 years of age. Just 8.71% (*N* = 19) of donors had both anti-A and anti-B high IgG titers. 63% of components were transfused to patients during study. None of the recipients showed any hemolytic reactions.

Summary / Conclusions: In this study, our donor population has a predominance of low isoagglutinin titers. Antibody titers equal to or greater than 128 were considered "high-titers" and critical. In view of their implicated relationship with hemolytic transfusion reactions, accurate measurement of isoagglutinin titers in donors of the "O" blood type is both essential and recommended. Defining critical titers will help formulate institutional guidelines on ABO-incompatible platelet transfusions. The use of automation in ABO titer assays removes subjective, inter-observer, or inter-laboratory variations, and use of DTT in technical procedures offering consistent, precise, and less labor-intensive assays in resource-constrained settings.

P514 | Unraveling the complexity of fetomaternal transfusion—a case report

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Background: Fetomaternal transfusion, the transfer of fetal red blood cells into the maternal circulation, remains a critical aspect of maternal-fetal immunology. Understanding the causes and potential risks associated with this phenomenon is crucial in managing pregnancy complications.

Aims: This case elucidates a specific case where massive fetomaternal transfusion and inadequate dosage of RhD Immunoglobulin led to the formation of anti-RH1, anti-RH2 and anti-RH12 antibodies.

Methods: The investigation included a comprehensive set of immunohematological techniques as serological typing of ABO and RH (Grifols, CH), direct (DAT) and indirect antiglobulin test (IAT) (ID-system, BioRad/Grifols, CH and SCARF), elution and antibody titration (in-house). We also conducted a G-absorption/elution technique to differentiate between anti-RH1, anti-RH2 and anti-RH12 (in-house).

Results: Due to decreasing child movements, the patient was hospitalized in the 38th week of pregnancy, initially showing a negative antibody screening. Three days later she gave a spontaneous stillbirth with massive fetomaternal transfusion, reflected by mixed field reactions in maternal RH determination. Double dosage of RhD Immunoglobulin was administered. Anti-D detected in both maternal serum and eluate, supported the suspected perinatal fetomaternal hemorrhage. However, three months later, three alloantibodies were differentiated: Anti-RH1, Anti-RH2, and Anti-RH12.

Summary / Conclusions: The subsequent formation of anti-RH1, anti-RH2 and anti-RH12 antibodies after massive fetomaternal transfusion raised questions about the adequate dosage of RhD immunoglobulin. The ensuing discussion revolves around determining the optimal preventive measures to avert anti-D alloimmunization. Unfortunately, the extent of the fetomaternal hemorrhage was not determined with a suitable method (eg. Kleinhauer-Betke or flow cytometry) as it is recommended to administer additional doses of RhD Ig accordingly to the amount of fetal red blood cells. With sufficient administration of RhD Immunoglobulin the alloimmunization against the anti-RH1 could have been prevented. Further, clear distinction between anti-RH1, anti-RH2 and/or anti-RH12 is indispensable to provide accurate recommendations for future pregnancies. The subsequent pregnancy, marked by vigilant monitoring and control measures, ultimately resulted in the birth of a healthy baby boy showing the phenotype RH:-1,-2,-3,4,5,-12.

P515 | Abstract withdrawn

P516 | Frequencies of blood group antigens and phenotypes in the Northern Vietnamese population

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Background: Blood safety is the top priority in transfusion medicine. However, patients in Vietnam are only transfused with ABO and RhD compatible blood products which could pose a threat to induce alloimmunization. For the possibility of providing increased selected antigen matching, knowing the population's various blood group antigen

(BGA) frequency is of great value. Very limited studies have been conducted in the past with inadequate sample sizes to demonstrate the antigen frequency of many of the blood group system antigens.

Aims: Our study was performed to provide improved information about the frequencies of antigens and phenotypes of major blood groups in Vietnamese donors.

Methods: Blood samples from 1218 blood donors were collected to test for a variety of blood group antigens in the following blood group systems: RH, KEL, FY, JK, LE, MNS, P1Pk and LU. Column agglutination tests using prefilled cards with antisera or monoclonal antisera added to the card column to test for the red cell antigens. Antigen and phenotype frequencies were calculated and expressed as percentages.

Results: Amongst the Rh antigens, e was the most common (96.96%) followed by D (96.72%), C (92.86%), c (43.10%), and E (32.59%) with D+C+E-c-e+ (54.02%) being the most common phenotype while D+C+E+c-e+ the second most frequent. The percentage of donors that tested positive for the remaining blood group antigens evaluated is: K(0.0), k(100.0), Fy^a(99.34), Fy^b(14.61), Jk^a(71.59), Jk^b(75.62), M(81.20), N(63.05), S(5.58), s(100.0), P1(22.25), Le^a(26.85), Le^b(84.65), Lu^a(0.0), Lu^b(100.0) and Mi^a(9.20). In the FY system, the phenotype Fy(a+b-) was 85.39%. Only eight of the 1218 donors phenotyped as Fy(a-b+). The most common phenotype in the JK blood group system was Jk(a+b+) at 47.62%. In the Lewis blood group system, the major phenotype found was Le (a-b+) (60.43%), while Le(a+b+) was found in 24.22%. In the MNS system, M+N+(46.63%), S-s+ (94.42%), were most frequent.

Summary / Conclusions: This study shows the frequencies of twenty-one blood group antigens in Northern Vietnam donors. Knowledge of red cell antigen phenotype frequencies can help prepare indigenous cell panels, provide antigen-negative blood to patients with multiple alloantibodies, and potentially prevent alloimmunization using better antigen matching methods.

P517 | Abstract withdrawn

P518 | Abstract withdrawn

P519 | A secondary IgM-mediated warm autoimmune hemolytic anemia with a catastrophic presentation

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Background: Autoimmune hemolytic anemia (AIHA) is caused by autoantibodies against red blood cell antigens. Warm AIHA, due to antibodies that are active at body temperature, is the most common type of AIHA and are almost always IgG, although IgA and warm-acting IgM have been reported. An evaluation for an underlying disorder is indicate in most individuals, especially older adults and those

who have evidence of an infectious, autoimmune or lymphoproliferative disorder.

Aims: Clinical case report of an IgM-mediated warm autoimmune hemolytic anemia secondary to a lymphoproliferative disorder.

Methods: Collection of clinical data in SCLínico[®] and ANASIBAS[®] applications.

Results: Woman, 55 years-old, with Hashimoto disease, presented to the emergency department with progressive worsening asthenia and palpitations with 7 days of evolution. The laboratory findings documented hemolytic anemia (hemoglobin 5.3 g/dL), with increased lactate dehydrogenase, indirect bilirubin and reticulocyte count and decreased haptoglobin. The blood smear did not show spherocytes or schistocytes. Organ function was assessed and was normal. To distinguish immune from nonimmune causes, a direct antiglobulin test was performed with polyspecific and monospecific reagents (anti-IgG, anti-IgA, anti-IgM, anti-C3c and anti-C3d) by gel microcolumns from BioRad (BioRad, Diamed GmbH, Cressier) and an agglutination was found on both tests (3+ for polyspecific and 2+ IgM and 3+ anti-C3d for monospecific reagents). Cold-induced symptoms were excluded, and a warm-acting IgM AIHA was assumed. Corticotherapy was started (prednisolone 1mg/kg), with a satisfactory clinical and laboratory response. Four weeks later a hemoglobin level of 13.5 g/dL was achieved. However, anemia relapsed whenever corticotherapy was tapered. Secondary causes were being evaluated, when three months later she was brought to the emergency department due to a syncopal episode. A recurrence of the disease was diagnosed, with a hemoglobin of 3.4 g/dL and a multi-organ dysfunction. The patient was admitted to intensive care unit. A bone marrow biopsy was performed and revealed a diffuse large B cell lymphoma. Chemotherapy was started, but the patient had an unfavorable evolution and deceased 20 days after admission.

Summary / Conclusions: This report shows a rare case of IgM-mediated warm AIHA. In concordance with the literature, that describes that most cases are associated with an underlying condition, this patient AIHA was secondary to a diffuse large B cell lymphoma and had a catastrophic outcome. With this case we intend to highlight the relevance of exhaustive investigation for secondary causes.

P520 | Abstract withdrawn

P521 | Evaluation of a compact, centrifuge-free immunoagglutination device for pre-transfusion testing

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Background: The traditional tube method for pre-transfusion testing, including ABO & RhD typing, irregular antibody screening, and cross-matching, heavily depends on manual operations. The interpretation of results, particularly post-centrifugation, is considered the most challenging and non-standardized aspect. In Taiwan, Delta Electronics has developed a compact device named "Card Mixer," an automatic liquid mixing device designed for programmable blending of blood cells and serum without requiring a centrifuge. This innovation expedites the agglutination reaction. The device, equipped with a single-use dedicated cartridge containing six microchannels, allows operators to use a pipette for mixing patients' plasma or anti-serum with 2-5% red blood cells individually. Results can be obtained within 10 min.

Aims: To Assess the performance of this immunoagglutination device in accurately conducting pre-transfusion experiments.

Methods: For ABO & RhD Typing: According to manual tube method as standard, we would like to evaluate the accuracy of blood grouping and reaction intensity through immunoagglutination device. For irregular Antibody Screening: According to the widely-used Manual Polybrene method in Taiwan, we aim to assess the accuracy of screening for irregular antibodies through an immunoagglutination device utilizing Manual Polybrene.

Results: For ABO & RhD Typing: We collected and examined 400 specimens. This immunoagglutination device exhibited 100% concordance with the standard manual tube method in forward typing for ABO grouping and Rh typing. In serum typing, a total of 19 specimens (4.75%) needs the second-stage enhanced reaction because their agglutination intensity scores were less than 2+. Following the second-stage reaction, three specimens (0.75%) remained no agglutination. For irregular Antibody Screening: We prepared 25 irregular antibodies positive samples, including anti-Mi^a, anti-D, anti-E, anti-P1, anti-Le^a, anti-N, anti-c, anti-M, and anti-Jk^b, representing commonly observed irregular antibodies in the Taiwanese population and 25 antibodies negative samples for immunoagglutination device testing. There were six false negative tests noted, resulting in a specificity of 100%, but a sensitivity of only 76%.

Summary / Conclusions: For non-specialized blood bank laboratory technicians, this for immunoagglutination device provide a relatively straightforward and feasible method for pre-transfusion experiment. In a small clinic with transfusion service, this device enables less experienced healthcare professionals to perform comprehensive pre-transfusion tests and contribute reducing human error and enhancing transfusion safety.

P522 | First report of phenotype prevalence of five major Rh blood group antigens in the blood donor population of Peshawar, Pakistan

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Background: After ABO, Rh is the most clinically significant and complex blood group system. It's warm-reacting antibodies can result in mild to severe haemolytic transfusion reactions as well as haemolytic diseases in the foetus and newborn. The frequency and distribution of Rh antigens vary greatly across the globe. Less research has been done in Pakistan on the frequencies and immunogenicity of C, c, E, and e. There have only been five studies in Pakistan to date that have assessed the frequency of Rh major antigens; however, no data from the Khyber Pakhtunkhwa province has ever been reported. It is pertinent to mention that the combined sample size of these five studies was 1,694, much less than our sample size of 13,148.

Aims: To investigate the prevalence of five major Rh blood group antigens.

Methods: A total of 13,148 blood donor samples were analyzed for Rh D, C, c, E, and e antigens using monoclonal reagents (Bio-Rad Laboratories) and tested against known positive (heterozygous antigens from antibody screening cells) and negative antigens according to manufacturer instructions. The frequencies were calculated by direct counting on a spreadsheet programme, and the Statistical Package for Social Sciences was used to conduct a meta-analysis for comparison of results with national and international studies. All results were standardised to percentages.

Results: The findings of the current study revealed that 92.17% of donors were RhD-positive. The e antigen showed the highest frequency of 98.31%, followed by the D antigen and the C antigen (92.17% and 78.37%, respectively). Comparing these results to earlier Pakistani studies and international data showed big differences in the frequencies of the antigens. These differences may be due to the region's diverse ethnic population and geography.

Summary / Conclusions: The findings emphasize the importance of local antigen frequency data for efficient blood transfusion strategies, especially for thalassaemia patients requiring frequent transfusions.

P522 - Table 1: Rh antigens frequency compared with earlier studies from Pakistan

Institute and City	C	c	E	e
Karim et al., 2015, Aga Khan University Hospital, Karachi	87%	57%	19%	99%
Anwar et al., 2016, National Institute of Blood Disease and Bone Marrow Transplantation, Karachi	89.6%	62.8%	22.6%	97%
Mahmood et al., 2018, Armed Forces Institute of Transfusion, Rawalpindi	87.53%	61.18%	21.41%	98.59%
Tariq et al., 2022, National Institute of Blood Disease and Bone Marrow Transplantation, Karachi	80.6%	66.5%	25.1%	97.8%
Khan et al., 2022, Chughtai Institute of Pathology, Lahore	70.20%	47.70%	26.90%	98.54%
Saba et al., 2023, Regional Blood Centre, Peshawar (current study)	78.36%	45.69%	25.14%	98.30%

P522 - Table 2: Rh antigens frequency compared with other countries

Author and Year	C	c	E	e
Yu et al., 2016, China	88.81%	58.43%	50.78%	92.28%
Shahverdi et al., 2016, Iran	77.04%	73.58%	30.47%	96.66%
Shah et al., 2018, India	91.0%	50.5%	16.5%	100%
Al-Riyami et al., 2019, Oman	74.3%	77.3%	28.2%	98.3%
Owaidah et al., 2020, Saudi Arabia	59%	86%	21%	97%
Orhan et al., 2022, Türkiye	68%	76%	14%	86%
Saba et al., 2023, Pakistan (current study)	78.36%	45.69%	25.14%	98.30%

P523 | Evaluation of inconclusive results in blood group serology

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Background: Despite the spectacular developments in molecular techniques, serological techniques are still esteemed in blood grouping for being cost-effective and reliable in determining the blood group phenotype. However, the discrepancies between forward and reverse grouping and weak phenotypes of D antigen can be challenging in particular cases. **Aims:** In this multicentral study, inconclusive results in ABO grouping and Rh typing were evaluated and the potential impacts of different techniques were also investigated.

Methods: Annual (2023) blood grouping data from three immunohematology laboratories served in different university hospitals were reviewed. For the determination of ABO grouping and RhD typing commercially supplied column agglutination gel cards and reagents (Across

Gel, DiaPro, Turkey) were used in all facilities. Additionally, the microplate technique based on red blood cell magnetization (Diagast, France) was also performed in a limited number of samples. All analyses were conducted by using automated platforms for both column agglutination and microplate techniques those were OCTO-M and QWALYS 3 analyzers, respectively. The chi-square was carried out for statistical analysis.

Results: In total, 153,409 blood samples were analyzed. Female/Male ratio was 1.6 (93725/59778). The mean and median for age were 47 and 47 (min 1, max 87), respectively. ABO discrepancy was detected at 0.054% (83/153503) and weak D phenotype was detected at 0.02% (31/153503). Cases with ABO discrepancy were most likely A antigen variants by detection of 0.04% (62/153503). In 21 cases (0.014%), the weakness of expected isohemagglutinins was observed. In five cases it was unable to identify the ABO group serologically because of chimerism probably related to recent transfusions. The microplate technique was used as an alternative method in randomized limited samples ($n = 3025$) to check the performance of routine systems. In all facilities, coherent results were obtained between the column agglutination and microplate techniques.

Summary / Conclusions: The frequencies for ABO variants and weak D were found similar to the findings of various studies. Even though the same automated platforms and gel cards are used and the same algorithms are followed in every facility, the frequency of inconclusive results was significantly lower in one of the transfusion centers which had processed far more tests over a hundred thousand samples ($p < 0.001$). Besides serological studies, molecular workup must be also considered to clarify the inconclusive results in blood grouping.

P524 | Serological evaluation of incompatible crossmatches encountered in pre-transfusion testing

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Background: The evolving landscape of transfusion medicine has seen a substantial increase in the adoption of immunohematology (IH) analyzers. While the type and screen policy has become increasingly prevalent due to its efficacy, the traditional practice of routine

crossmatching maintains its critical role. This study was aimed to study the incidence of all the incompatible crossmatches encountered in our laboratory and to study the serological reasons for all incompatible crossmatches and screening processes in transfusion medicine, ensuring patient safety and enhancing the effectiveness of transfusion outcomes.

Aims: To study the incidence of all the incompatible crossmatches encountered in our laboratory. To study the serological reasons for all incompatible crossmatches.

Methods: This study was meticulously designed as a cross-sectional study. The study was conducted in the Department of Pathology in collaboration with the Department of Immunohematology and Blood Transfusion, Swami Rama Himalayan University, Swami Ram Nagar, Dehradun over a period of 10 months after obtaining an informed written consent from the patients and ethical clearance from the institutional ethics committee. Study samples were taken from blood samples presented in blood centre for requirement of blood for cross matching. Spanning a period from March 1, 2023, to December 31, 2023. Total 46 participants were included in the study.

Results: Upon the evaluation of patient samples, a relatively small fraction, exhibited incompatibility, translating to a 0.8% incidence rate. Notably, the incompatibility was predominantly observed in females, accounting for 60% of the cases. A significant portion, approximately 85%, of these patients had a history of repeated transfusions. Among the incompatible samples, only 38% yielded a positive result on the indirect anti-human globulin test or during antibody screening. Notably, 17% demonstrated panagglutination and were subsequently managed with the administration of Rh, Kell phenotype, or best-matched red cell units

Summary / Conclusions: The findings from this extensive study elucidate the significant role of antibodies against the Rh system, highlighting it as the most common cause of incompatibility in red cell transfusions. The results underscore the need for meticulous and evolved crossmatching and screening processes in transfusion medicine, ensuring patient safety and enhancing the effectiveness of transfusion outcomes.

P525 | Immunohematology in resource-constraint setting—a single center experience from Karachi, Pakistan

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Background: Antibody screening is a test that detects antibodies in the blood that may react with different blood groups. It is important for safe blood transfusions and diagnosing some diseases. The prevalence and specificity of antibodies vary by region, ethnicity, genetics, and exposure to transfusions, pregnancies, and infections. Local data on antibody screening and patient characteristics are required to guide blood transfusion policies and practices. However, there is a lack of published data on antibody screening outcomes and the demographics of the general population Pakistan. Karachi is a large and diverse city that faces challenges in providing adequate and safe blood transfusion services. In this paper, we report the results of antibody screening in the general patient population.

Aims: The objective of this study was to determine the frequency of red cell alloantibodies by column agglutination gel card method.

Methods: This was a cross sectional descriptive study covering over a period of one year from January 2023 till December 2023. This study was conducted at Chughtai Laboratory Karachi, Pakistan. A total of 402 samples were received in our centre for antibody screening, included samples of all ages and genders. We performed ABO and Rh D blood typing on the samples. Then, we used the column agglutination gel card method with three screening cell panels to detect any red cell alloantibodies in the samples. We confirmed the positive results using 11 cell panels as per the manufacturer's guidelines.

Results: 93.5% of females and 6.5% of males were processed for antibody screening. Out of total 402 samples, 17(4.2%) samples were identified with alloantibodies. Out of total positive samples following are the identified alloantibody described below: Anti -D = 6, Anti D + Anti C = 3, Anti E = 2, Anti Fya = 1, Anti M = 1, Anti K = 1, Weak Positive = 1, Autoantibody = 2.

Summary / Conclusions: This study has implications for the management of transfusion and pregnancy in the screened population. The high prevalence of anti-D and anti-C suggests that there is a need for more comprehensive and accessible Rh immunization programs for pregnant women, as well as more extensive Rh typing and matching for blood donors and recipients.

P526 | Effect of storage time for monoclonal anti-D product from Central Blood Transfusion Service Indonesian Red Cross

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Background: The use of monoclonal reagents antisera A, antisera B and Antisera D is the most important examination in the confirmation of blood type. To obtain quality reagents and appropriate inspection results, it is necessary to check the stability test in the reagent storage. Antisera can be used or applied within a certain period of time, I conducted a stability test study of the use of monoclonal antisera D on the products of the Central Blood Transfusion Service Indonesian red cross.

Aims: This study aims to determine the stability of monoclonal antibody research on the degree of agglutination.

Methods: The test method used in this study is the tube method and slide test method with examination criteria based on WHO standards namely potency test, avidity test and specificity test.

Results: Storage stability test results of Antisera D monoclonal products from the central blood transfusion service Indonesian red cross, for the potency test with LOT code D 030523, the highest titer results in August, November 2023, and February 2024 still showed stability at a titer of 1:1024 in the O cell test. In all the test results antisera d monoclonal potential lot code D 030523 is still relatively good and stable in accordance with the feasibility that has been established according to WHO with a minimum titer of 1:128. The results

of Antisera D monoclonal specificity test on LOT Code D 030523 showed that the degree of agglutination that occurred in the re-examination in August, November 2023 and February 2024 was 4+ both in the rh group R1R1, R2R2, R1r, and R2r cell tests. while the results of the Rh group rr cell test were negative. The results of the Antisera D monoclonal avidity test with LOT Code D 030523 showed that the time required for its implementation in August and November 2023 in the O cell test was 4 seconds with the agglutination degree set at 3+ after 2 minutes. while the time required to perform agglutination in February 2024 is 5 s with the degree of agglutination performed is 3+ after 2 min of reaction. Although there is an increase in avidity time, but the results obtained are still within the standard time range set by WHO which is less than 10 seconds and the degree of agglutination formed is still in accordance with the reagent quality graduation standard of 3+ after 2 minutes of reaction.

Summary / Conclusions: Based on the results of research that has been done, it can be concluded that the stability test results of monoclonal reagent antisera d affect the storage of antisera on agglutination binding power. The importance of stability test in reagent storage is to find out whether the reagent is still suitable for use or not before the expiration date.

P527 | Abstract withdrawn

P528 | Abstract withdrawn

P529 | Study of unexpected antibodies characteristics in blood donors at Cho Ray Blood Transfusion Center, Vietnam

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Background: Cho Ray Blood Transfusion Center has implemented unexpected antibodies (UAs) screening in blood donor samples from 2015. Unexpected antibodies are considered one of risks leading to the blood transfusion reaction. The UAs screening help detect positive cases and prevent timely transfusion for patients to ensure the blood transfusion safety.

Aims: Investigating the prevalence and characteristics of unexpected antibodies (UAs) in blood donors at Cho Ray Blood Transfusion Center.

Methods: Cross-sectional descriptive study with convenient sampling. The study comprised in three steps: 23,933 blood donor samples were screened for IAs from August 2023 to November 2023. Using the microplate technique on the NEO system with Capture-R Ready-Screen kits (Pool panel cell) from IMMUCOR (USA). 27 reactive samples with Pool panel cell were continuously tested by Capture-R Ready-Screen (3 panel cells) kits from IMMUCOR (USA) on the NEO system. 21 reactive samples with 3 panel cells donors were finally identified with Capture-R Ready-Indicator Red cells kits from IMMUCOR (USA) on the NEO system.

Results: Among the 23,933 blood donor samples at Cho Ray Blood Transfusion Center, the prevalence of positive UAs screening was 0.11%, undetermination was 0.04%, and negative was 99.85%. There was a significant gender difference in UAs occurrence among donors with $p < 0.001$. The prevalence of donors with one type of UAs was 52.38%, with anti-E found the most common UAs at 33.33%, anti-Lea (14.28%), anti-M (4.76%) and 10 cases with unidentified UAs (47.62%) by using Capture-R Ready-Indicator Red cells kits from IMMUCOR (USA).

Summary / Conclusions: The prevalence of blood donors with unexpected antibodies was 0.11%. Anti-Lea, anti-M, anti-E were identified in donor samples which have important related to blood transfusion safety.

P530 | Undetected anti-D via column agglutination, identified by solid phase red cell adherence—a case report

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Background: The detection and identification of unexpected red blood cell (RBC) antibodies play a pivotal role in patient safety during a blood transfusion. Many institutions opt for an automated platform to perform this testing. Instruments on the market either employ column agglutination (CAT) or a solid-phase red cell adherence assay (SPRCA). The selection of blood bank automation can depend on several factors: turn-around time, ease of use, maintenance time, etc. However, the ultimate deciding factor should be patient safety.

Aims: The purpose of the studies presented was to find the best approach to immunohematology diagnostics for the patient, ensuring patient's safety.

Methods: The two cases presented are based on a retrospective examination of extant clinical and laboratory data, emphasizing discordant antibody identification. Within our institutional framework, CAT (Grifols, Barcelona Spain) constitutes the primary screening modality, complemented by SPRCA (Werfen, Barcelona Spain) as the principal supplementary approach.

Results: The first is a case report of a 32-year-old male individual who sought medical attention at our facility's emergency department subsequent to a traumatic incident. Due to the severity of the hemorrhage, four units of crossmatch compatible RBCs were ordered. The patient had no history of transfusion at our facility. The patient's blood type was determined to be A Negative with the specific Rh phenotype denoted as r (dce). In anticipation of the transfusion, a pretransfusion CAT antibody detection test yielded a negative result. Furthermore, a comprehensive CAT antibody identification panel employing papain-treated red blood cells was performed, with negative results. Follow-up investigations utilizing SPRCA were performed, and an anti-D was clearly identified. The second case is of a female patient, aged 87, who sought admission to our facility in anticipation of a scheduled surgical procedure. According to the patient's family, she had never been transfused. Her birthing

history is para 3, gava 2. The patient's blood type was determined to be O Negative with the specific Rh phenotype denoted as r' (dCe). In accordance with our procedures, the initial antibody detection test was performed in CAT with no unexpected RBC antibodies detected. This was further confirmed by negative results in two comprehensive CAT antibody identification panels. Supplementary investigations utilizing SPRCA clearly identified an anti-D. Upon the patient's readmission 27 days later, pretransfusion testing yielded consistent results: anti-D was identified in SPRCA but remained undetectable in CAT.

Summary / Conclusions: This pivotal outcome highlights the distinctive efficacy of SPRCA in detecting unexpected RBC antibodies that might escape detection with CAT. The findings accentuate the potential limitations of relying solely on conventional CAT methodologies, emphasizing the need for a more comprehensive and nuanced approach in antibody identification. The implementation of SPRCA as a primary screening modality or at least incorporating SPRCA into routine pretransfusion testing protocols could enhance the sensitivity of antibody detection, thereby contributing to improved patient care and safety. This case serves as a compelling argument for the reconsideration and potential revision of blood bank practices to prioritize the inclusion of SPRCA in antibody screening procedures for optimal clinical outcomes.

P531 | Case report of an un-expected antibody (anti-f) uncommon in Indian settings—confirmed by additional immuno-hematological work-up

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Background: The f-antigen is one of the Rh blood group system's compound antigens. There is RHCE*ce allele that encodes c, e and f antigen. The f antigen is expressed on RBCs with c and e on the same protein, e.g., on R₁r (Dce/ce), R₀R₀ (Dce/Dce) red blood cells (RBC). It is not expressed when c and e are on separate Rh proteins. e.g., on R₁R₂ (DCE/DcE) red blood cells. Anti-f is produced on exposure to the f (ce) antigen on RBC. There are very few cases reported worldwide since the presence of anti-f is often masked by anti-c or anti-e and is not generally found as a single antibody.

Aims: This is a case report of anti-f identified and confirmed in a 58-year-old female who came to our institute for a routine health check-up.

Methods: One EDTA sample was received for group and screen. Antibody screen and identification was done using 3-cell panel and 11-cell panel respectively. The identification of alloantibody was further supplemented by additional immuno-hematological workup.

Results: Blood grouping was reported as B RhD positive. Antibody screen came out to be positive, while auto-control was negative. Antibody identification done on the 11-cell panel showed the pattern of anti-f at AHG (Anti-human globulin) phase. The identification of anti-f was further supplemented by the following additional tests: (1). Rh antigen typing: On Rh antigen typing, the results were C+, E-, c-, e+. As the patient was negative for c-antigen, she is negative for f-antigen and can, therefore, develop allo-antibody against f-antigen. (2). Antibody identification panels: Enzyme (Ficin) treated panel: Antibody identification was repeated in parallel with "untreated panel cells" as control cells and "enzyme (Ficin) pre-treated panel cells" as reagent cells. Both the antibody identification panels showed the pattern of anti-f and there was an increase in the strength of the reaction from 3+ (untreated) to 4+ (enzyme-pre-treated). In-house constructed Rh panel: Authors constructed an in-house Rh panel and the patient's serum was tested with these panel cells. The reactions are summarized in Table 1.

(3). Look-back and phenotyping of donor RBC units: On tracing transfused RBC units, the phenotype of the two donors' red cells was found to be D+ C+ c- E- e+ and D+ C+ c+ E- e+. Transfusion of the second unit (D+ C+ c+ E- e+) might have led to the subsequent formation of anti-f in the patient. (4). Molecular typing: The molecular typing done predicted a phenotype of Rhe positive and Rhc negative, thus confirming the absence of f antigen.

Summary / Conclusions: The present case reports an uncommon allo-antibody against a compound antigen ce (f-antigen), expressed on the RBC when both c and e are present on the same haplotype. The identification of anti-f is uncommon as the presence of anti-f is often masked by anti-c or anti-e and is not generally found as a single antibody. In the case of need for transfusion, these patients should be provided with f-antigen negative blood. It is not imperative to give blood that is negative for both c and e antigens. Transfusion with either c-antigen negative or e-antigen negative blood would be adequate since all c-negative or e-negative individuals are also f-antigen negative.

P532 | Red blood cell alloimmunization in sickle cell patients in Kilifi, Kenya

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P531 - Table 1: In-house constructed Rh panel

Phenotype of panel cells	Genotype	Reaction
R ₁ R ₁	DCE/DCE	0
R ₂ R ₂	DcE/DcE	0
R ₁ R ₂	DCE/DcE	0
rr	dce/dce	3+
R ₀ r	Dce/dce	3+
R ₀ R ₀	Dce/Dce	3+

Background: Sickle cell anaemia (SCA) is a severe monogenic disorder with sub-Saharan Africa bearing 75% of the global burden. Blood transfusion is one of the key management strategies for management of anaemia and acute crises in people living with sickle cell anaemia. In Kenya alloantibody screening or extended matching beyond ABO and Rhesus D is not undertaken prior to transfusion and is likely to increase the risk of developing red cell antibodies against the unmatched antigens (alloimmunization). Blood for transfusion is a

limited resource hence the need to understand if alloimmunization may be contributing to repeat transfusions in sickle cell patients.

Aims: The study aimed at establishing the burden of red blood cell alloimmunization in SCA patients in Kilifi County.

Methods: The study was conducted at Kilifi County Hospital and utilized two designs; (i) a retrospective cohort of 106 participants with 322 transfusion events between 2002 and 2022 that had 255 archived plasma samples. (ii) a cross-sectional survey of recently transfused sickle cell clinic attendees ($N = 142$). Plasma samples were screened for alloantibodies and further analysed for alloantibody identification using the ID-Diacell and ID-diapanel cells (DiaMed GmbH; Switzerland, Bio-Rad).

Results: In the retrospective cohort, 54% were female and the median age was 4.9 (IQR, 2.3-7.9) years. The number of transfusions received ranged from 1 to 13 with the median age at first transfusion being 2.3 (IQR, 1.0-4.8) years. 24 (9.4%) samples were alloantibody positive with an alloimmunization rate of 13.2%. Eight alloantibodies were identified; anti -e ($N = 2$), -E, -M, -S, -s, -Lu^a & -Le^b. Five participants had pan-reactive alloantibodies, three had antibodies of unidentified specificities (AUS) and one had an autoantibody. 46% of the participants in the cross-sectional survey ($N = 142$) were female and the mean age range was 1-46 years. The prevalence of alloimmunization was 4.9% and five alloantibodies were identified (anti -e, -E ($N = 2$), -D & -Le^a). Two participants expressed pan-reactive and AUS respectively while an additional two had autoantibodies.

Summary / Conclusions: The rate of 4.9% observed in the cross-sectional survey was similar to the rates reported by other studies. The alloimmunization rate observed in the retrospective cohort was higher than previously reported in the East African region (2.9%-8%). This highlights the importance of study design in the determination and identification of alloantibodies. Further analysis is on-going to explore the risk factors for development of alloimmunization and the expression of minor blood group antigen phenotypes in the retrospective cohort and cross-sectional participants respectively.

P533 | The distribution of KEL blood group antigens among healthy blood donors in Saudi Arabia

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Background: Jazan Province of Saudi Arabia is common with inherited hemoglobinopathies, including sickle cell disease and thalassemia patient. Those patients may need frequent blood transfusion units. Therefore, compatible blood is required to administer in order to preclude any risk of red cell alloimmunization and hemolytic transfusion reactions. KEL blood group system is considered to be of clinical significance following the ABO and RH systems. It has many antigens that can lead to those issues.

Aims: This study aims to investigate the frequencies of the KEL blood group antigens (K, k, Kp^a, and Kp^b). Furthermore, the KEL phenotypes, K/k and Kpa/Kpb, were determined.

Methods: A total of 138 anonymous healthy Saudi blood donors from Prince Mohammed bin Nasser Hospital in Jazan Province, Saudi Arabia were enrolled to this study. Anticoagulated blood samples were taken and gel card technique was performed using antigen profile-II to investigate the K, k, Kp^a, Kp^b antigens.

Results: The antigens prevalence was as follows: K ($n = 9$, 6.52%), k ($n = 137$, 99.28%), Kp^a ($n = 1$, 0.72%), Kp^b ($n = 138$, 100%). The frequencies of the KEL phenotypes were K+k+ ($n = 8$, 5.80%), K+k- ($n = 1$, 0.72%), K-k+ ($n = 129$, 93.48%), Kpa+Kpb+ ($n = 1$, 0.72%), and Kpa-Kpb+ ($n = 137$, 99.28%).

Summary / Conclusions: We determined the prevalence of the KEL blood group antigens as well as the phenotypes in Jazan Province of Saudi Arabia. These outcomes may assist in establishing a national database of blood groups. In addition, it may reflect on transfusion practices by providing compatible blood units and avoid the risk of alloimmunization.

P534 | A2 subgroup of blood group A—a cause of discrepancies between forward and reverse ABO testing, a case report

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Background: ABO remains the most important blood group system in transfusion medicine. Errors in patient identification may lead to ABO-incompatible transfusions, so a full ABO group must be performed on all samples from first-time patients, with the exception of neonates. Approximately 80% of all group A (or AB) individuals are A1 (or A1B). The remaining 20% are A2 (or A2B) or weaker subgroups. Differentiation of A1 and A2 phenotypes is determined serologically using anti-A1 lectin. Anti-A1 antibody is detected in A2 group individuals in only 1% to 8% of cases. The subgroups of A can create apparent discrepancies between the red cell (forward) and serum (reverse) grouping results which can not only delay the transfusion but also contribute to errors. An incompatible ABO transfusion can be fatal.

Aims: Description of a case report of a patient with blood group discrepancy due to a subgroup of A in the AB blood group. A 58-year-old male patient went to the emergency department due to aggravated chronic diarrhea associated with acute kidney injury. He had a prior medical history of diabetes, recurrent pancreatitis and stable iron-deficiency anaemia, with no prior transfusion registry in our hospital. During hospitalization, there was a decrease in hemoglobin levels and required transfusion with a red cell concentrate unit.

Methods: Initially, pre-transfusional tests were performed with the anti-sera, panel and Card-ID system (BIO-RAD®), manually and in IH-500 equipment in EDTA-K3 venous blood sample. ABO reverse determination used commercially available known A1, A2, B and O cells. For subgroup identification, we used anti-H lectin monoclonal (BIO-

RAD©). LISS/Coombs (37°C) and NaCl (22°C) gel card systems were used to test the compatibility between patient sera and donor cells.

Results: ABO group test showed anti-A: 4+, anti-B: 0 and anti-AB: 4+ agglutination; however, reverse grouping test showed agglutination with A1 cells: 3+, B cells: 4+ and no agglutination with A2 cells and O cells. A discrepancy between the forward and reverse blood groups was verified. The patient tested positive for anti-D. The automated method gave similar results to the manual method. The patient's cell sample was performed with anti-H lectin, which a 4+ reaction at 22°C for 15 minutes. For control, anti-H was also tested with commercial A1, A2, B and O cells. Compatibility tests with group A units were carried out at 37°C and 22°C. They were negative at 37°C by manual and 1+ agglutination by automated methods at 37°C and both were 4+ with A units at 22°C. Taking into consideration the risk of haemolytic transfusion reaction and the limited red cell supply, it was decided to transfuse the patient with group O red cell concentrate.

Summary / Conclusions: This case shows a patient with subgroup A with antibody and investigation of an A subgroup. It advises that forward and reverse grouping should always be carried out. If a discrepancy is found, careful serological investigation should be performed. Improper subgroup identification can lead to wrong selection of blood for transfusion and delayed hemolytic transfusion reaction with consequent reduced post-transfusion cell survival. Modern genetic tests allows faster and safer identification and resolution, but depend on the availability of reference centers.

P535 | Hemolytic anemias due to warm antibodies experience in a tertiary hospital

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Background: Autoimmune hemolytic anemia (AHAI) due to warm antibodies is produced by antibodies generally of IgG nature, which react preferentially at 37°C. These antibodies behave in most cases like a panagglutinin, since they are directed against polymorphic epitopes that are present in all red blood cells. In some cases, the antibody may be directed against a specific antigen and thus the specificity of the autoantibody can be established. Autoantibodies directed against the e antigen (autoanti-e) have generally been reported.

Aims: To perform a retrospective study of the cases AHAI by warm antibodies in our center in order to know the specificity of the autoantibodies detected as well as to analyze the general characteristics of the patients.

Methods: A total of 124 patients with AHAI by warm antibodies were diagnosed in our center between January 1, 2013, and December 31, 2023. The study of the polyspecific Coombs test was performed using QuidelOrtho® Ortho BioVue System cards with subsequent

automated analysis in the Ortho-Vision equipment. The monospecific study was performed by manual technique, using Grifols® DG Gel DCscan cards. The eluate study was performed using the Immucor® ELU-KIT following the manufacturer's instructions. Resolve Panel QuidelOrtho® for identification of irregular antibodies were used to establish the specificity of the eluate.

Results: Of the 124 patients tested, 5 patients (4%) had autoantibody specificity. In 4 cases an anti-D autoantibody was identified (3%) and in the remaining case an autoantibody with anti-D+ anti-C specificity was identified (1%). In the remaining 119 cases (96%), the eluate revealed the presence of a nonspecific panagglutinin. Regarding the general characteristics of the series, 52% (N = 63) patients were female and 48% (N = 61) were male. The median age was 63 years (0-96). Sixty percent of the cases (N = 74) were classified as AHAI secondary to various processes. Among the different diseases detected, in 46% of the cases (N = 34) it was associated with lymphoproliferative syndromes, in 39% of the patients (N = 29), AHAI was detected in the context of different autoimmune diseases and in the remaining 15% (N = 11) the patients had been diagnosed with different solid neoplasms. In the remaining 40% (N = 50) a primary entity could not be identified and were classified as idiopathic AHAI.

Summary / Conclusions: Only 4% of the AHAI had an autoantibody with specificity, the most frequently detected being an anti-D autoantibody. The general characteristics of the series are similar to those previously reported. The differences in specificity in our series and what has been previously published could be justified by the small number of cases presenting specificity. It is useful to perform the autoantibody identification study in order to optimize transfusion in those patients who require it.

P536 | A comprehensive exploration of ABO Blood group and Rh(D) phenotypes frequencies at a single center in Pakistan for informed healthcare and beyond

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Background: Blood grouping is crucial in medical settings for transfusions and transplants, focusing on the ABO and Rh systems. The ABO system classifies blood into A, B, AB, and O based on genetically inherited antigens A and B on red blood cells. The Rh system, independent of ABO, distinguishes between Rh-positive (e.g., A+, B+) and Rh-negative (e.g., A-, B-) individuals based on the Rh factor (D antigen). Rh incompatibility, significant in pregnancy, may lead to hemolytic disease of the newborn (HDN), necessitating Rh immunoglobulin (RhIg) administration to Rh-negative pregnant women. Understanding ABO and Rh is essential in medical contexts to ensure transfusion compatibility and prevent complications in pregnancy, highlighting their pivotal role in safeguarding individual well-being.

Aims: To determine and classify individuals into distinct blood types based on the presence or absence of A and B antigens (ABO system) and the Rh factor D.

Methods: This cross-sectional study was conducted in a Chughtai Medical Centre from January 2023 to January 2024. 4845 Samples were received for Blood Group and Rh D Factor. The blood group and Rh factor D testing involves obtaining a blood sample from individuals through sterile procedures and collecting it in anticoagulant-containing tubes. ABO blood grouping reagents (anti-A and anti-B antibodies) and Rh blood grouping reagents (anti-D antibodies) are prepared according to manufacturer instructions. Using the tube method, the individual's blood sample is tested for ABO blood groups by adding anti-A and anti-B sera, with agglutination indicating the presence of specific antigens. Rh blood grouping involves testing for the Rh factor using anti-D serum. Weak D(Du) testing further confirms Rh Negatives. All Blood Groups undergo confirmation through repeat testing, with subsequent meticulous documentation. Quality control checks with known blood samples ensure testing reliability. Data analysis involves assessing the frequency distribution of ABO blood groups and Rh D factors, offering insights into population blood group diversity.

Results: 4845 samples were tested, the blood group distribution were as follows: Out of which A (24.6%), B (33.8%), O (32.7%), and AB (9.0%). 92.41% samples were Rh D Positive. In Rh D Positives ABO group distribution are as follows

- A (24.48%), A Sub (0.04%), B (33.59%), O (32.81%), AB (9.06%)
- 7.60% samples were RhD Negative distribution are as follows
- A (25.81%), B (35.86%), O (30.70%), AB (7.60%)

Summary / Conclusions: In conclusion, the analysis of 4845 samples from a single center in Pakistan revealed a notable distribution of ABO blood groups and Rh (D) phenotypes. B blood type dominated at 33.8%, followed by O (32.7%), A (24.6%), and AB (9.0%). A significant majority (92.41%) of samples were Rh D positive, with B being the most prevalent (33.59%) among them. Among the 7.60% Rh D negative samples, A blood type was predominant (25.81%), followed by B (35.86%), O (30.70%), and AB (7.60%). These findings underscore the importance of understanding regional blood group prevalence for informed healthcare decisions, particularly in transfusions and transplants. The data presented serves as a valuable resource for optimizing blood matching protocols and improving overall medical care in Pakistan.

P537 | Blood provision for a patient with anti-IH and subsequent laboratory improvements

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Background: Anti-IH is a complex antibody. It is typically benign in nature and has a preference for colder temperatures. Though clinically insignificant, Anti-IH can create a complicated serological

investigation which can lead to delay in the safe provision of blood. For Anti-IH formation there must be co-expression of both I and H antigens on the red cell surface. The antibody may be detected in individuals with A1B, A1 or B blood groups. Strength of reactivity depends on H antigen expression. H antigen expression is greater with O and A2 red cells when compared to other blood groups. Anti-IH has been implicated in cold agglutinin disease and more rarely in haemolytic transfusion reaction. The IH antigen is either not expressed or shows weak expression with cord red blood cells. Anti-IH is always present as Auto-antibodies that are enhanced by enzyme. They are IgM in nature and common in the serum of pregnant group A1 individuals.

Aims: A 23 year old female patient, at 33 weeks gestation was admitted to ICU with suspected sepsis and required urgent blood transfusion. The hospital reported pan reactivity in the antibody investigation panel. The aim is to present the serological work performed, the outcome for the patient and the laboratory process improvements for detection of this antibody going forward.

Methods: Standard serological and typing techniques were employed for ABO/D grouping, Rh/K typing, antibody investigation and compatibility testing by BioRad gel column technique. The presence of Anti-IH was confirmed by use of cord red blood cells along with rare reference cells and tube techniques.

Results:

Blood group: BRhD+, Phenotype: C-E-K-, IAT: Pan reactive 2+, ENZ-IAT: Pan reactive 3+, Saline 18°C: Pan reactive 4+, PWT 37°C: Weak reactions, Auto: Negative, DAT: Negative, Adsorptions: R1R1 (x3) Non reactive, r'r (x3) Non reactive, REST: Weak reactions, Compatibility: Incompatible IAT XM with O red cells, Compatible IAT XM with B cells, Saline RT XM incompatible, Rare SCARF cells: One Negative reaction obtained with a Hy-, Jo(a-) Js(a-) cell, Cord cells (oii): Negative, Bombay cells (Oh): Negative

Summary / Conclusions: The RCI were able to provide cross-matched blood for this patient, thankfully she recovered from her sepsis and left the ICU. She safely delivered her baby by ELCS at 33 weeks. Although the investigation of such an antibody can take a considerable amount of time it is necessary to circumvent transfusion reaction. Anti-IH can have potentially catastrophic consequences owing to the timely investigation and expertise devoted to identifying such an antibody. There are however techniques that should be employed in an opportune manner to ID this antibody. The use of cord and Bombay red cells should be engaged early in an investigation where Anti-IH is suspected. Thermal amplitude studies are very useful for demonstrating the antibodies preference to react at lower temperatures (18°C) than higher temperatures (37°C). Group B and O control cells should be used for saline investigations. These techniques should be utilised first before time-consuming adsorption studies. ABO identical units should be selected where possible as these units have the best chance of being compatible for a patient with a suspected Anti-IH. The lengthy investigation of this antibody has resulted in an improved laboratory process for identifying anti-HI going forward, which will ensure quicker blood provision.

P538 | Immunohematological complications in beta-thalassemia major adult patients

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Background: Patients with beta-thalassemia major (BTM), require life-long recurrent transfusions. BTM and its treatment can be responsible for metabolic, immunological or overload complications, which can jeopardize vital prognosis, function and quality of life.

Aims: The aim of our work is to describe the hematological complications of adult polytransfused patients with BTM.

Methods: This was a monocentric descriptive longitudinal study enrolling adult patients with BTM followed at the biological haematology department of La Rabta Hospital. Patients were recruited over a 6-month period. We took the medical history from the medical records and performed immunohaematological tests: direct coombs test and irregular antibody test.

Results: We included 43 patients. The sex ratio was 1.15 and the mean age was 27.3 ± 4.8 years. Mean pre-transfusion hemoglobin was 7.9 g/dL [5.4-9.8]. Autoimmunization was found in 47% of patients, alloimmunization in 7% and an auto and alloimmunization in 12%. Positive direct coombs test was more frequent in patients with age at first transfusion less than 12 months ($p = 0.035$). Positive direct coombs test was more frequent in splenectomized patients than in non-splenectomized patients, $p = 0.025$. There was no statistically significant relationship between first transfusion at an age < 12 months and the presence of alloimmunization ($p = 0.407$). The mean number of packed red blood cells transfused per year was 25 per year, with extremes ranging from 20 to 30 per year. The most represented number of packed red blood cells transfused per year was 24 per year in 37% of patients ($N = 16$). The mean interval between transfusions was one transfusion of two packed red blood cells every 14 days, with extremes ranging from 10 to 17 days.

Summary / Conclusions: BTM is responsible for various complications inherent to the disease or its treatment. Multidisciplinary care and early screening of these complications would improve the quality of life and monitoring of patients.

P539 | Frequency of blood groups with clinical importance in blood donors at Hospital de Clinicas, Montevideo Uruguay

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Background: The fundamental objective of transfusion medicine is to provide high quality blood components and products safely to all patients. Although institutionally we have established blood donor selection criteria, standard operating procedure for different processes of the blood transfusion process and an hemovigilance

program has been implemented, we cannot exempt transfusion itself from possible risks, such as transfusion reactions. Aiming always at the patient and donor's safety, blood samples are routinely taken and both serological and immunohematological tests are performed. We consider it is fundamental to know how clinically significant antigens of blood group systems are distributed in our population (especially our blood donors) to accurately determine the probability of finding blood units with certain antigenic characteristics to prevent possible immunizations due to exposure to non-self antigens. In addition, developing an investigation of the prevalence of clinically significant blood group antigens in voluntary donors, collaborates in characterizing the stock that blood banks have and enabling a more rational use of it. Our country does not have a centralized system for Blood Banks at the national level, where this information about the blood group's prevalence is reflected, so we must turn to international data. Through this work we seek to generate statistical data showing the antigenic frequencies of the main blood group systems, selecting the population of blood donors from the "Hospital de Clínicas" in Montevideo, Uruguay as target population.

Aims: General objective: Study clinically significant blood groups in blood donors. Specific objectives: Determine the frequency of the most important antigens of ABO, Rh, Kell, Kidd and Duffy blood group systems in blood donors.

Methods: This is a cross-sectional descriptive study, in which a total of 1127 blood donors from the blood bank of the Hospital de Clínicas were studied. The procedure was performed together with the extraction of routine samples for serological and immunohematological studies, with prior authorization from the donor via informed consent. Prior to the study of samples, an inventory of the available reagents

P539 - Table: Results.

Antígeno	Frequency obtained	Daniel's Frequency
A	37.00%	40.00%
B	8.80%	11.00%
AB	3.20%	4.00%
O	51.00%	45.00%
D	87.50%	85.00%
C	69.30%	68.00%
c	76.30%	80.00%
E	29.80%	29.00%
e	98.20%	98.00%
Cw	1.40%	2.00%
Kell (K)	4.00%	10.00%
k	100%	99.80%
Fya	55.80%	68.00%
Fyb	77.20%	80.00%
Jka	77.30%	76.00%
Jkb	80.60%	74.00%

was made and quality controls were thoroughly checked on them, allowing their use to carry out the project. The results were recorded in paper and digital format, using an Excel spreadsheet system and then, depending on the absence or presence of the antigen, the frequency of the blood group antigens mentioned above and the frequency of the phenotypes found in the blood donor population were calculated.

Results:

Summary / Conclusions: The results obtained were entered into a table shown below and compared with the antigenic frequency described in the international bibliography. The data obtained was contrasted with the described antigenic frequency in the international bibliography, not showing significant changes in the comparative prevalences, reaffirming the European migrant origin of our population.

P540 | Abstract withdrawn

P541 | Anti-D plus anti-C or anti-G? A diagnostic challenge

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Background: The G antigen, belonging to the Rh system, is present in more than 85% of the caucasian population, especially in individuals who are RhD and/or RhC positive. The reactivity for antigen G is determined by the Ser103 residue, encoded by RHD and the C allele of the RHCE genes. For this reason, from a serological point of view, the Anti G antibody presents a specificity coincident with an anti-D plus anti-C, and it is capable of causing hemolytic disease of the fetus and newborn, usually milder than that caused by anti-D and/or anti-C. The anti-G antibody is an antibody formed in almost all cases by D-negative, G-negative patients with the genotype rr (dce) who have been exposed to an immunizing event. The clinical implication lies in differentiating the presence of anti-G or anti-D plus anti-C antibodies, as it is essential to prevent maternal alloimmunization by anti-D, allowing for the optimization of the use of anti-D immunoglobulin, as well as defining the transfusion of packed red blood cells, if necessary. We describe the case of a 46-year-old female patient, caucasian ethnicity, group A Rh(D) negative, with an immunological history of 5 pregnancies, who had received two transfusions of A Rh(D) negative red blood cells in the past is described. During a new routine study, an antibody with specificity coincident with a probable anti-D + anti C was discovered. Immunohematological studies were performed to determine whether it corresponded to an anti-G antibody or an anti-D antibody plus an anti-C antibody.

Aims: Differentiate the specificity of the antibody between an anti-G or an anti-D antibody plus an anti-C antibody.

Methods: This is a descriptive observational study, clinical case report type. Differential adsorption/elution (cloroquine) techniques were

performed initially with R2R2 cells and subsequently with r'r cells in order to distinguish the presence of Anti G from the presence of Anti D plus Anti C. For antibody identification, gel microtechnique ID-Card LISS /Coombs were performed following the manufacturer's instructions (BIO-RAD (MR) IDDiapanel Set of 11 vials for IAT and NaCl test. LOT 859202 77 1 Expiration 13/11/2023).

Results: The presence of an anti-G antibody plus an anti-C antibody is identified in the patient plasma, ruling out the presence of anti-D.

Summary / Conclusions: The adsorption/elution technique is an effective method to allow for the differential diagnosis between an anti-G antibody and an anti-D plus anti-C antibody, which favors the correct clinical management of these patients.

P542 | Abstract withdrawn

P543 | Abstract withdrawn

P544 | Treatment of anaemia in a mother with a rare phenotype and two alloantibodies developed—anti-C and anti-e

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Background: Anaemia is the most common medical complication in pregnancy. If the pregnancy is not controlled, this anaemia can manifest itself during and after childbirth, thus directly endangering the life of the mother and potentially the newborn.

Aims: To show how a woman who has just given birth was cared for when a rare phenotype and two alloantibodies were proven only after delivery.

Methods: Since the frequency of this phenotype in the population is 2.10%, we had to type a large number of erythrocyte doses to find an antigen-negative match for therapy. Typing was done manually on the plate, and negative doses were confirmed by the test tube method. Screening and identification of antibodies was done by the gel method on the IH-500 device with BioRad's erythrocyte panel.

Results: Out of a total of 103 serotyped blood doses, only four units were compatible, of which the mother received two doses. From this third uncontrolled pregnancy, a male child was born, blood type: BRhD+ with a CcEe phenotype, while the DAT was negative on the third day after delivery when we finally got the child's sample. We found one blood dose of ORhD-, ccEE phenotype, but the child did not receive a transfusion, but the elevated bilirubin values were corrected by UV lamp therapy.

Summary / Conclusions: It is necessary to perform screening of all pregnant women, not only RhD-, in order to avoid these situations and to be ready with appropriate typed blood doses in case of an emergency positive screening. In this way, the morbidity/mortality of both the mother and the child would be reduced.

P545 | A patient with red blood cell reactive antibodies against CD36 which could be identified with a platelet specific antibody detection assay

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Background: The CD36 protein is expressed on platelets and known as glycoprotein (GP)IV or the NAK antigen. This antigen has a high (> 99%) prevalence and is absent (type 1 deficiency) in few people from African or Asian descent. CD36 is expressed by various cell types, including monocytes and platelets and cells of the erythroid lineage, but with very low amounts on mature red cells. CD36 was recently ratified as a red cell blood group system (ISBT045) by the ISBT. Antibodies against CD36 have been reported to cause fetal neonatal alloimmune thrombocytopenia (FNAIT), platelet transfusion refractoriness and hemolytic disease of the fetus and newborn (HDFN). Because of the low expression on mature red blood cells, these antibodies are hard to detect with standard red blood cell immunohematology methods.

Aims: To confirm the presence of anti-CD36 antibodies with a more sensitive method.

Methods: Hemagglutination testing was performed with standard tube and gel column IAT methods. Antigen typing was performed with commercial or inhouse developed reagents. Platelet specific antibodies were detected with the Pak LX assay from Immucor.

Results: A female of African origin was routinely tested during her first pregnancy and antibodies were detected that were weakly reactive with all except her own red blood cells. The presence of an antibody against a high prevalence antigen was suspected. Extensive investigations to determine the antibody specificity, during three pregnancies, remained inconclusive. Interestingly the patient was found to have the rare Jo(a-) phenotype, however the presence of anti-Jo^a was excluded. Three healthy children were born, without any signs of HDFN, in each consecutive pregnancy. During her fourth pregnancy the antibodies were still present and weakly reactive with red blood cells. With newly gained information on CD36 expression on red blood cells the possibility of anti-CD36 specificity was investigated. The red blood cells and platelets of the patient were found to be CD36-negative, and the antibodies were nonreactive with CD36-negative red cells. Since the red cell antibodies were only weakly reactive, a more sensitive technique was sought to confirm the CD36 specificity. The PAK Lx assay is used to detect anti-platelet antibodies and includes beads specifically expressing GPIV/CD36. The antibodies of the pregnant woman showed strong reactions with these beads which confirmed the CD36 specificity.

Summary / Conclusions: The use of a platelet antibody specification assay (PAK Lx) made it possible to improve the sensitivity for detecting the presence of anti-CD36 antibodies that are only weakly reactive in standard red blood cell serological methods. Because anti-

CD36 can cause HDFN and FNAIT, the woman has been referred to a specialized perinatal center.

P546 | Abstract withdrawn

P547 | Performance evaluation of an immunohaematology analyzer in Central Blood Transfusion Service Indonesian Red Cross

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Background: Pre-transfusion testing includes serological testing such as blood type, antibody screening, and cross-matching in the laboratory. There are two methods for pre-transfusion examination, tube test, and CAT (Column Agglutination Test). Media in the CAT there are 2 types, sephadex gel and glass beads. Vitros Ortho Vision Immunohematology use glass beads

Aims: To determine the performance and suitability of tools and reagents in interpreting the results of pre-transfusion blood tests

Methods: 4 parameters were examined, blood group examination of 100 samples, crossmatch examination of 300 samples, antibody screening examination of 100 samples, and DCT examination of 100 samples.

Results: Based on the results of the examination, the results obtained are for blood group examination a total of 100 samples consisting of blood group A positive of 17 samples, B Positive of 27 samples, O Positive of 33 samples, AB positive of 13 samples, and B negative of 10 samples, all in accordance. In the Crossmatch examination, 100 samples were incompatible and 200 samples were compatible with fully compatible results. The antibody screening consisted of 27 positive and 73 negative samples with all corresponding results. DCT (Direct Coombs Test) consists of 58 negative samples, 36 positive samples on IgG, and 6 positive samples on C3. All examination results are in accordance with the comparison using the standard purpose of examination using a tube test.

Summary / Conclusions: Based on the results of the examination of 4 tests, blood group examination, crossmatch examination, antibody screening examination, and DCT (Direct Coombs Test) in accordance with the results tested by tube test method or as a goal standard for immunohematology

P547-A | Abstract withdrawn

P547-B | Transfusion challenges in patients treated with magrolimab: A case report

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Background: Magrolimab is a humanized monoclonal IgG4 anti-CD47, used in clinical trials for malignancies treatment. CD47 blockade

inhibits its antiphagocytic signal, enhancing cancer cell clearance by macrophages. Since CD47 is highly expressed on red blood cells (RBC), anti-CD47 drugs can lead to anemia and interfere in pretransfusion (pretx) testing by causing nonspecific panagglutination in ABO typing and antihuman globulin (AHG) tests. Mitigation strategies include: perform baseline ABO typing, extended pheno- or genotyping and irregular antibody (IA) screening before starting Magrolimab; use monoclonal AHG lacking anti-IgG4 in indirect antiglobulin tests; perform RBC adsorptions. A recent study on adsorption using Burkitt lymphoma-derived Daudi cells presented promising results.

Aims: Case report of a patient treated with Magrolimab, displaying altered pretx tests.

Methods: Review of the patient's medical and analytic record. Immunohematology and pretx tests were performed using the column agglutination technique and an automated immunohematology analyzer (IH-1000 Bio-Rad), namely ABO/RhD typing (ABO/Rh for Patients ID-Card, NaCl ID-Card and ID-DiaCell ABO, Bio-Rad), Rh/K phenotyping (Rh-Subgroups+Cw+K ID-Card, Bio-Rad), IA screening (LISS/Coombs ID-Card and ID-DiaCell I-II-III, NaCl ID-Card and ID-DiaCell IP-IIP-IIIP, Bio-Rad), Direct antiglobulin testing (DAT) and crossmatch (LISS/Coombs ID-Card, Bio-Rad). IA screening and crossmatch were also performed using the tube method and a monoclonal AHG (Anti-IgG murine monoclonal, Werfen).

Results: A 54-year-old patient diagnosed with hypopharynx carcinoma, with no transfusion history, was treated with chemotherapy + radiotherapy, achieving metabolic remission. Due to disease progression, he was included in a clinical trial, starting treatment with Carboplatin + 5-Fluorouracil + Pembrolizumab + Magrolimab (day 1, 8, 15). A previous immunohematology study revealed: Group AD cEe, K negative; IA screening and DAT were negative. On D1 of Magrolimab, the pretreatment blood count was: Hb 10.7g/dL, MCV 97.9fl, RDW 14%; and posttreatment: Hb 7.6g/dL (remaining parameters not measured). On the following day, reassessment blood count was: Hb 9.7g/dL, MCV 102.7fl, RDW >25%. The patient remained asymptomatic and hemodynamically stable. On D15, due to Hb 6.6g/dL, RBC transfusion was requested. Pretx tests showed: AD (forward group); nonspecific panreactivity (4+) in reverse group and IA screening; incompatible crossmatch (4+); negative DAT. IA screening and crossmatch performed by tube method, using AHG lacking anti-IgG4, showed persistent panagglutination. Since the Transfusion Service (TS) was not previously informed that the patient would start Magrolimab, extended pheno- or genotyping had not been performed. RBC units matching patient's historical ABO Rh Kell phenotype were issued. Over the course of 4 months the patient received 13 packed RBC with no transfusion reactions. Later on, he died due to disease progression.

Summary / Conclusions: Pretx laboratory findings were concordant with anti-CD47 interference. This case addresses the importance of communication between clinicians and TS, and to obtain a pretherapy sample for baseline immunohematology studies, with aim to provide extended antigen-matched units and prevent alloimmunization. AHG lacking anti-IgG4 wasn't effective, raising the need to implement new laboratory techniques to ensure safe and timely transfusions.

Immunohaematology—red cell immunohaematology: molecular

P548 | Anti-Fy3 as a game of chance lowering the odds

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Background: ACKR1 encodes a transmembrane glycoprotein, previously known as DARC, that carries the five antigens of the Duffy blood group system. The antithetical antigens, Fya and Fyb, are encoded by codominant *FY*01* and *FY*02* alleles, which differ by a single nucleotide polymorphism (SNP), c.125G>A. This missense mutation produces a codon for glycine in the *FY*01* allele and a codon for aspartic acid in the *FY*02* allele at position 42 (p.Gly42Asp). The other three antigens – Fy3, Fy5, Fy6 – are conformational epitopes as opposed to sequence-specific epitopes, therefore lacking antithetical antigens. In individuals of African ancestry, the Fy(a-b-) phenotype is most common, caused by homozygosity for a silenced *FY*B* allele (*FY*02N.01*), the GATA box mutation. Very rarely Europeans and Asians with Fy(a-b-) red blood cells (RBC) are homozygous for inactivating mutations with premature stop codon. These mutations in homozygosity prevent Duffy antigen expression on any cell in the body and thus are true Duffy null phenotypes. Consequently, these individuals are at risk of being alloimmunized when exposed to RBC expressing FY antigens. The *FY*02N.01* allele in people of African ancestry encodes the Fyb antigen but is silenced by a point mutation in the promoter region. This SNP disrupts the binding site for the erythroid-specific GATA-1 transcription factor and prevents ACKR1 gene transcription on erythrocytes only while permitting expression on nonerythroid cells. Consequently, individuals carrying at least one copy of the *FY*02N.01* allele can be transfused with Fy(b+) RBC units because they are not at risk of developing anti-Fyb. However, on rare occasions they make unexpected anti-Fy3. This antibody is predominantly of the IgG1 subclass and has been responsible for acute and delayed hemolytic transfusion reactions.

Aims: Assess the inability of Sickle Cell Disease (SCD) patients with a Fy(b-) phenotype secondary to FYB-GATA mutation to produce anti-Fyb or anti-Fy3.

Methods: We followed Fy(b-) SCD patients, chronically transfused, who were given Fy(b+) RBC units. The patients' genotype was *FY*01/FY*02N.01* or *FY*02N.01/FY*02N.01*. Phenotyping was performed by gel test () and FY/GATA genotyping was performed by a reference laboratory.

Results: The SCD patients did not make anti-Fyb. However, a few of them made potent anti-Fy3, with antibody specificity verified at a reference laboratory.

Summary / Conclusions: These results show that although Fy(b-) SCD patients carrying the silenced *FY*B* allele can safely receive Fy(b+) RBC because they do not make anti-Fyb, the same is not always true for anti-Fy3. Since Fy3 is exclusively conformational and not a

sequence-specific antigen, we conclude that most of the times the patient's immune system tolerates the tertiary structure of this amino acid sequence expressed on the transfused red cells. This immunological tolerance shall be induced by the histologic expression of Duffy protein. These individuals have a Fy(b-) RBC phenotype, but they are tolerized by the Fyb antigen expressed in their nonerythroid tissues, despite potential slight differences in its spatial assembly. However, with the patient being chronically transfused, someday his immune system could find donor erythrocytes whose Fy3 three-dimensional conformation would appear different enough to be recognized as nonself, resulting in alloimmunization and consequent production of anti-Fy3.

P549 | Feasibility study of the use of red cell and platelet genotyping assays with a new multiplexing platform

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Background: BLOODchipID is a high-throughput platform for blood groups genotyping based on Luminex[®] xMAP technology (Luminex[®] 200™). The ID CORE XT, ID HPA XT and ID RHD XT are multiplex assays for the simultaneous detection of genetic variants that provide predicted phenotypes of the red blood cell and platelet antigens.

Aims: The Luminex[®] FLEXMAP 3D[®] System is a more versatile instrument with some user-friendly features for end-users in blood transfusion centers. In order to evaluate the usability and the compatibility with the new instrument, several analytical tests were carried out.

Methods: ID CORE XT (including ID CORE CONTROL), ID HPA XT, and ID RHD XT assays were tested with positive and negative controls and a panel of reference samples that covers the genetic variants interrogated by each assay, using two FLEXMAP 3D[®] instruments in comparison with the currently used Luminex 200 system.

Results: Regarding usability, FLEXMAP 3D was considered easy to be used in the laboratory with the BLOODchipID platform; and a 96-well plate read time was reduced by more than half. Regarding performance, the results obtained in genotypes and predicted phenotypes were 100% correct for all samples tested in comparison with the current instrument Luminex 200. However, some adjustments will be necessary for ID HPA XT to be compatible with the FLEXMAP platform.

Summary / Conclusions: The use of the BLOODchipID platform with FLEXMAP 3D[®] System was feasible. Further validation studies will be performed to confirm the safety and performance in the new instrument.

P550 | Validation and performance of a new software version for data management of red cell and platelet genotyping assays

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Background: The progression in blood group genotyping technologies demands robust software solutions for data management and analysis. BLOODchipID Software (BIDS XT) represents a significant step forward, integrating with ID CORE XT, ID HPA XT and ID RHD XT Analysis Software to organize, manage and/or transfer sample information and data results and reports. A new software version (BIDS XT v3) has been developed to include usability and cybersecurity improvements.

Aims: The primary aim of this study is to assess the functionality, reliability, and user-friendliness of BIDS XT v3 in conjunction with BLOODchipID assays, ensuring its suitability for diagnostic laboratories' demanding environments.

Methods: The study involved testing BIDS XT v3 software functional requirements, including installation, configuration, batch processing, result accuracy, and system communication. This was conducted through multiple ID CORE XT, ID HPA XT and ID RHD XT runs with both positive and negative controls and a panel of reference samples to simulate real-world laboratory conditions and assess the software's performance across a range of situations.

Results: BIDS XT v3 demonstrated a 100% match between its results and those from the corresponding Analysis Software and the previous version of BIDS XT. The software met all functional requirements, successfully managed batch processing, and ensured accurate result reporting without significant anomalies. The software proved to be user-friendly, facilitating streamlined workflows, reliable control management, and effective communication with Luminex and Laboratory Information Systems.

Summary / Conclusions: The validation of BIDS XT v3 confirms its efficacy and reliability as a comprehensive data management solution for blood genotyping laboratories using BLOODchipID products.

P551 | Deletion of c.241_243TTC in A4GALT associated with p phenotype

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Background: The phenotype p in the P1PK blood group system is infrequently reported but holds clinical significance. These occurrences generate naturally-occurring anti-PP1Pk antibodies, resulting in acute hemolytic transfusion reactions (AHTR). Different mutations in the A4GALT gene lead to the p phenotype. Identifying it through serological and molecular biology methods is crucial, followed by adapting a blood transfusion plan accordingly.

Aims: To determine why a donor's blood isn't compatible in cross-matching, identify antibodies to high-incidence antigens and elucidate the genetic background of the p phenotype in the P1PK blood group system through Sanger sequencing.

Methods: Blood typing was conducted using gel cards. Antibody screening and identification were carried out using the conventional tube method and anti-human globulin in gel cards. Cross-matching

was performed to assess compatibility with other donor blood samples. The presence of the P1 antigen was confirmed using monoclonal anti-P1 antibodies. Sequencing of A4GALT and B3GALNT1 exons was accomplished using the Sanger method.

Results: The 48-year-old blood donor was typed as A and RhD positive. In cross-matching with numerous other donor samples, major compatibility was negative, while minor compatibility displayed strong agglutination and severe hemolysis (4+H). Antibody screening and identification revealed the presence of anti-PP1Pk antibodies. These antibodies exhibited no reaction with p red blood cells and had a titer of 1:64. The donor's RBCs tested negative for the P1 antigen. Sanger sequencing identified a homozygous A4GALT genotype as c.241_243delTTC/c.241_243delTTC. Genotyping of B3GALNT1 showed no mutations.

Summary / Conclusions: The donor's RBC phenotype was determined to be p by serological testing. The sequenced allele with c.241_243delTTC in A4GALT was previously reported and designated as A4GALT*02N.01.02 in P1PK blood group alleles by ISBT. The homozygous genotype of this case resulted in p.Phe81del in amino acid sequence, potentially deactivating the enzyme galactosyltransferase. The tested coding sequence was assigned an accession number OR900206 in NCBI GenBank. Conducting a comprehensive analysis of the pedigree and gathering a larger pool of comparable cases to elucidate the genetic and ethnic background, as well as studying the transcription and translation processes, would be interesting and meaningful when samples are available in future.

P552 | Three new XK alleles accounting for the McLeod phenotype in the Kx blood group system

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Background: Kell glycoprotein and Xk protein, which are encoded by the KEL and XK genes, respectively, are linked by disulfide bonds on the red blood cell (RBC) membrane. The McLeod phenotype, an extremely rare Kx-deficient phenotype, is caused by a mutation of the XK gene, which also affects Kell glycoprotein expression.

Aims: We report the details of three novel XK alleles in blood donors with the McLeod phenotype who were identified during routine donor screening.

Methods: Japanese blood centers routinely screen for the rare K_o and McLeod phenotypes using an automated blood typing system (PK7300) with in-house mouse monoclonal anti-Ku (CBC-117) or anti-K14 (CBC-120) antibodies. When negative or weak agglutination patterns were observed, further tube tests using Kell-related antibodies and immunoblotting using in-house mouse monoclonal anti-Kx antibodies (CBC-365) were performed. The XK gene was analyzed by polymerase-chain reaction and Sanger sequencing.

Results: Three blood samples from male blood donors with the McLeod phenotype were identified by serology and immunoblotting. All three RBC samples showed weak agglutination with anti-k, -Kp^b, -Js^b, and -Ku as compared with control RBCs, and no irregular antibodies were observed in their plasma. The apparent band at approximately 37 kDa, which corresponds to the Xk protein, was not observed by immunoblotting. Sanger sequencing of the XK gene revealed that one individual had a missense single nucleotide variant (SNV), c.245+5G>A. Another individual had an 8-bp insertion, c.885_892dupGTCTGCTG (p.-Val298Glyfs*6). The remaining individual had a 4617-bp deletion of g.9063-g.13679, including the entire exon 2. The c.245+5G>A SNV might alter splicing of the donor site of exon 1, which is a similar phenomenon to that in the allele known as XK*N.25 (c.508+5G>A) with aberrant splicing. The c.885_892dupGTCTGCTG (p.Val298Glyfs*6) insertion generates a stop codon in exon 3. The g.9063-g.13679 deletion includes the entire exon 2, which is a similar phenomenon to that in the XK*N.04 allele. However, the deletion size reported in XK*N.04 is different (7453-bp, g.89705-g.97158).

Summary / Conclusions: We identified three novel XK alleles, c.245+5G>A, c.893ins (p.Val293Glyfs*6), and del Exon 2 (p.Arg82_Ser170del), from Japanese blood donors with the McLeod phenotype.

P553 | Role of decentralized molecular testing devices in immunohematology

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Background: Allo-immunization remains a major risk in immunohematology that can complicate red blood cell transfusion, especially for patients requiring chronic transfusions. Conventional pre-transfusion testing through serological methods may be inadequate in certain clinical situations. Molecular typing offers an alternative method, allowing for the prediction of erythrocyte phenotype from genotype. However, current platforms are more suited for large-scale screening and require highly skilled personnel. Additionally, they entail long processing times and are generally not performed on-site, limiting their application in emergency situations. When quick decision-making is necessary, tests that can be conducted nearby or on-site can be advantageous.

Aims: We have developed a lateral flow strip for rapid visual detection of seven alleles from three clinically relevant blood group systems (FY*01, FY*02, FY*02N.01, GYPB*03, GYPB*04, JK*01, and JK*02),

with an execution time of approximately 1 hour. Our test is tailored for use in immunohematology laboratories, such as pre-transfusion testing for patients when serological methods have failed or in emergency scenarios.

Methods: Ninety-eight blood samples were obtained from patients at the Immunohematology Laboratory (Montpellier, EFS Occitanie). Samples were processed with our LFD assay to determine genotypes and to predict phenotypes. The obtained results were then compared with those derived from serology and the ID CORE XT blood group genotyping assay (Progenika Biopharma-Grifols).

Results: All samples were successfully genotyped using the LFD assay, with a concordance rate of 99.7% compared to standard hemagglutination. However, two predicted phenotypes involving the FY and MNS blood group systems showed discrepancies. In the FY blood group system, one patient with FY*01/*02 genotype according to our LFD assay was typed as FY:1,-2 via serology. Further analysis with the ID CORE XT blood group genotyping assay revealed the presence of an FY*02M.01 allele, resulting in a weak expression of the FY2 antigen (Fyx) that was missed by serology. In the MNS blood group system, one sample was genotyped heterozygous GYPB*03/GYPB*04 using our molecular assay, but exhibited an MNS:-3,4 phenotype. Subsequent analysis with the ID CORE XT assay identified the presence of a silent GYPB*03N.03 allele, which was not assessed in our assay.

Summary / Conclusions: Our assay provided precise predictions of phenotype for FY1, FY2, S, s, JK1, and JK2 antigens in clinical samples, demonstrating no invalid results, unlike the 19.7% rate observed with the ID CORE XT assay. Additionally, our panel of SNPs was enhanced to detect silent GYPB*03N.01 and GYPB*03N.03 alleles. With its user-friendly operation and quick turnaround time, the LFD assay shows great potential for adoption in clinical laboratories, especially for patient management scenarios where serological typing fails to provide conclusive results or essential information.

P554 | Genetic background of anti-CD99 producers in Japanese

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Background: The Xg blood group system is comprised of two antigens, Xg^a (XG1) and CD99 (XG2) encoded by two closely linked homologous genes XG (X chromosome) and CD99 (X and Y chromosomes) located on the pseudoautosomal region 1 (PAR1). XG on the Y chromosome is disrupted because of lacking the exons 4 through 10. The expressions of Xg^a and CD99 are coregulated by a single nucleotide polymorphism (SNP) called rs3111031 in the GATA-1 binding region. Anti-Xg^a producers were males with a 114 kb genomic deletion on the X chromosome including the XG gene (esv2662319). Anti-Xg^a is likely a naturally occurring antibody,

because the producers have never been transfused. On the other hands, all anti-CD99 producers were females with a history of pregnancy. To date, five CD99- individuals with anti-CD99 had the molecular bases characterized. Four individuals were homozygous for various deletions spanning exon 2-8 in their CD99, and one was homozygous nonsense mutation in CD99. We identified five unrelated anti-CD99 producers in Japan from 1992 to 2023.

Aims: We investigated the genomic background of the anti-CD99 producers in Japan. This study was approved by the ethics committee of the Japanese Red Cross Society (#2023-018).

Methods: Standard serological techniques were used. Antiglobulin tests were read in tubes after centrifugation using anti-human IgG. Genomic DNA was obtained from five anti-CD99 producers using a QIAamp DNA Blood Mini Kit (Qiagen, Tokyo, Japan). To identify the deleted region, we conducted PCR with the following primer pairs; specific for CD99 intron 2, CD99 exon 5, XG exon 1, XG exon 8, GYG2 exon 1, and GYG2 exon 3. Long PCR using forward primer of specific for CD99 intron 2 and reverse primer of specific for GYG2 intron 3 was performed using PrimeSTAR[®] GXL DNA Polymerase (Takara, Shiga).

Results: Five archived antibodies gave the expected strength variation with the cells of predicted high and low CD99 expression in the anti-globulin test. IgG subclass of the five samples was IgG1. We confirmed that all anti-CD99 producers retained CD99 intron 2 and GYG2 exon 3 but lost CD99 exon 5, XG exon 1&8, and GYG2 intron 1. Next, we tried to amplify the predicted deleted region. The length was 127 kb normally. Interestingly, all five anti-CD99 producers showed a clear 12 kb band, whereas the normal controls showed only nonspecific faint bands. We sequenced the 12 kb PCR products and identified the deleted region chrX:2,717,469-2,832,051 (114,583 bp) on the human GRCh38/hg38 Assembly. This deletion results in the loss of a nearly 115 kb on the X chromosome that spans CD99 exons 2 through 11, total of XG and GYG2 exon 1. All five producers were confirmed homozygous for a large deletion allele. No anti-Xg^a were detected in the sera of the five anti-CD99 producers, although the entire gene encoding XG was deleted.

Summary / Conclusions: We identified a large deletion of approximately 115 kb including CD99 and XG on the X chromosome, resulting in CD99- phenotype with anti-CD99 production. Interestingly, no anti-Xg^a were detected in the five anti-CD99 producers. All five individuals were from the Kyushu prefecture (Southwestern island of Japan), so this deletion allele may be endemic to this region. Although anti-CD99 may have been produced during pregnancy, it was not confirmed as the causative of the hemolytic disease of the fetus and newborn.

P555 | Genetic variants in the CD59 blood group system—an exploratory reference genome dataset study

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Background: The CD59 glycoprotein is highly expressed in the membranes of many cell types. It protects the cells from complement-

mediated cell lysis by inhibiting the assembly of complement membrane attack complex. Lack of a functional CD59 protein have been shown to cause intellectual and developmental disabilities, recurrent ischemic strokes, neuropathy, and chronic hemolysis. Recent large scale sequencing projects are generating datasets of human variation across global populations. Worldwide differences in the distribution of genetic variants can affect a multitude of diseases at the population level. Understanding global genetic diversity can provide insights into the mechanisms of underlying disease and help develop public health decision-making models. Variants in the *CD59* gene have not been collated and compared for their deleterious and disease-causing effects.

Aims: We aimed at a comprehensive analysis of *CD59* gene from publicly available databases to identify population-specific variants. Bioinformatic analysis will aid in the discrimination between deleterious and neutral variants and their prioritization for pathological investigation.

Methods: Variants in the *CD59* coding sequence (exons 4, 5 and 6) and the splice sites were systematically compiled from 4 major populations in 6 whole genome and whole exome databases. The populations represented were African, Caucasian, Asian, and Latin American. PredictSNP metaserver was used to determine the functional impact of non-synonymous single nucleotide variants (SNVs).

Results: Among 488,592 individuals analyzed, nucleotide variants were observed in only 6,491 individuals (1.33%). Most of the variants with >10 observations were synonymous and occurred in all 4 major populations analyzed. Among the non-synonymous variants, 9 had been deposited in ClinVar and 3 had been observed in patients and published. PredictSNP's deleterious classification of 5 out of 9 SNVs correlated well with the diseased clinical outcome in ClinVar. The most common computationally predicted deleterious variant was the non-synonymous p.Pro128Ser present in the 1 in 11,187 Caucasians and 1 in 19,685 Latin Americans, yet has not been reported in a patient.

Summary / Conclusions: We systematically collated genetic variants observed in multiethnic individuals from global sequencing datasets and applied a computational tool to investigate their effect and assist in their classification. The inability of PredictSNP in accurately classifying 4 out of 9 ClinVar SNVs as deleterious implies that current computational algorithms are not reliable enough to predict the pathogenic potential of a genetic variant. To improve clinical care, automated learning algorithms should be supplemented with reliable 3D protein structure and high quality genetic and clinical data to train predictive models. Comprehensive collation and frequency assessment of variants in global sequencing datasets in conjunction with computational algorithms can help identify and prioritize variants in genes susceptible to developing disease for *in vitro* functional studies. Our study is the first report compiling the *CD59* variants and contribute to improve our understanding of the frequency of *CD59* variants across the populations and facilitate the development of population-specific genomic medicine.

P556 | Full-length *RHD* and *RHCE* haplotypes by Nanopore sequencing of five overlapping, generic long-range PCR amplicons

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Background: Elucidating the allelic composition of the long and highly homologous *RHD* and *RHCE* genes on the haplotype level, which can be transfusion-relevant, is a challenging endeavor. So far, it required very laborious techniques like cloning the allele into a suitable vector or performing transcript (cDNA) analysis in reticulocytes. Thus, it is common to only deduce the most likely genetic allele from phenotype data. For instance, in a case of weak RhC and normal Rhc antigen expression, an identified heterozygous non-synonymous variant would be assigned to the *RHCE**C allele. Newest long-read sequencing methodology finally facilitates *RHD* and *RHCE* haplotype reconstruction by single-molecule sequencing of long-range PCR amplicons.

Aims: Here, we developed a long-range PCR approach using generic primers, designed to co-amplify the homologous *RHD* and *RHCE* genes. The use of generic primers reduces the risk of unnoticed allelic dropout in case of *RHD/CE* hybrid alleles.

Methods: To fully cover exons 1 to 10 from *RHD* (~57 kb) and *RHCE* (~59 kb), we designed five generic primer pairs to co-amplify both genes. PCR amplicon lengths ranged from 12.3 to 15.2 kb for *RHD*, and 13.3 to 15.2 kb for *RHCE*. Overlaps of PCR amplicons comprised at least 1.1 kb and were chosen, whenever possible, to lie within polymorphic regions as haplotype reconstruction requires heterozygous variants. The five PCR products per sample were equimolarly pooled and barcoded to allow Oxford Nanopore sequencing of multiple samples on the same flow cell. Bioinformatic analysis involved reference-based variant calling and subsequent phasing using an extracted 165-kb HG38 reference sequence of the entire *RHD* and *RHCE* gene locus. We tested the method with two donors known to carry heterozygous *RHCE* C/c, E/e and c.733C>G variants, the latter causative for V+ and VS+ antigen formation. Phenotypically, both donors showed DCcEe, but the C antigen was weak in one of them.

Results: Both samples showed good distributions of read depths along both genes for all PCR-amplicons (least covered amplicons with 250× and 150×, respectively). Allelic distribution patterns of variants clearly showed that the generic PCR amplicons of both genes were sufficiently different to unequivocally map to the respective gene reference sequence. Genetic phasing worked well, which not only allowed the re-construction of full-length haplotypes for both genes, but also elicited converse results regarding the *RHCE* allele backgrounds of the c.733C>G variant in both donors. While the donor with the serological weak C showed a classical ce^s allele (*RHCE**01.20.01 | *RHCE**04), the other donor showed the V and VS-causing variant on a *RHCE**C

background (*RHCE*02.30* | *RHCE*03*). The complete sequence information for *RHD* allowed to rule out alternative complex alleles like (C) *ce^s*. Based on these results, we speculate that the observed weak C in the *ce^s*/CE sample is most likely a consequence of the R(z)-haplotype where DCE all lie on the same allele.

Summary / Conclusions: Here, we present an amplicon-based Nanopore sequencing method suitable to gain full-length *RHD* and *RHCE* haplotypes. By relying on generic PCR primers, the method not only reduces the number and complexity of PCRs but also the risk of unnoticed allelic dropout in case of hybrid alleles, which are prevalent in the Rh blood group system.

P557 | Characterization of a novel *RHCE*CeCW* variant allele

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Background: The RH blood group system contains more than 50 blood group antigens with some of them being considered highly immunogenic. Hence, a correct assignment of the RH status of blood donors and patients is crucial. However, weakened expression of antigens can challenge serological investigations and make correct classifications difficult. A detailed characterization of variant alleles at the molecular level can help to clarify discrepant serological results.

Aims: Samples of three first-time donors were investigated to resolve discrepant RH phenotyping results.

Methods: Blood group phenotyping was performed using standard serological column agglutination testing (DG Gel Double Pheno, DG Gel ABO/Rh, DG Gel Rh Pheno+Kell, Diagnostic Grifols, S.A.; DiaClon Rh-Subgroups + Cw + K, DiaClon Rh-Subgroups + K, Bio-Rad). Genotyping required semi-automated DNA extraction on a Qiagen EZ1 Advanced platform and manual total RNA isolation using the NucleoSpin RNA Blood kit (Macherey-Nagel, Germany). Commercially available SSP-PCR kits were used to detect common *RHCE* variants (RBC-CDE; RBC-Ready Gene, *RHCE* variants, Inno-Train, Germany). Genomic DNA and reverse-transcribed cDNA were amplified and sequenced using published and in-house designed primers.

Results: All three donor samples were initially tested by column agglutination and showed a positive RH2 phenotype with clone Anti-C MS-24. In contrast, the reaction with clone Anti-C P3 × 25513G8 was negative for all samples (Grifols). Further investigations revealed a weakened RH2 antigen expression demonstrated by column agglutination with clones Anti-C MS-273 and P3 × 25513G8 (BioRad). In addition, all three donors were RH8 (*C^W*) positive resulting in the common phenotype RH:1,P2,-3,4,5,8. Commercially available PCR-kits did not detect any conclusive *RHCE* variant. However, sequencing of genomic DNA revealed a heterozygous missense mutation c.1139C>T in exon 8. This results in the amino acid substitution p.-Ser380Phe located in the twelfth transmembrane domain of the mature glycoprotein according to a recently published model (Floch *et al.*, Transfusion, 2021). Allele-specific amplification and sequencing

of *RHCE*02* cDNA demonstrated that c.1139T is located on the *RHCE*02.08.01* allele encoding RH8. Furthermore, a significant lower abundance of *RHCE*02.08.01* cDNA compared to the cDNA of the additional allele *RHCE*01* was detected. The new allele *RHCE*02.08.01.1139T* (accession number OQ884365), to the best of our knowledge, has not been reported previously.

Summary / Conclusions: Here we report the allele *RHCE*02.08.01.1139T* causing an amino acid substitution within the last transmembrane domain of RH2. A significantly lower expression of RH2 in the three donors was observed compared to samples with unchanged *RHCE*02.08.01*. A likely explanation is the amino acid change p.Ser380Phe. In addition, a decreased level of mRNA of the mutant allele was observed in comparison to *RHCE*01* mRNA. Interestingly, all three donors harbouring the novel allele reside in the same specific region of Switzerland and it is likely that more donors with this phenotype will be identified there. Thus, a simple SSP-PCR approach was developed for a rapid analysis of suspect cases.

P558 | The universal blood donor typing array for large-scale genotyping of red cell, platelet and leukocyte antigens shows high concordance with test-of-record in an international multi-ethnic donor cohort

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Background: Matching for blood cell antigens between donor and recipient is important to prevent alloimmunisation and haemolytic transfusion reactions. To facilitate extended matching, the Blood transfusion Genomics Consortium (BGC) has developed the Axiom Total Blood Typing Solution to simultaneously genotype human erythrocyte (HEA), platelet (HPA) and leukocyte (HLA) antigens.

Aims: To determine assay accuracy and reproducibility in a multi-ethnic cohort of two Axiom genotyping arrays and the accompanying integrated analysis package (IAP) designed for blood donor typing in a real-world setting of three blood service laboratories.

Methods: DNA samples were collected from blood services from Australia, Canada, England, Finland, the Netherlands, U.S. and South Africa. Identical sets of 6946 samples were genotyped using the GeneTitan-MC instrument (Thermo Fisher) at two blood centers with a 20,000 probe Universal Blood Donor Typing array

(UBDT_PC1). 3,938 samples were typed with the UK Biobankv2.2 array which includes the UBDT_PC1 transfusion focused probes. The array genotypes were analysed using IAP-v2.0. Ancestry was inferred from the data to determine performance within the major ethnic groups. HEA, HPA, and HLA class I/II types were inferred by the IAP bloodTyper and HLA*IMP:02 modules and results were compared to the donor test-of-record. Resolution of sample discordances was performed by SNP-based tests and/or gene sequencing.

Results: Inferred phenotypes of 6,775 samples passing the IAP quality control were compared to donor record types. For HEA, concordance levels of 99.89%, 99.90% and 99.88% across 124,030, 124,629 and 78,092 phenotypes were observed between three test sites. For HPA, concordance was 99.64%, 99.57% and 99.65% across 1389, 1417 and 1433 comparisons although HPA-3 was not able to be determined. For 767 samples with previous HLA results, concordance was 99.7%, 98.7% and 99.7% for HLA class I A, B and C and 96.9%, 99.9%, 98.9% for class II DPB1, DQB1 and DRB1. Lower concordance for DPB1 was due to an outdated reference table. Reproducibility of genotype calls for 53 HEA antigens across 369,181 comparisons in a unified dataset consisting of 6,672 of the samples was 99.93%. Comparison of the 53 HEA and 8 HPA types with donor records resulted in 181 discordances in 165 samples with 44 in RH, 32 in MNS, 29 in DO, 25 in FY, 21 in JK, 10 in LU, 8 in CO, 4 in KEL, 2 in LW and 6 in HPA-1,2,5 and 15. Most (54%) were incorrect donor record types and 38% due to incorrect array interpretations requiring reprogramming.

Summary / Conclusions: We present data that the transfusion genotype module incorporated in UBDT_PC1 and UKBB_v2.2 AxiomTM arrays can produce highly accurate HEA, HPA and HLA genotypes simultaneously and at scale. Both arrays have been trialled using an ancestrally diverse panel of DNA samples, with 34.8% (2,417/6,946) samples from donors of non-European ancestry determined by genotype. A high level of reproducibility for both the genotype calls and inferred phenotypes between the typing labs for 6,672 repeated samples has been shown with 99.93% and 99.98% concordance, respectively. Importantly, all genotyping in this study has been conducted in accredited blood service molecular laboratories, demonstrating that this technology can be implemented for routine donor typing. The development and validation by BGC of a comprehensive genotyping array to densely type donors of the main ancestry groups for HEA, HPA and HLA will support blood services to issue better matched blood.

P559 | Correlation Between RHD variants, Duffy phenotypes and FY*B^{ES} in mixed population

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Background: The variability of alleles encoding variant antigens in RH is wide and highly important to transfusion medicine. The types of variant RHD vary can be gathered in clusters that are influenced by

the individual ethnic ancestry. Relying on self-declared race or skin color is not accurate in highly mixed populations. FY*B^{ES} is an African ancestry marker that can be easily assessed by the Duffy phenotype followed by DARC genotyping. Our hypothesis is that FY*B^{ES} can be used by blood banks to select donor population that needs to be tested to identify RH variants in case of variant-compatible transfusion as well as to screen for RH variants in case of patients presenting with serological weak D.

Aims: To associate Duffy phenotypes, FY*B^{ES} and RHD variant alleles in a racially mixed population

Methods: All donors were phenotyped for RhD and RhCE by microplate hemagglutination (NEO Immucor, Norcross - GA, USA) and confirmation of weak D by Solid phase Capture-R select (NEO Immucor, Norcross - GA, USA). Fy(a) and Fy(b) phenotyping was performed by IAT-gel test (Grifols, Barcelona). Genotyping was performed using PCR-SSP, Multiplex-PCR, PCR-RFLP, and Sanger Sequencing (RHD). Samples displaying the Fy(a-b-) or Fy(a+b-) phenotypes were genotyped by PCR-RT for the c.-67T>C (FY*B^{ES} or FY*02N.01), which alters the FY*B promoter gene, resulting in non-expression of the Fy^b protein solely on red blood cells.

Results: 311,998 blood donor samples were included. Of these, 295 were identified as serological weak D and 486 as D-negative (serological) with positive C and/or E antigens. 26 of the selected RhD- samples exhibited RHD. Among the 295 D-weak donors, some differential associations between Duffy phenotype and RHD variants were detected: 1-Fy(a-b-) or Duffy null phenotype (n = 29/295) was mainly linked to weak D type 2 (n = 11/29; 38%), weak partial 11 (n = 4/29; 14%), and RHD*DAR (n = 2/29; 7%). Even though this phenotype is typically associated with African ancestry, in this mixed population Caucasian variants were prevalent; 2- Fy(a-b+) individuals (n = 95/295) had higher frequencies of weak D type 2 (n = 47/95, 49%), followed by weak D type 1 (n = 13/95, 14%); and weak D type 38 (n = 12/95, 13%); 3- Fy(a+b-) individuals (n = 76/295) presented diverse types of weak D and weak D type 2 was the most common (n = 22/76, 29%); 4- Fy(a+b+) individuals (n = 95/295) also mostly associated with weak D type 2 (n = 36/95, 38%). Fy(b-) samples were tested for FY*B^{ES} (n = 104/295). 25% of the Fy(b-) samples were FY*B^{ES}/ FY*B^{ES} and presented the following RhD variants: weak D type 2 (n = 12/26; 46%), weak partial 11 (n = 2/26; 8%) and, interestingly, only one sample was RHD*DAR (n = 1/26; 4%). 34% of the Fy(b-) samples were FY*B^{ES}/ FY*A with the following RhD variants: weak D type 2 (n = 8/35; 23%), RHD*DAR and RHD*DAU (n = 8/35; 23%), followed by other weak D variants. 38% of the Fy(b-) samples were FY*A/ FY*A and was mostly associated with weak D type 2 (n = 13/39; 33%).

Summary / Conclusions: HD variants underlying weak-D typically associated with Caucasian population can be found with considerable prevalence among individuals presenting with the ancestry marker FY*B^{ES} in highly mixed populations. Individuals phenotyping as Fy(b+) and with serological weak-D exhibited only Caucasian-associated altered RHD. However, among Fy(b-) individuals, Caucasian and African-associated altered RHD were identified.

P560 | Haplotypes of the CR1 gene coding for the Knops blood group system characterized by long read sequencingP Bugert¹, J Schneider¹, C Lang¹, G Rink¹, H Klüter¹¹Institute of Transfusion Medicine and Immunology, Heidelberg University, Medical Faculty Mannheim, German Red Cross Blood Service Baden-Württemberg - Hessen, Mannheim, Germany

Background: The antigens of the Knops blood group system (KN; ISBT 022) are located on the complement receptor 1 (CR1), a single pass membrane protein with four Long-Homologous-Repeats (LHR-A, -B, -C, -D). All KN antigens identified so far are located in LHR-C and LHR-D. Antibodies to KN antigens are usually not clinically significant but they are common in patients. Massively parallel sequencing (MPS) of all 39 exons of the CR1 gene in patients with suspected alloantibodies to KN antigens revealed several CR1 variants of unknown significance (VUS). The variants were located in different CR1 exons and the phasing of the haplotypes remained unknown.

Aims: The identification of haplotypes adds more to sequencing data in order to characterize the molecular basis of blood group antigens. However, MPS by using short read sequencing technologies is not suitable to phase haplotypes of distant variants. We established long read Oxford Nanopore Technologies (ONT) sequencing for the LHR-C and -D region of CR1 for haplotype phasing.

Methods: The CR1 LHR-C and -D region was amplified by long range PCR with 5 overlapping PCR products from genomic DNA samples of 10 patients with at least 2 distant VUS. ONT sequencing was performed on MinION with R10.4.1 flow cells (Oxford Nanopore Technologies, Oxford, UK). After base calling, the Chopper software program was used to filter the data for a minimum quality score of 9. Filtered FASTQ files were aligned to the chromosome 1 sequence of the human reference genome GRCh38.p14 by using the Geneious Prime software (Version 2023.2.1; Biomatters Inc., Boston, MA, USA).

Results: PCR amplification of the LHR-C and -D region was achieved from all samples with amplicon sizes of 10,601 to 12,797 bps. Long range sequencing of the large amplicons confirmed the variants in the exons previously identified by MPS. Further variants were identified in the overlapping amplicon regions and were used for haplotype phasing of distant exon variants. In 6 samples we were able to identify the CR1 haplotypes including exon and intron variants with a distance of up to 41,699 bps. E.g., haplotype 1: c.3623A (exon 22, p.1208His, KN11) – c.4843A (exon 29, p.1615Ile, KN9) – c.5480C (exon 33, VUS); haplotype 2: c.3623G (exon 22, p.1208Arg, KN12) – c.4843G (exon 29, p.1615Val, KN10) – c.5480G (exon 33, VUS).

Summary / Conclusions: Long read ONT sequencing of large PCR products is suitable for haplotype phasing of the CR1 gene. The knowledge of the haplotypes could be useful especially in cases with alloantibodies against an unknown KN antigen and two or more VUS in CR1. We were able to allocate 4 VUS to haplotypes of the LHR-C and -D region.

P561 | Genomic exome sequencing to resolve complex blood group problemsG Lopez^{1,2}, G Millard³, E Roulis¹, M Sarri¹, Y Liew³, T Powley³, J Daly⁴, R Flower^{1,5}, C Hyland^{1,5}¹Research and Development, Australian Red Cross Lifeblood, Brisbane,²School of Health, University of the Sunshine Coast, Sippy Downs, ³Red Cell Reference Laboratory, ⁴Pathology Services, Australian Red Cross Lifeblood, ⁵School of Biomedical Sciences, Queensland University of Technology, Brisbane, Australia

Background: The emergence of blood group (BG) genomics has aided the discovery of more BG systems and revealed new antigens within existing systems. Currently 45 systems comprising 362 antigens are registered on the ISBT website in tables which are maintained by the ISBT Working Party for Red Cell Immunogenetics and Blood Group Terminology. There is also a genetic diversity overlaying this antigenic diversity with about 1800 alleles registered on these tables. We previously reported on a targeted exome sequencing panel designed to predict comprehensive BG, platelet and neutrophil profiles in a single test (Roulis *et al*, Transfusion 2020). This system subsequently contributed to defining 5 novel red cell antigens by immunohematology investigations of either antenatal or transfused patients. Targeted BG exome sequencing also has the potential to resolve samples presenting with equivocal serology and/or anomalous genotyping calls by the accredited single nucleotide polymorphism (SNP)-array systems.

Aims: To report on the utility for targeted exome sequencing to resolve n = 451 samples that remained unresolved by serology and/or SNP-array typing.

Methods: Blood group serology was performed by standard accredited methods; genotyping was performed using the BioArray™ molecular BeadChip™ assays (Immucor). The sequencing panel covered 38 BG systems as well as transcription factors GATA1 and KLF1. Massively parallel sequencing (MPS) was performed using an Illumina DNA prep enrichment workflow and sequenced on the Illumina MiSeq. Variant call files were generated using CLC Genomics Workbench (QIAGEN), from BAM files aligned to the GrCh37/Hg19 human reference genome. Variants were compared to the ISBT BG allele database to provide a predicted phenotype call.

Results: Among 451 samples presented for MPS, 265 (58.8%) had unresolved Rh typing results: 191/265 (72.1%) comprised a range of over 70 diverse alleles listed by ISBT and associated with weak D, Del or negative phenotypes; 41/265 (15.5%) carried no variant sequences; 27/265 (10.2%) carried novel or rare single nucleotide variants (SNVs) not previously listed on the ISBT database; 6/265 (2.3%) represented possible chimeras explaining mixed field reactions. The remaining 186/451 (41.2%) samples were referred for investigations across 23 BG systems. Among 40 samples sent for ABO investigation, 31 were known ISBT allelic variants; 5 had a potential novel or rare allele and 4 were either a

chimera or unresolved. The JK, MNS, KEL, FY, LU and GE systems comprised the next largest group of allelic variants, noting that among 8 samples with GE requests, 3 involved exon changes. One sample referred for XK investigation carried the rare ISBT XK*N39 allele. Finally, among 12 samples referred for investigation in the LU system, 1 had a potentially novel KLF1 SNV.

Summary / Conclusions: Targeted BG exome sequencing is effective in resolving aberrant serology and/or genotyping results. The ISBT BG allele data base is a critical resource in predicting the phenotype from the MPS variant calls. Even so, the allelic diversity present in the diverse Australian population is not yet completely captured and a proportion of samples remain unresolved and require further molecular studies. As this progresses, it will be important to register findings on public databases to further support the integration of genomics into red cell reference investigations and to support the transfusion community.

P562 | Mixed erythrocyte phenotypes - an indirect sign of clonal hematopoiesis?

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Background: Mixed-field agglutination (MFA) is common in transfused patients and bone marrow transplant recipients. MFA can also be detected in patients with no immediately obvious explanation.

Aims: This retrospective analysis examines such cases referred to a RBC Genomics Reference Laboratory over a period of 13 years (2009-2022).

Methods: We included all patients presenting MFA or modified phenotypes for ABO, RH or other RBC antigens, with no history of transfusion or allograft and no underlying causal variant. We collected patient age, the existence of a hematology/oncology disease or a twin, transfusion history and the results of serological and molecular typing (real-time PCR, Sanger sequencing). A literature review was conducted to identify possible mechanisms and assess clinical and transfusion implications.

Results: The literature reports 2 genetic mechanisms. Chimerism is the presence of cells from different individuals in the same organism, including permanent twin chimerism, transitory microchimerism during pregnancy and iatrogenic chimerism (e.g post-transplant) while blood cell mosaicism is more frequent and refers to the development of a cellular clone following genetic alterations, notably loss of heterozygosity (LOH), usually in myeloid hemopathies. The significance of the fortuitously discovered LOH in asymptomatic patients is unknown. Only recently has the extent of somatic variation in normal

tissues become apparent. Despite its normal occurrence in aging individuals, current evidence suggests that clonal hematopoiesis of indeterminate potential (CHIP) constitutes a risk factor for cardiovascular disease, and potentially for hematologic progression. Somatic mutations in blood cells could affect nearly 1% of subjects under 50 years. 76 cases were identified: 33 men and 43 women (25 women < 50 years including 10 who were pregnant). 70 were adults (median of 64 years) and 6, children. The discrepancy could be explained for 40/76 patients: 6 twins and 34 patients with a history of onco/hematologic malignancy, leaving 36/76 patients with no apparent cause. 69/76 cases involved the RH system, 4 the ABO system, 3 the KEL system and 1 the JK system. In the RH system, we mainly observed MFA, but 7/69 cases were referred for a weak expression, an acquisition or a loss of RH antigen. Most cases of this series involved the RH system probably because ABO discrepancies are usually managed with group O blood and not further investigated in our settings. Analysis of our data suggests 9 cases of chimerism with anomalies in several blood group systems (including 6/9 hematopoietic inter-twin chimeras) and 63 cases of mosaicism (59/63 suspected LOH, and 4/63 cases of complex chromosomal anomalies). Among suspected LOH cases, 41/59 led to partial or complete loss of the R1 (DcE) haplotype, 7/59 of the R2 (DcE) haplotype, and 5/59 loss of RHD. None of the patients formed alloantibodies to the altered antigen(s), despite transfusions in 22/76 patients.

Summary / Conclusions: Although cases of secondary changes in blood groups have been reported in hematologic neoplasms, such anomalies in healthy subjects have only recently been reported. This series, the largest described to our knowledge, suggests that these anomalies are more common than expected and may be an indirect sign of clonal hematopoiesis, including in healthy patients. Prospective studies are needed to assess the risk of eventually developing an underlying hematologic disease.

P563 | Abstract withdrawn

P564 | RHD genotyping—a six years' experience in Mestre Blood Bank

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Background: A considerable number of RHD alleles responsible for weak and partial D phenotypes have been identified. Serologic determination of these phenotypes is often doubtful and makes genetic analysis of RHD gene highly desirable in transfusion recipients and pregnant women.

Aims: We report the experience of Mestre Blood Bank in analysis of the RHD gene in six years from 2018 to 2023.

Methods: Subjects for RHD gene analysis were selected for presence of a serological weak D phenotype, defined as reactivity of RBCs with an anti-D reagent giving no or weak ($\leq 2+$) score in initial testing but agglutinating moderately or strongly with antihuman globulin. These

P564 - Table 1: D weak and D variant observed in Mestres Blood Bank from 2018 to 2023

D Weak	Number (%)	D Partial	Number (%)
D weak type 1	227 (40.1%)	D partial III	3 (0.5%)
D weak type 2	46 (8.3%)	D partial IV	1 (0.2%)
D weak type 3	9 (1.6%)	D partial V	4 (0.7%)
D weak type 4	4 (0.7%)	D partial VI	27 (4.9%)
D weak type 11	47 (8.5%)	DAR	1 (0.2%)
D weak type 15	34 (6.1%)	DFR2	3 (0.5%)
D weak type 18	4 (0.7%)	DAU4	1 (0.2%)
		DNB	2 (0.4%)

samples were selected for genotyping using the microarray-based method “Bead-Chip” supplied by Immucor.

Results: From 2018 to 2023, we selected, for RHD gene analysis, 555 subject with D weak phenotype; 86 subjects (15.5%) were D positive and 56 (10.1%) were D negative, without variant, in 413 subjects a D weak or a D variant was observed. See table I for further description.

Summary / Conclusions: Many serological weak D phenotypes are associated RHD gene mutations leading to one or more amino acids substitutions in the RhD protein predicted to be within or below the RBC membrane causing decreased antigen expression on the red cell surface. Prevalence of serological weak D phenotypes varies by race and ethnicity. Serological weak D phenotypes are the most common D variants detected in Caucasians (0.2%-1.0%), the majority, as in our series, are associated with weak D type 1, 2 or 3. Our data confirmed a high prevalence of weak D type E and type 2, but we observed a high prevalence of type 11 and 15 and of the uncommon type 18 too. Studies indicate that transfusion recipients with weak D type 1, 2 or 3, in the homozygous or hemizygous state, are not at risk for forming allo anti-D when exposed to conventional RhD-positive blood units. Partial D genotype are associated with amino acid substitutions in the RhD protein on the RBC surface and lack D epitopes. The most common partial D phenotypes in Europe are DNB, DVI, and DVII. Our data confirmed a high prevalence of D partial type VI. Studies indicate that transfusion recipients D partial are at risk for forming alloanti-D when exposed to conventional RhD-positive blood units. As matter of fact the subject with D Partial DAR, a pregnant woman, developed and anti-D.

P565 | Navigating the complexity—assessing the risk of immunization for patients with RHCE variants

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Background: The presence of alloantibody poses a challenge for the support of patients needing transfusions or in prenatal care. This can be problematic for patients carrying genetics variants altering the expression of high frequency antigens or homozygote for a specific

allele, as it can increase the risk of hemolytic reaction or hemolytic disease of the fetus and newborn. Many patients from the Black communities, including patients with SCD, carry *RHCE* alleles affecting the expression of the e (RH5) and the high-frequency antigen hr^B (RH31). The most common alleles are from the ceVS subcategory (*RHCE*01.20...*), which are characterized by the nucleotide change c.733C>G that encodes for the VS antigen, a partial e and c (RH3) antigen, and a very weak or negative expression of hr^B. However, the risk of alloimmunisation of individuals with *RHCE* variants and the clinical significance of the resulting alloantibody is still debated.

Aims: This study aims to evaluate the risk of alloimmunisation in patients with an altered expression of the e and hr^B antigens.

Methods: A cohort of 51 patients referred to our provincial Immunohematology Reference Laboratory (IRL) from 2016 to 2021 with a C-E- partial e partial c and weak or negative hr^B phenotype was selected. All patients had an *RHCE* genotype composed of the *RHCE*01.20.01*, .02 and/or .03 allele. Clinical and serological data as well as the transfusion history in the province of Quebec (Canada) were gathered.

Results: Patients were divided into three categories according to the reason of the referral: 20 were followed for prenatal care, 15 had a sickle cell disease (SCD) diagnosis and 16 were referred for other or unspecified reasons. No anti-c, anti-e or anti-hr^B was found in prenatal care patients, regardless of their genotype. We found that 80% (12/15) of the patients with SCD received at least one transfusion which is a higher transfusion rate than the 40% (7/16) observed in the last category. No anti-c was found in patients with a transfusion history in our province. An anti-c was identified in one patient without recorded transfusion. We found that 50% (6/12) of transfused patients with SCD developed at anti-e. In contrast, no anti-e was detected in patients from the prenatal and the other category. Three anti-hr^B were identified in our cohort: 1 in a hr^B+^{vw}/− patient with SCD and 2 in hr^B− patients with SCD. However, 58% (7/12) of transfused patients with SCD also had a warm autoantibody of undetermined specificity, among them 4 patients that also had an anti-e.

Summary / Conclusions: We found that the risk of developing an anti-e or an anti-hr^B in patients with an altered expression of the e and hr^B antigen is low for individuals in prenatal care or with a diagnosis other than SCD. The risk is higher in patients with SCD, as over 50% of transfused patients developed an anti-e or anti-hr^B. However, the presence of autoantibodies limits our ability to determine the clinical significance of these antibodies in some patients, as recent transfusion or lack of material limits the characterization of the autoantibodies. For those patients, a case-by-case approach and an individual assessment of the risk of alloimmunisation are needed.

P566 | Molecular epidemiology of the *RHD* gene in serological D-negative individuals in Indonesia—from basic knowledge to clinical applications

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Background: Genetic polymorphism of the RH blood group system has been extensively reported in Europe, America, and East Asia, but remains to be described in details in many other regions/countries in the world, as it is known to be highly dependent on ethnicity. Southeast Asia, which accounts for 8% of the total world population, has been poorly explored so far. D-negative (D-) phenotype is typically rare in Southeast Asia (<1% in the general population) and supply of these D- red blood cells (RBCs) is a recurrent challenge in the region. Apparent serological D- RBCs may carry actually D antigen, and fundamental knowledge of the molecular basis resulting in this phenotype may help for optimizing diagnostics and selection of these RBCs.

Aims: In this work, we sought to investigate the molecular basis of serological D- phenotype in a subset of individuals from Indonesia, the most populous country in Southeast Asia with >270 million inhabitants.

Methods: Blood samples from Indonesian individuals originating from Medan (Sumatra Island), Yogyakarta and Surabaya (Java Island) were typed by routine serological techniques, and 238 apparent D- samples were selected for further investigation. After informed consent, genomic DNA was extracted with a commercial kit. The *RHD* gene was analyzed by complementary techniques to identify structural variants and single nucleotide variants, i.e. a quantitative multiplex PCR of short fluorescent fragments (QMP5F) and direct sequencing, respectively. Allele frequency was recorded and reported here.

Results: A total of 18 variant *RHD* alleles with various mutational mechanisms was found. As expected in such a situation, the whole deletion of the *RHD* gene (i.e. *RHD**01N.01) was by far the most common variant allele (n = 386; allele frequency: 0.8109), followed by the Asian type DEL allele (43; 0.0903), and the *RHD**01N.03 and *RHD**01EL.44 hybrid alleles (10 alleles each). All other alleles were found four times or less. Interestingly, three novel alleles were identified at the hemizygous state in six samples: 1/ a complex genomic structure, named *RHD**01N (ex8del), involves a deletion of a >2 kilobase-region, including full exon 8, which is very partially replaced by a short homologous intronic region of *RHCE*; 2/ a splice site variant, as shown by functional analysis, defining the *RHD**01(c.635-1A) allele; and 3/ a frameshift variant characterizing the *RHD**01(c.1220del) allele. It is also worth mentioning that two alleles showed no evidence of variant(s) in their sequence.

Summary / Conclusions: For the first time, this study provides molecular data about the genetic polymorphism of the *RHD* gene in a country that remained unexplored before. Beyond the fundamental findings,

we showed interestingly that 16.4% serological D- individuals actually carry the Asian type DEL allele, and are thus thought to be of the DEL phenotype. In accordance with recent recommendations of the literature, we propose that these Asian type DEL patients should be considered and treated as conventional D+ patients. Also, implementing a systematic test (e.g. molecular assay) at the national level for identifying those patients and the donors presenting with an apparent D- phenotype for a proper selection and supply of true D- RBCs is to be achieved. Finally, it will be important in a near future to reinforce the cooperation between countries that share common issues at the intraregional level, such as Thailand that shows a comparable pattern.

P567 | Software update enables improved *RHCE*/*RHD* genotyping on allele level for real-time PCR system

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Background: Individuals from African descent often carry variant *RHCE* and *RHD* alleles. To handle the high number of SNPs investigated for a detailed genotyping including variants in a routine setting a sophisticated software-based solution is indispensable. With this, the alloimmunization risk may be lowered, as the absence of high prevalence antigens in patients and the presence of low prevalence antigens in donors could be predicted with more accuracy.

Aims: The RBC-FluoGene CDE eXtend kit is a real-time PCR assay based on the TaqMan probe technique. It comprises 43 *RHD* and 30 *RHCE* SNPs. The analysis with the FluoGene Software (FGSW) update version 1.8 has been improved for calling a heterozygous *RHD* result, calling a genotype for *RHCE* on an allele level in ISBT nomenclature and the prediction of the respective phenotypes.

Methods: 16 DNA samples from African descent were genotyped with RBC-FluoGene CDE eXtend kit (lot R920823) and the result displays of FGSW versions 1.7 vs. 1.8 were compared.

Results: The FGSW version 1.8 calls for all samples *RHD* and *RHCE* genotyping results in numerical ISBT nomenclature as listed in table 1 (*RHD*) and table 2 (*RHCE*). In the previous version 1.7 only the most likely heterozygous combinations of two variant alleles (e.g. Dpsi/DAU as shown in sample no. 10) were displayed by the analysis software, but others gave no call (samples no. 2, 6, 7, 8, 12, 13, 15, 16; table 1). In version 1.8, the software calculates all possible heterozygous combinations if the Dd filter option is deactivated. For results comprising a *RHD**01 allele and a variant allele (samples no. 4, 5, 6, 8, 9, 13; table 1), FGSW version 1.8 also

P567 - Table 1 (shortened)

Sample	RHD result display in FGSW version 1.7#	RHD result display in FGSW version 1.8*§
No. 2	RHD Allele no call	<i>RHD</i> *04.01/10.05
No. 3	RHD Allele 08N.01 (PseudogenePsi)(Dpsi)	<i>RHD</i> *01N.01/08N.01
No. 7	RHD Allele no call	<i>RHD</i> *03N.01/10.04
No. 16	RHD Allele no call	<i>RHD</i> *01N.06/10.05

P567 - Table 2 (shortened)

Sample	RHCE result display in FGSW version 1.7	RHCE result display in FGSW version 1.8 #
No. 2	RHCE 1006G, 1025C, 1025C>T, 340C, 48G>C, 577A, 667G, 697C, 712A, 733C, c, e	RHCE*01.01/01.02.01
No. 3	RHCE 1006G, 1025C, 254C>G, 340C, 48G, 48G>C, 577A, 667G, 697C, 712A, c, e	RHCE*01.01/01.06.01
No. 7	RHCE 1006G, 1006G>T, 1025C, 340C, 48G>C, 577A, 667G, 697C, 712A, 733C, 733C>G, c, e	RHCE*01.01/01.20.03
No. 16	RHCE 1006G, 1025C, 340C, 48G, 48G>C, 577A, 667G, 697C, 712A, 733C, 733C>G, c, e	RHCE*01.01/01.20.02.01 RHCE*01.01/01.20.01

displays the possible combinations of two variant alleles. These combinations are very unlikely but cannot be excluded.

Table 2 shows the display of each positive specificity (C, C^w, c, E, e, plus SNPs) in FGSW 1.7 compared to the RHCE genotyping result in numerical ISBT nomenclature in FGSW 1.8. For 15 out of 16 samples the version 1.8 calls a clear RHCE genotyping result. For sample no. 16 (table 2) the analysis software gives two possible combinations depending on the allelic location of the SNP 48G>C either on the RHCE*01.01 or RHCE*01.20.02.01 allele. For both combinations the predicted phenotype 1 is identical (RH:-2,-3,4,5,10,19,20,3), but a difference is seen in the predicted phenotype 2, which could be a regular e+ in case of a RHCE*01 allele or a e+(partial) in case of a RHCE*01.01 allele.

Summary / Conclusions: Complex blood group systems require investigation of an increasing number of SNPs for a better characterization of variant alleles. To handle RHD and RHCE genotyping in a routine setting, a sophisticated software solution is essential.

P568 | Diversity of RHD underlying serological weak-D phenotype in Brazilian admixed population and its implications on transfusion protocols

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Background: RH complexity in transfusion medicine varies across populations. Caucasians typically exhibit weak-D phenotypes 1, 2, or 3, with low alloimmunization risk. However, in African populations, weak D type 4 cluster dominates. Brazil's diverse population—Caucasian, African, and Indian—poses challenges in defining protocols for D immunoprophylaxis and transfusions, particularly for women and children, due to varying ancestral predominance by region.

Aims: To describe the RHD variants underlying serological weak D or D-negative (with either C+ or E+) phenotypes in blood donors of four regions of Brazil with different ethnic background.

Methods: A total of 311,998 blood donor samples were collected at Imunolab (São Paulo, Brazil) stemming from the following Brazilian regions: Southeast, Northeast, Midwest and South over the course of one year. The South part of Brazil is typically described as Caucasian, and the Southeast and Northeast as mixed (African and Caucasian). All donors were RhD typed by microplate hemmagglutination (NEO Immucor, Norcross - GA, USA) and confirmation of weak D was performed with solid phase (Capture-R select, NEO Immucor, Norcross - GA, USA). RHD genotyping was conducted using PCR-SSP, Multiplex-PCR, PCR-RFLP, and Sanger Sequencing.

Results: 295 donors were classified as serological weak D and included in the study. Among these samples, 88% exhibited at least one altered RHD. Both weak-partial D and weak D RHD were found at the following distribution: 19% of the samples showed RHD alleles encoding partial-weak RhD phenotypes of weak D type 4 cluster (RHD*DAU4, RHD*DAR2.1 and RHD*DAR1.2); 67% were categorized as weak D type 1, 2, or 3 variants, and 14% comprehended other types of RHD (38, 11 and 15). The RHD*DAR (weak and partial D) variant was more prevalent in the Southeast (45%) and Midwest (36%) regions in comparison to the others, while the RHD*DAU4 (weak and partial D) allele had higher prevalence in the Northeast (39%) and Southeast (28%) regions. The fact that the Northeast and Southeast regions of Brazil had different slavery routes may justify these differences. The weak D type 38 and weak partial 11 were more prevalent in the South (18 and 12%, respectively) and Southeast (12 and 9%, respectively). The distribution of weak D type 1, 2 and 3 was: Southeast (35%), Midwest (31%), South (17%) and Northeast (17%). A total of 486 donors included in the study were D-negative (serological) with C and/or E antigens. Of these, 26 presented RHD: 19% genotyped as RHD*08N.01/RHD*01N.01 and 31% genotyped as RHD*03N.01/RHD*08N.01. Among the RHD*03N.01/RHD*08N.01 samples, 25% were from the Southeast, 50% from the Northeast, and 25% from the Midwest, with no samples from the South. Regarding RHD*08N.01/RHD*01N.01 samples, 60% were from the Southeast, 20% from the Northeast, and 20% from the South.

Summary / Conclusions: More than 30% of the RHD encoding serological weak D in Brazil are associated with partial D phenotype. Even though the distribution of weak-partial RHD varied between the Brazilian territory, all studied regions presented a significant proportion of partial D phenotype among the serological weak-D, emphasizing the need for RH genotyping in case of reduced RhD expression, especially in pregnancy to guide anti-D immunoprophylaxis.

P569 | Homozygosity for a novel A4GALT null allele resulting in p phenotype and production of anti-PP1Pk in an Indian blood donor

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Background: The A4GALT gene encodes the galactosyltransferase enzyme responsible for production of the red cell carbohydrate antigens P^k and P¹ (of the P1PK blood group system) from their

precursors. Homozygosity or compound heterozygosity for inactivating mutations in A4GALT results in the rare p phenotype, associated with production of anti-PP1P^k antibody. The ISBT currently list nearly 40 different A4GALT null alleles caused by deletions, missense and nonsense mutations. P₁ and P₂ phenotypes are determined by transcriptional regulation, with P₂ individuals being homozygous for a polymorphism in A4GALT intron 1 (rs5751348) which results in reduced A4GALT transcript levels and a lack of P1 antigen production. **Aims:** To present results from serological and molecular investigations of a case of anti-PP1P^k with a novel molecular background of the p phenotype, including a family study.

Methods: Samples from a 32-year-old Indian blood donor were investigated due to the presence of a pan-reactive alloantibody. Blood samples from the donor's sister and mother were also tested. Serological investigations were performed by standard tube LISS IAT and direct agglutination techniques. Genomic DNA was extracted and sequencing of the A4GALT gene was performed.

Results: The donor was found to have the p phenotype (P1–, P–, P^k–) and his plasma contained anti-PP1P^k, reacting with moderate strength by LISS IAT with untreated cells and stronger with papain treated cells. Moderate reactivity was also observed by direct agglutination at both 18°C and 37°C. Three examples of p cells were compatible with the donor's plasma and no additional antibodies were detected. Cells from the donor's ABO compatible sister were found to be incompatible with the donor's plasma. Sequencing of the A4GALT intron 1 polymorphism associated with P1/P2 expression revealed the donor to be homozygous for NM_017436.7:c.-188+3010G, associated with the A4GALT*01 allele (P1+P^k+). Sequencing of the coding region of A4GALT also revealed homozygosity for c.109A>G (p.Met37Val, characteristic of the A4GALT01.02 allele) and c.526G>A (p.Asp176Asn; rs371893172; freq. 0.0007). Two additional homozygous silent mutations were identified; c.903C>G (p.Pro301 =) and c.987G>A (p.Thr329 =). The rare c.526G>A mutation does not appear in the ISBT list of known A4GALT null alleles and thus appears to characterise a novel allele, carried on an A4GALT01.02 background in this case. The sister was found to be heterozygous for this novel allele (carried together with an A4GALT*02 allele), whilst surprisingly the mother did not carry the allele and had a homozygous A4GALT*02 (P2) genotype. Later enquiries revealed the mother was not in fact the biological mother of the donor, explaining the perceived discrepant sequencing results.

Summary / Conclusions: We have identified a novel null allele of A4GALT, c.[109A>G, 526G>A, 903C>G, 987G>A], found in an Indian blood donor carried in the homozygous state. The rare c.526G>A missense mutation appears to cause a lack of functional galactosyltransferase enzyme being encoded by the A4GALT allele, resulting in a p phenotype and production of anti-PP1P^k. This allele adds to the catalogue of known genetic backgrounds of this rare phenotype.

P570 | Variation in ABO intron/exon splice sites affect the mRNA forms using minigene technique in vitro

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Background: ABO incompatibility can cause severe hemolytic reactions and hemolytic diseases. In addition to the common normal A, B, O and AB phenotypes, there are some proportion and wide variety of ABO variants (subtypes) in the population. Elucidating the causes of their formation is helpful to accurately identify and regulate these variant antigens and improve the safety of blood transfusion.

Aims: Understanding the influence of variation in ABO intron/exon splice sites on the mRNA transcription splicing.

Methods: Genomic DNA of each variant individual was collected, and three discontinuous sequences of ABO gene were amplified, including exon 1, X, 7 and their flanking intron sequences, with × being exon adjacent to the variation site. Then the three purified discontinuous sequences were seamlessly cloned onto pcDNA3.1 (+) plasmid to construct the minigene reporter plasmid. The plasmids were transformed into Trans1-T1 receptor cells respectively, coated on 100µg/ml ampicillin resistant LB plate. On the second day, a single colony was selected and shaken in liquid LB medium, plasmid DNA were extracted for Sanger sequencing, wild type and mutant DNA were selected respectively for electroporation to K562 cells, and RNA were extracted 48 hours later. After reverse transcription into cDNA, PCR were performed, and finally Sanger sequencing analysis were performed to detect the splicing mode of variations.

Results: c.98+3A>G, c.155+5G>A, c.204-1G>A and c.374+5G>A variations were found in ABO variant individuals in our laboratory. Here, we successfully constructed wild type and variant minigene splicing reporter plasmids for the above variation sites. Through the electroporation, RNA extraction, cDNA synthesis, PCR and the final Sanger sequencing, the results showed that c.98+3A>G led to deletion of exon 2, c.155+5G>A led to deletion of exon 3, c.204-1G>A led to deletion of exon 5, and c.374+5G>A led to deletion of exon 6, compared with the wild type.

Summary / Conclusions: Variation in ABO intron/exon splice sites lead to produce different mRNA forms with adjacent exon sequences deletion through RNA splicing, resulting in weak ABO antigen expression in variant individuals.

P571 | A novel large deletion in the ABO gene resulting in blood group O

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Background: ABO is the clinically most important blood group system, but genotyping remains a challenge. In addition to the major ABO

alleles, numerous variants responsible for both weak A and B subgroup expression, as well as group O phenotype, have been identified. The overwhelming majority of the latter allelic variants abolish ABO glycosyltransferase activity due to a common single-base deletion, c.261delG. Other reasons underlying the null phenotype exist but apart from the well-characterized c.802G>A, most will complicate genotyping efforts. Here we describe a novel large deletion of 2242 bp in the ABO gene, identified in samples from a pregnant woman and a Czech family, resulting in blood group O.

Aims: To investigate the underlying molecular cause for the absence of A antigen on the RBCs from four individuals.

Methods: Blood samples from a pregnant woman and an unrelated family with three generations were investigated independently. Standard serological techniques were used. Red blood cells (RBCs) from two of the family members were tested with monoclonal anti-A, -B and -H by flow cytometry. The ABO locus was analysed by PCR-ASP/PCR-RFLP genotyping and direct DNA sequencing of all seven exons and parts of the introns.

Results: The three family members' RBCs showed no reactions with anti-A in forward typing and two of them presented anti-A1 in addition to anti-B in reverse typing. The pregnant woman's RBCs were negative with anti-A in the forward typing and the reverse typing showed 2+ with A₁ RBCs while A₂ RBCs were negative, hence also presenting anti-A1 and anti-B in plasma. Initial attempts towards ABO genotyping showed heterozygosity for ABO*A1 in the woman and the family members, which prompted further investigation. By flow cytometry, the vast majority of RBCs stained as the group O control, although a very small portion (<0.1%) of the cells expressed low levels of A antigen, reminiscent of a microchimeric pattern. Anti-B was negative and anti-H strongly positive, as expected. The same pattern was seen for both family members tested. ABO genotyping showed ABO*A1.01 in combination with either ABO*O.01.01 or O.01.02 in all individuals tested, which is usually consistent with normal expression of A antigen. Sequencing of all seven ABO exons and parts of the introns revealed a large deletion of 2,242 bp, from c. 29–177 in intron 1 to c.156–241 in intron 3, encompassing parts of intron 1, the whole of exon 2, intron 2, exon 3 and nearly all of intron 3.

Summary / Conclusions: A large deletion leading to a novel O allele was identified in one individual and the same deletion was also found in three additional samples from a family. No known relationship exists between the index case and the investigated family members. The large deletion probably results in a shortened stem region in the A₁ glycosyltransferase and complete absence of its transmembrane domain (amino acids 17–37). This will change the overall structure of the enzyme and severely decrease its activity, if expressed at all. A similar but apparently different deletion encompassing 2,169 bp and involving sequences from intron 1 to intron 3 was described in another family with group O phenotype without anti-A in plasma (Matzhold et al., Transfusion, 2016). Finally, it can be discussed if these kinds of alleles with large deletions encompassing exons 2 and 3 are indeed O alleles or give rise to a very weak form of blood group A with lack of or weakening of anti-A in plasma.

P572 | Blood antigen genotyping using MALDI-TOF MS technology to establish the selected donor registry in Poland

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Background: The access to a blood donor registry with extended blood group phenotypes/genotypes is necessary for finding compatible units for immunized patients with unique multi-specific alloantibodies or antibodies to high-frequency antigens. From 2021, within the program "Ensuring the self-sufficiency of the Republic of Poland in blood and blood components for the years 2021–2026", the establishment of such donor registry has been developed including blood donors with homozygous phenotypes in Rh, Kell, Kidd, Duffy or MNS blood group systems, confirmed by genotyping, with determined the high/low-frequency antigen genotypes (HFA/LFA), clinically relevant for the Polish population.

Aims: The aim was high-throughput blood antigen genotyping of phenotyped blood donors using matrix-assisted laser desorption/ionization, time-of-flight mass spectrometry (MALDI-TOF MS) to establish the selected blood donor registry.

Methods: 1) Results of selective phenotyping of Jk^a/Jk^b, Fy^a/Fy^b, S/s, M/N antigens in Rh(C,c,E,e), K-, K+k- homozygous donors, performed between 2021 and 2023 by 17 of Regional Blood Transfusion Centers (BTCs), reported to the Institute of Hematology and Transfusion Medicine (IHTM) to select samples for further molecular screening 2) Genotyping of 4000 selected donors (homozygous in at least 2 tested blood group systems, excluding M/N; or Jk^a or Fy^a negative phenotypes) was performed on MassARRAY Dx Analyzer 4 using Hemo ID™ Donor Quick Screen (DQS) and Custom Rare Panels (including 14 additional SNVs for HFA/LFAs as well as platelet HPA-1) and analyzed by Typer software (Agena Bioscience). Rare HFA negative/HPA-1a negative genotypes have been successively confirmed from the next donation by phenotyping and molecular tests: ID CORE XT/ID HPA XT (Grifols) or *in-house* qPCR.

Results: In a 3-year period, BTCs reported the results of Jk^a/Jk^b, Fy^a/Fy^b, S/s and M/N phenotyping of 27 369 donors and 58% of them have been selected for further genotyping. To the end of 2023, one third of selected donors was sent to IHTM. The molecular screening of 4000 selected donors: in 96.4% confirmed their phenotypes (in the rest: 92 donors with confirmed discrepant pheno/genotypes; 53 waiting for further explanation); revealed 59 donors with rare blood groups (1.5%) such as YT*B (20), LU*02.14 (12), CO*B (10), RHCE*02.08 (9), LW*B (4), KN*B (2), LAN negative genotypes (2) and identified 92 HPA-1a-negative donors (2.3%). To the end of 2023 nine (15%) rare blood group donors and thirty (32.6%) HPA-1a-negative donors were confirmed by phenotyping and molecular tests from the next donation.

Summary / Conclusions: Blood antigen genotyping with MALDI-TOF MS technology allowed to establish the selected blood donor registry adjusted to Polish immunized patients. In 2023 the presented registry was used successfully to find the donors compatible for alloimmunized patient with anti-Lu:8. In addition, including of HPA-1

genotyping into HFA/LFA panel enriches the Polish blood service giving the access to HPA-1a-negative donors.

P573 | Rapid clarification of suitable blood supply in an anemic patient with inconclusive serologic and genotypic results for the RhCE blood group

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Background: In rare cases, standard methodology in routine diagnostics fails to unequivocally determine matched blood for patients in need of blood transfusion. If this happens in a time-sensitive setting, the most parsimonious transfusion recommendation to avoid alloimmunization is usually selected. We present here a case of an anemic patient with unknown transfusion history whose erythrocytes showed strong reactions (4+) with anti-D, anti-C, anti-c and anti-e, as well as a positive direct antiglobulin test (DAT). The detection of an anti-C antibody in the serum and eluate led to further investigations. Molecular analysis for important *RHD* and *RHCE* variants showed, beside the expected heterozygosity for *RHCE**C/c and homozygosity for *RHCE**e, the variant c.667G>T in exon 5. In the absence of knowledge on the allelic background of this variant, allele frequencies as well as the observed phenotype suggest that the allele *RHCE**01.07 (also known as *RHCE**ceMO), reported as being associated with partial c, is the more likely genetic set-up than the allele *RHCE**02.22 associated with weak C.

Aims: Here, we evaluated a newly developed Nanopore sequencing approach designed for constructing full *RHD* and *RHCE* haplotypes through co-amplification of both genes in generic long-range PCRs. In particular, we assessed its suitability to resolve the present case within one working day.

Methods: The patient was serologically typed for D, C, c, E, e, C^w and K by hemagglutination in gel cards. Standard serological methods such as DAT and indirect antiglobulin tests on test cells with and without papain treatment were used for antibody specification. Molecular genotyping was done by commercial PCR-SSP (sequence-specific priming) kits for *RHD* and *RHCE*. The amplicon sequencing approach was based on long-range PCR with five primer pairs that simultaneously amplify both *RH*-genes. To phase variants in both genes, amplicons (12 – 15.5 kb) were designed to overlap by at least 1 kb. Sequencing library preparation was carried out according to guidelines by Oxford Nanopore Technologies (ONT). Filtered sequences were mapped against a tailored region of HG38 reference including *RHD* and *RHCE*.

Results: Protocols for the long-range PCR and ONT library preparation fitted well into half a working day. Sequencing for ~4 h on a MinION flowcell resulted in more than 20,000 target reads and, despite a relatively unequal distribution among fragments, a coverage above 200× for all PCR-products. Phasing of reads mapping to *RHCE* unexpectedly assigned c.667T to the *RHCE**C allelic background (*RHCE**02.22).

Consequently, the patient's final transfusion recommendation was blood from a ccD. ee (or ccddee) donor. Regarding the anti-C antibody, its presence in the eluate more than three months after the last transfusion clearly favors an auto-origin. Given the patient's strong serological reactions with anti-C, the previously reported phenotypic information for the *RHCE**02.22 allele (weak C) may be incomplete.

Summary / Conclusions: Amplicon-based long-read sequencing by ONT proved a promising tool in urgent situations where optimal blood supply depends on unambiguous genetic allele information. To our knowledge, no other method can accurately elucidate the allelic background of the *RH* locus within approximately one working day, from DNA extraction to entire haplotype sequences.

P574 | The role of ABO blood group system in the occurrence of venous thromboembolism

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Background: The association of blood group antigens, especially those from the ABO system and some diseases is well known. The alleles of the ABO locus have functional effect on the level of some plasma coagulation factors which contributes to a thrombophilic condition and increase the risk for occurrence of venous thromboembolism.

Aims: To examine relationship between ABO blood genotype with the occurrence of thromboembolic disease, as well as to assess the risk for thrombosis in individuals with non OO blood group genotype in comparison to those who poses it.

Methods: This prospective case control study included 52 patients with a confirmed diagnosis of VTE and a control group of 50 healthy subjects who do not have a personal or family history of thrombosis. In addition to the serological ABO typing, ABO alleles from ABO blood system (A1, A2, O1, O2, B) were determined by molecular methods (PCR-SSP and RT-PCR).

Results: The most frequent genotype in the VTE patients is O1A1 with 40.4%, while in control group is O1O1 with 32%. The genotype O1A1 is most frequent in patients with pulmonary embolism (PE) with 55.6% in comparison to patients with deep venous thrombosis (DVT) in which it is 35.7% and to the control group with 24%. The results from ABO phenotyping and genotyping performed in patients with VTE show significant connection between non OO genotypes and the occurrence of VTE, in manner of increased thrombotic risk. Non OO genotypes have significantly higher prevalence (86.5%) in patients with VTE in comparison to OO genotypes with 13.5 % ($p < 0.05$).

Summary / Conclusions: The results confirm association of ABO blood group system with the occurrence of venous thromboembolism. ABO alleles can be useful VTE risk assessment tool. We suggest ABO genotyping as additional test in the thrombophilia assessment panel.

P575 | A novel JK*02 null allele in a patient of Indian origin

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Background: The Kidd (JK) blood group system currently contains three antigens expressed at the surface of red blood cells (RBCs): the polymorphic, antithetical Jk^a (or JK1) and Jk^b (or JK2) antigens; and the high prevalence JK3 antigen. Alloantibodies directed against Kidd antigens are considered to be clinically relevant. Kidd antigens are carried by the solute carrier family 14 member 1 glycoprotein, a urea transporter encoded by the SLC14A1 gene. So far, more than 60 variant alleles carrying molecular variants and altering partially or totally the expression of either antigen have been reported officially (ISBT 009 JK blood group alleles v8.1), but many others are likely to be found in the future.

Aims: In this work, we sought to investigate the molecular basis of an atypical JK phenotype in a patient of South Asian (Indian) origin.

Methods: Blood group testing (including ABO-RH-K and extended typing) and irregular antibody screening were carried out. Genomic DNA was extracted from a whole blood sample and first analyzed by a commercial technology (BioArray HEA BeadChip Kit, Immucor) for genotyping the c.838 nucleotide position of the SLC14A1 gene that defines Jk^a/Jk^b antigen expression. Second, direct sequencing of all ten exons of the gene (NM_015865.7) was carried out. When necessary, the PCR product was subcloned into a plasmid vector for phasing.

Results: An Indian patient suffering from infectious disease with severe thrombocytopenia required transfusion with two RBC units. He had a history of lupus nephritis, as well as autoimmune hemolytic anemia (AIHA) and pulmonary venous thrombosis (PVT). Upon testing, autoantibodies already reported before were identified. Routine serological typing yielded two different results for JK typing: 1/ Jk(a-b-) and 2/ Jk(a+^wb-). Molecular analysis by the commercial kit identified a heterozygous genotype at position c.838, suggesting that this patient carries two variant alleles on a different background: one JK*01 (or JK*A) and one JK*02 (or JK*B). In addition to this heterozygous position, further investigation by Sanger sequencing identified two missense variants at the heterozygous state: 1/ c.130G>A (p.-Glu44Lys) in exon 3 (rs2298720; gnomAD v4.0.0, minor allele frequency in South Asians [MAF] = 0.2545), and 2/ c.929T>G (p.Leu310Arg) in exon 8, a very rare variant that is more frequent in South Asians (rs756906464; MAF = 0.0002526). Subcloning and sequencing confirmed that the latter variant is carried by the JK*02 allele. In addition, the frequent synonymous c.588A>G variant (rs2298718; MAF = 0.9332) was found at the homozygous state.

Summary / Conclusions: On the basis of both the serological and molecular data, our findings suggest that this patient likely carries: 1/ the JK*01W.06 allele, characterized by the common c.130G>A variant, which is known to weaken dramatically JK antigen expression (Jk^a here), and c.588A>G; and 2/ a novel JK*02 null allele, namely JK*02 (c.588G, c.929G), which completely abolishes the expression of the Jk^b antigen. Although we cannot definitely exclude the deleterious effect of another variant located in a regulatory region of the gene, we conclude that the p.Lys310Arg change at the protein level is causative for the Jk(b-) phenotype. Indeed, more than 20 JK null alleles with one or more missense variants have been already reported. Overall, we recommend transfusing this patient with Jk(a+b-) RBCs in a clinical context for preventing alloimmunization.

P576 | Screening for del phenotype in RhD negative donors in national blood centre, Malaysia

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Background: The DEL phenotype represents a very weak form of the D variant, detected only by the adsorption and elution technique of anti-D or by molecular methods. DEL-phenotype donors mistyped as RhD negative can lead to alloimmunization if transfused to patient which can be harmful especially in female of reproductive age. DEL blood donors are typed as D-negative because DEL typing methods are not currently used in routine practice for donor screening in Malaysia. Molecular techniques have now been used to identify DEL variants. The DEL (K409K) is the most prevalent allele, and it is frequently seen in the East Asian population.

Aims: To determine the frequency of DEL phenotype in RhD negative blood donor population at National Blood Centre, Malaysia.

Methods: A total of 372 RhD-negative blood donor samples were included in this study, which was conducted over a period of 6 months (January to June 2023). All these blood samples were tested for extended Rh typing, including C, c E, and e antigens, by column agglutination techniques, as well as for adsorption and elution testing. The eluate and the last wash supernatant were used for an indirect antihuman globulin test (IAT) against O RhD-positive and O RhD-negative cells by gel technique using the LISS Coombs' AHG gel cards (Biorad, Switzerland). Those donor samples that were found positive on adsorption and elution testing were also further investigated by the column agglutination technique for the direct antihuman globulin test (DAT), weak D testing, and auto-control tests. As confirmation, molecular testing was performed using single-specific primer-PCR (SSP-PCR) (Bagene, Germany).

Results: Out of the total 372 RhD negative donor samples tested, 28 (7.5%) samples were found to give positive results for the DEL phenotypes by the adsorption and elution technique. It was found that 27/28 (96.4%) donor samples had the phenotype Cde/Cde (r'r), and 1/28 (3.6%) had the phenotype Cde/Cde (r'r'). All 28 samples that

were found to be positive for the DEL phenotype also gave negative results for DAT, weak D testing, and auto-control. For molecular testing, it showed that all samples were confirmed to be DEL variants (K409K).

Summary / Conclusions: We concluded from this study that the frequency of DEL phenotype in our RhD negative donor population at National Blood Centre, Malaysia is 7.5%. The findings of this study will be of great benefit and importance in the future local blood transfusion service. We conducted this study as NBCs in Malaysia to determine the frequency of the DEL phenotype among RhD-negative blood donors. A comprehensive DEL screening protocol is required to prevent the avoidable immunization of susceptible patients.

P577 | Journeying through RhD variants—14 years of study and anti-D sensitization

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Background: Rhesus D antigen is a critical determinant of transfusion compatibility and pregnancy management. A serological weak D phenotype is the expression of an amino acid substitution in the RhD protein or an RHD-RHCE-D gene conversion resulting in a D variant antigen. They are classified as weak D or partial D, depending on whether there is a quantitative or qualitative change. According to most authors, individuals with weak D types 1, 2, 3, 4.0 and 4.1 can safely receive D+ red blood cell (RBC) units and do not require anti-D prophylaxis, while the remaining patients should be treated as D negative due to the known or unknown (but feared) risk of anti-D alloimmunization. Surprisingly, there are few cases of presumed D-positive patients developing anti-D antibodies.

Aims: We aim to report the demographic distribution, transfusion and obstetric management of D variants over fourteen years, focusing on some cases of anti-D sensitisation.

Methods: We retrospectively collected demographic, immunohematologic, transfusion and immunization data from all patients serologically classified as weak D (from 1+ to 3+), whose diagnosis could imply a relevant change in their transfusion or obstetric management. Selected samples were first analysed in H12O¹ Immunohaematology lab using the "DiaClon ABO/D + Reverse Grouping" card (BioRad) and then sent to our reference Immunogenetics centre (CTCM²) to define subtypes of the RHD gene by PCR-SSO and PCR-SSP (ID RHD XT Grifols, Spain; and RBC-FluoGene Verify Extend Inno-train, Germany). Coombs direct, autocontrol, adsorption and elution tests were done in positive screening samples.

Results: We included 153 patients, median age 35 (28-46), 121 (79%) females. The reason for RhD typing was pregnancy in 94 cases (61%), pre-transfusion testing in 32 patients (21%) and other reasons in 27 patients. Partial D variants were 17% of the total (see Table 1). cEe, Cce and ce phenotypes were associated with non-risk variants in 97.3%, 77.6% and 51.3% respectively. 27 patients were transfused, 19 (70%) pre-genotyped and 11/13 received D-RBCs when they could have received D+. Molecular diagnosis helped us to save 7 D- RBCs out of the 25 that could have been saved. In addition, anti-D was detected in six patients (4%) in the post-transfusion scenario (Table 2). Auto-Anti-D was detected in three patients with weak D, due to evidence of autoagglutination. Allo-Anti-D was detected in the two DIIIc patients who received D+

P577 - Table 2. Anti-D patients

RHD variant	Autocontrol test	Direct Coombs test	Auto/ Allo antibody	RhD+ RBC units transfused
Weak type 1	+w	+w	Auto	2
Weak type 2	1+	3+	Auto	1
	+w	+w	Auto	8
Weak type 1	Neg	Neg	Allo	2
Partial DIIIc	Neg	Neg	Allo	4
	Neg	Neg	Allo	17

P577 - Table 1. RhD variants.

		All patients (n = 153)	% 2+ serologic RhD reaction	CcEe phenotype (%)
Weak D, n (%)	Type 1	51 (33.3)	96	Cce (86)
	Type 2	37 (24.2)	86	cEe (95)
	Type 4.0 (DAR3), 4.1 (DAR4) and 4.2.0 (DAR1) families	25 (16.3)	72	ce (92)
	Type 59	11 (7.2)	64	Cce (100)
	Type 3	8 (5.2)	88	Cce (100)
Partial D, n (%)	DAR, DAR2 and DAR2.1	12 (7.8)	67	ce (92)
	DVI type IV	5 (3.9)	60	ce (60)
	DIIIa, DIIIc	3 (1.4)	33	Cce (67)
	DAU-4	1 (0.7)	0	Ce (100)

(because of a 3+ hematic RhD reaction). Last patient was weak type 1 (just one allele) and the other allele deleted, which explains this unexpected event.

Summary / Conclusions: Although international transfusion guidelines for D-variant patients are well established, cases of anti-D immunization may occur. Accurate differentiation between autoantibodies and alloantibodies is crucial for transfusion strategies and neonatal care in these patients.

P578 | Molecular characterization of RhD variants with serological reactions of mixed field in blood donors and recipients from Southern Brazil

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Background: D variant frequency varies among ethnic groups, and knowledge of the main variants present in a specific population is important in transfusion practice. Currently, RH genotyping has been recommended to resolve serologic atypical reactions or discrepancies found in the routine and to differentiate weak and partial Rh antigens.

Aims: The aim of this study was to perform molecular analyses on DNA samples from blood donors and recipients from the South of Brazil, who exhibited atypical reactions of mixed field in RhD typing.

Methods: RhD typing was performed using column agglutination techniques (Ortho Clinical) and gel testing (Bio-Rad) on blood samples from donors and patients. The molecular analysis of these samples was conducted using conventional PCR techniques, RHD BeadChip (Immucor), and genomic sequencing.

Results: The blood samples from 22 blood donors and 2 patients showed serological reactions of mixed field with a reactivity of 1+. Molecular analyses showed in blood donors, eight (36%) Weak D type 18, five (23%) Weak D type 1, four (18%) Weak D type 2, one (5%) Weak D type 3, one (5%) Weak D type DAR, one (5%) Weak D type 38, and one (5%) Del 37. In patients, one (50%) Weak D type 1 and one (50%) Weak D type 2 were identified. A family study was conducted for the Weak D type 18 identified in donors, revealing 9 relatives with this phenotype.

Summary / Conclusions: We observed that the weak RhD type 18 variant, although generally uncommon in other regions and populations, exhibited a surprisingly high frequency among the blood donors studied in the Southern region of Brazil. These findings highlight the importance of serological and molecular characterization of the RhD antigen in this specific region, as the prevalence of this variant may differ significantly from other geographic areas and suggests the possibility of a founder effect. Our study emphasizes the importance of molecular investigation when serological variations such as mixed field are identified in the RhD phenotyping routine.

P579 | Molecular basis of RhD negativity in Indians: Development of population specific RHD genotyping strategy

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Background: Alloantibodies against the Rh blood group antigens are known to be a major cause of HDFN and HTR. Among Rh antigens, the D antigen is the most immunogenic. The high degree of homology, opposite orientation and proximity shared by *RH* genes (*RHD* and *RHCE*) promotes the formation of numerous variants. Depending on the reagents and the techniques used, D variants give varied results in RhD typing in different laboratories and create confusion. Knowing the correct RhD status is clinically important, as the D variant individual is considered RhD positive as a donor and RhD negative as a patient/recipient of blood transfusion. Hence, to overcome the limitations of serology, D negative samples should be screened using a DNA-based approach to exclude RhD variants for preventing alloimmunization. Genotyping requires knowledge about the molecular mechanism causing D negativity in a given population. The *RHD* gene deletion, pseudogene (*RHD Ψ*) and the *RHD-CE-D* hybrid formation are the main cause of D negativity. The percentage distribution of these different mechanisms varies with the population. *RHD* genotyping strategies based on molecular methods have to be developed by few countries to determine the correct RhD status. About 5-7% of Indians have a D-negative phenotype. However, the molecular mechanisms responsible for RhD negativity in Indians is not known.

Aims: The aim of the study is to determine the molecular basis of D negativity in apparently RhD negative Indians and thereby develop a suitable DNA-based diagnostic strategy for correct *RHD* typing in Indians.

Methods: Over 3023 apparently RhD negative individuals were screened with a panel of anti-D reagents and typed for presence of C, c, E & e antigens. All the samples were primarily screened for *RHD* exon 5, 10 and weak D type 150 using indigenous Multiplex PCR to identify D variants. All samples were further tested for all *RHD* exons by the quantitative multiplex polymerase chain reaction (QMPSF) assay for copy number analysis to identify presence of *RHD-CE-D* hybrids. Samples positive for one or more *RHD* exon(s) were further screened by *RHCE* QMPSF. Sequencing was performed on uncharacterized samples.

Results: 1.2% D variants were identified in RhD negative individuals tested. QMPSF studies showed 2% of the RhD negative samples harboured *RHD* exons, of which 1.74% carried the gene in *RHD-CE-D* hybrids form and 0.23% showing single nucleotide changes. All samples with the rr phenotype showed complete deletion of the *RHD* gene. Out of 253 RhD negative samples positive for 'C' and/or 'E' antigens, 23% showed presence of one or more *RHD* exons. *RHD-CE* (3-9)-D hybrid was the most frequent hybrid allele identified followed by *RHD-CE*(3-8)-D and *RHD-CE*(4-9)-D.

Summary / Conclusions: This comprehensive study has successfully determined the mechanism of D negativity in Indians. Predominant

mechanism of RhD negativity in Indians is *RHD* deletion & presence of *RHD-CE-D* hybrids. Based on the data generated, we proposed three different strategies/ algorithm for correct RhD typing in Indians. Performing C, c, E, e phenotyping and genotyping for only 8% of C/E+ RhD negative samples (no pooling). Complete genotyping strategy for reference laboratory (distinguishing between D negative and D variants phenotype with *RHD-CE-D* hybrids). RHD genotyping all serological RhD negative samples by pooling method. The strategy will facilitate decision making for blood transfusion and can also be applied for fetal *RHD* typing.

P580 | Abstract withdrawn

P581 | One novel single nucleotide polymorphism c.424A>G on A1.02 gene in ABO glycosyltransferases leads to Aweak phenotype

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Background: The dysfunction of the ABO glycosyltransferase (GT) enzyme, which is caused by mutations in the ABO gene, can lead to weak ABO phenotypes.

Aims: In this study, we have discovered a novel weak ABO subgroup allele and investigated the underlying mechanism to causing its Aweak phenotype.

Methods: The ABO phenotyping and genotyping were performed by serological studies and direct DNA sequencing of ABO gene. The role of the novel single nucleotide polymorphism (SNP) was evaluated by 3D model, predicting protein structure changes, and *in vitro* expression assay. The total glycosyltransferase transfer capacity in supernatant of transfected cells was examined.

Results: The results of serological showed the subject was Aweak phenotype. A novel SNP c.424A>G (p. M142V) based on ABO*A1.02 was identified, and the genotype of the subject was AW-var/O.01 according to the gene analysis. *In silico* analysis showed that the SNP c.424A>G on the A allele may change the local conformation by damaging the hydrogen bonds and reduce the stability of GT. *In vitro* expression study showed that SNP p.M142V impaired H to A antigen conversion, although it did not affect the generation of GTA.

Summary / Conclusions: One novel AW allele was identified and the SNP c.424A>G (p.M142V) can cause the Aweak phenotype through damaging the hydrogen bonds and reducing stability of the GTA.

P582 | The complex molecular mechanism investigation of a rare D- phenotype using long-read sequencing

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Background: Rh blood group is a complex and polymorphic blood group system with clinical significance. The D- phenotype is an extremely rare

RhCE variant that appears as a complete absence of RhCE antigen. The RhCE variants could be caused by diverse molecular mechanisms of *RHCE* and *RHAG* gene locus. The detection of *RH* genes is difficult because they are highly homologous and prone to complex structural variations. Long-read sequencing technologies may offer improvements in the characterization of genes that are currently difficult to assess.

Aims: The molecular mechanism of an individual with D- phenotype was revealed by three-generation long-read sequencing.

Methods: Standard haemagglutination methods were used to investigate the subjects' red blood cells. The *RHD*, *RHCE* and *RHAG* genes were amplified by segmented long fragment PCR to cover the full length of the genes. Single molecule real-time (SMRT) libraries were constructed from purified amplified fragments, including DNA damage repair, end-repair, adapter ligation and the final library was bound with sequencing enzymes and primers through Sequel Binding Kit 2.2 (Pacific Biosciences) and Internal Control Kit 1.0 (Pacific Biosciences). 150 pM DNA-polymerase complexes were finally loaded and sequenced with the Sequel II platform (Pacific Biosciences). A bioinformatic analysis for full-length *RHD*, *RHCE* and *RHAG* haplotype sequence analysis was processed according to the reference genome (ISBT).

Results: The proband was phenotyped as group D+C-E-c-e-. Molecular analysis performed in the proband was found to carry a normal *RHD* gene. The *RHCE* gene had structural change based on the background genotype *RHCE**Ce/*RHCE**Ce. The *RHCE* gene lacked a fragment spanning Intron 2 to Intron 3. Since *RHCE* and *RHD* homologous sequences were considered in the long segment amplification, the failure of this segment suggested that this *RHCE**Ce allele may have a large complex structural variation in this region. Additionally, one of the *RHCE**Ce variant allele had a variant c.569_572dupCTCT, resulting in frame shift and premature termination(p.Pro192Serfs*8). The combination of structural change and insertion variation given rise to two novel *RHCE**Ce variant alleles. Unfortunately, this structural change was not clear, and it was speculated that it may be a very complex *RHD-RHCE* gene exchange, which required follow-up long-read whole-genome sequencing to clarify. Interestingly, when performing haplotype analysis of *RHAG* gene in this sample, variations c.808G>A and c.861G>A in one allele were identified, forming a novel *RHAG* variant Rhmod or Rhnull allele. Since the proband was found to carry another normal *RHAG* allele, the function of the protein encoded by the novel variant allele requires further verification.

Summary / Conclusions: Novel *RHCE* and *RHAG* allele were identified in a Chinese individual with D- phenotype, which expanded our knowledge of the underlying molecular mechanism of RhCE variants and contributed to the improvement of clinical transfusion safety.

P583 | The screening and molecular analysis of the rare In(Lu) phenotype in Southern Chinese population

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Background: The RBCs of the individuals with the Lu(a-b-) phenotype usually seem to lack the Lu^a and Lu^b blood group antigens when tested by standard agglutination techniques, may arise from three different genetic backgrounds: homozygosity for recessive alleles at the BCAM gene locus, called Lu_{null}; hemizyosity for a recessive X-linked suppressor gene GATA1, called Lu_{mod}; or heterozygosity for inactivating mutations in the erythroid transcription factor gene KLF1, called In(Lu). KLF1 is an essential transcriptional activator that drives erythropoiesis. KLF1 variants can result in the “Inhibitor of Lutheran” or In(Lu).

Aims: To elucidate the frequency and molecular mechanism of the rare In(Lu) phenotype in southern Chinese population.

Methods: RBCs from blood donor samples (7206) were screened for Lu^b expression, and those with Lu(b+^u) or Lu(b-) phenotype were subsequently tested for Lu^a expression. Genomic DNA was isolated from samples with Lu(a-b-)/Lu(a-b+^u) phenotype and then sequencing of KLF1 and BCAM coding regions.

Results: 18 of 7206 donors were identified to be In(Lu), six different KLF1 variants were identified. Three were novel: c.914-4_917delCTAGGGGA, c.914-2delA, c.325C>T (p.Pro109Ser) & c.872C>A(p.Thr291Asn) present in three unrelated individuals. And one novel heterozygous mutation of BCAM gene(c.1346A>G, p.Glu449Gly) was identified.

Summary / Conclusions: The prevalence of the In(Lu) phenotype in the southern Chinese population was 0.25%(18/7206), and we identified 3 novel KLF1 alleles and 1 novel BCAM allele.

P584 | Identification of a novel KEL*01M allele encoding a K_{mod} phenotype in a blood donor from Portugal

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Background: The Kell blood group system comprises 38 antigens carried on zinc-dependent Kell metallo-endopeptidase, a type II transmembrane glycoprotein. The glycoprotein and all the antigens are encoded by the KEL gene, located on chromosome 7q34 and encompassing 19 coding exons. The rare phenotypes Kell null (K₀) and Kell mod (K_{mod}) are encoded by mutations in KEL and are characterised by either complete absence, or weak expression of Kell antigens, respectively. K_{mod} is used to describe a broad category of phenotypes in which Kell antigens are expressed very weakly, often requiring adsorption and elution tests

for detection. ISBT currently lists 25 such unique K_{mod} genotypes, usually resulting from homozygosity or heterozygosity for missense mutations or heterozygosity for nonsense mutations.

Aims: We report results from serological and genetic investigations of a 33-year-old male Portuguese blood donor of White European origin, with a weakened expression of K antigen, suggesting the presence of a variant allele.

Methods: Standard serological techniques were performed with anti-K sera, clone MS-56. Genomic DNA was isolated from whole blood. Genotyping was performed using Inno-Train Diagnostik GmbH RBC-Ready Gene KKD test system. All 19 exons of the KEL gene were amplified by PCR and bidirectionally Sanger sequenced.

Results: The donor's cells showed weak reactions by standard tube direct agglutination, and mixed field and weak reactions with Immucor NEO Iris and Echo blood group serology analysers. Strong reactions (4+) were observed with ORTHO AutoVue Innova system, and DiaClon Rh-Subgroups + K BioRad ID-cards. KEL genotyping and Sanger sequencing confirmed the donor to be heterozygous KEL*01/02 (K/k). Furthermore, the donor was heterozygous for a mutation c.974C>G in KEL exon 9, encoding p.Thr325Arg in the Kell glycoprotein. This mutation (dbSNP no. rs761304758) appears to be very rare, detailed in gnomAD database with a total allele frequency across all populations of 0.0000027 and no recorded homozygotes. The combined serology and genetic data indicate that it is carried on the KEL*01 allele, weakening the expression of the K antigen, and thus causing the observed K_{mod} phenotype in this donor.

Summary / Conclusions: Here we have described a novel K_{mod} allele identified in a case study of a Portuguese blood donor. The mutation c.974C>G, p.Thr325Arg, appears to be responsible for weakening of the K antigen in this case. This molecular background of K_{mod} phenotype is not yet recorded in the ISBT Kell blood group allele table, thus extending the number of known K_{mod} alleles which modify expression of Kell antigens.

P585 | Establishment of targeted next-generation sequencing based genotyping method for complete sequence analysis of ABO, RHD and RHCE genes and its application

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Background: Genomic variants in non-coding regions (enhancer, promotor-TSS, splice site, etc.) of blood group antigen coding genes may alter the antigen expression pattern, resulting in cases which are hard to interpret. Complete sequence analysis of red blood cell antigen coding genes which is able to detect both coding and non-coding region variants can help identifying the molecular basis of complex blood group serology cases.

Aims: To establish and apply targeted next-generation sequencing (NGS) based genotyping method that is able to analyze the complete sequence of ABO, RHD and RHCE genes.

Methods: Test samples with known ABO, RHD and RHCE genotypes (n = 4) were used for method establishment. Three gene specific

amplicon sets, which cover the complete sequence of *ABO*, *RHD* and *RHCE* gene respectively, were amplified, purified and quantified to create an equimolecular pool. For each gene and each sample, NGS libraries were prepared and sequenced following the manufacturer's protocol. Reads were aligned to hg19 (for *ABO* gene analysis), *RHCE*-masked hg19 (for *RHD* gene analysis) and *RHD*-masked hg19 (for *RHCE* gene analysis). Coding and non-coding region variants were called, filtered and compared with ISBT blood group allele table to perform *ABO*, *RHD* and *RHCE* genotyping of each sample. In addition to the test samples, two weak D and eight D- samples which were previously tested by Sanger sequencing were analyzed by targeted NGS for method validation.

Results: In test samples, all amplicons were successfully amplified except the cases which are expected to have structural variants (ex: deletion of the whole *RHD* gene), and NGS based genotyping results of *ABO*, *RHD* and *RHCE* genes were the same as expected. All *RHD* gene amplicons were successfully amplified in weak D samples and coding region variants which were identified in previous Sanger sequencing were also observed in targeted NGS. In D- samples, three samples with *RHCE-RHD* (3-8)-*RHCE* hybrid and one sample with *RHCE-RHD*(7-9)-*RHCE* hybrid showed missing *RHCE* gene amplicons in the corresponding regions. Three D- samples with no known D- causing variants showed missing *RHCE* gene amplicons in intron 2 ($n = 1$) or intron 2 & 7 ($n = 2$), therefore were excluded from the downstream analysis. One D- sample with no known D- causing variants was successfully amplified and sequenced. Targeted NGS result of this sample had discordance with Sanger sequencing result which showed heterozygous variants in exon 2. NGS data showed no heterozygous variants in exon 2 and two additional homozygous variants (c.1170C>T, c.1193T>A) in exon 9 were identified. Further effort to resolve the cause of the discordance is needed.

Summary / Conclusions: We successfully established the NGS based genotyping method for complete sequence analysis of *ABO*, *RHD* and *RHCE* genes. Coding and non-coding region variants were called with accuracy, which enabled us to perform high resolution genotyping. This method, however, is amplicon-based and is inappropriate for samples with structural variants due to the inexistence of PCR products. Three D- samples with missing intronic region amplicons suggest the possibility that variants which inhibited the amplification may have also caused the D- phenotype. One D- case which showed discordance may have been caused by sequence similarity between *RHD* and *RHCE*, hybrid gene formation or other unknown reasons. Further investigation of these samples with non-targeted NGS method like whole genome sequencing (WGS) is needed to reveal the exact genetic nature.

P586 | Abstract withdrawn

P586-A | Value of genomic DNA sequencing in the characterization of two exceptional cases of anti-Lu13 in Swedish and Syrian patients

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Background: The Lutheran blood group system (LU) consists of 28 antigens carried on a single-pass transmembrane immunoglobulin (Ig) superfamily glycoprotein (gp) encoded by *BCAM*. They are distributed across five extracellular Ig domains. *BCAM* is located over 12.5 kb on chromosome 19q13.2 and consists of 15 exons. Two isoforms are produced by alternative splicing: the full-length Lu-gp, and a shorter *BCAM* gp, in which intron 13 is retained, resulting in an earlier stop codon, and shorter cytoplasmic tail. Lu13 is a high-prevalence antigen in this system. The allele defining the LU:-13 phenotype, LU*02.-13, is defined by three single nucleotide variants: c.1340C>T (p.Ser447Leu, rs117737673), c.1671C>T (p.Ser557 =, rs28399658), c.1742A>T (p.Gln581Leu, rs28399659). Of these, only p.Ser447Leu is located extracellularly in the 5th Ig domain, and most likely defines the Lu13 antigen. The allele has a frequency of ~2% but despite this, only 4 examples of anti-Lu13 have been reported.

Aims: To sequence *BCAM* to identify the likely rare Lu phenotype in two unrelated female patients with Lu-related antibodies; the first, a 70-year-old, multiply transfused Swedish patient; the second, a 26-year-old pregnant woman of Syrian origin.

Methods: Standard serology methods were used for antibody identification. Genomic DNA isolated from whole blood was prepared for whole genome sequencing (WGS) using a PCR-free clinical protocol in case 1. WGS was subsequently performed on a NovaSeq 6000. Sequencing data were filtered and analysed using an established pipeline, and specific alleles assigned following manual inspection and data interpretation. Sanger sequencing of *BCAM* exons 9 to 13 was performed in case 2.

Results: Case 1: The antibody investigation showed an antibody to a high-prevalence antigen in the patient's plasma. The reactivity profile with different proteases narrowed the specificity to the LU system. The plasma was nonreactive with In(Lu) Lu(a-b-) red blood cells (RBCs), and the patient's RBCs typed Lu(a-b+), Lu:-14, Vel+, Lan+. Further Lu typing reagents were not available. Due to the patient's imminent transfusion needs, an established clinical WGS pipeline was used, with the purpose to investigate *BCAM*. The results revealed compound heterozygosity for the LU*02.-13 allele, and for c.711C>A (p.Cys237Ter, rs3810141), which defines LU*02N.03. No other changes in *BCAM* were observed. Case 2 also demonstrated a pan-reactive antibody, reactive 3+ at IAT, 1+ with papain-treated RBCs. All RBCs tested were reactive except In(Lu) Lu(a-b-) and one example of Lu:-13 RBCs, and the RBCs of her sister. Sanger sequencing showed homozygosity for LU*02.-13.

Summary / Conclusions: We used DNA analysis to characterize the molecular background of a high-prevalence LU antigen in two unrelated patients. We determined the first patient's genotype to be

LU*02.-13/ LU*02N.03, consistent with the LU:-13 phenotype and conclude that the patient's antibody is anti-Lu13. While the LU*02.-13 allele is relatively frequent in Europeans and other populations, anti-Lu13 is rarely seen. The LU*02N.03 allele is very rare and has only been described once in a Lu(a-b-) Japanese man. The clinical importance of antibodies to high-prevalence LU antigens varies. While no blood was required in the second case, 3 crossmatch-compatible Lu(a-b-) units were transfused to patient 1 uneventfully. Our report shows the value of using DNA analysis when specific typing reagents are not available, and that a well-established WGS pipeline can be an effective tool.

P587 | A novel c.1227+2T>C variant in RHD Gene in a case representing serological D-negative phenotype

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Background: Among over 40 identified DEL variants, RHD*1227A (RHD*01EL.01), which is relatively common in East and Southeast Asia, represents a serologically D-negative. The DEL variant caused by the skipping of exon 9 in the RHD gene, due to the loss of the donor splicing site, has not been reported yet.

Aims: This study reports a novel case of a Korean individual with a D-negative phenotype, suspicious of DEL phenotype, characterized by a homozygous splicing site variant in intron 9 of RHD gene.

Methods: An EDTA coated sample was sent to our reference laboratory for RHD gene testing. Upon receipt, we performed standard serologic tests and multiplex PCR, which included the promoter region, intron 4, exon 7, and exon 10 of RHD gene. Following this, we conducted Sanger sequencing on all exons, from exon 1 through exon 10, of RHD gene. NM_016124.4 (GRCh37) was used as the reference sequence. In-silico prediction cut off was set Δ Score > 0.2 of Splice AI.

Results: An eighty-four-year-old female with uremia underwent a transfusion test. In serologic tests, her Rh phenotype showed Cce. The Weak D test showed a trace result in the direct antiglobulin test, but the quantitative antiglobulin test for IgG also yielded a trace result. This suggests a false positive direct antiglobulin test, raising suspicion of a D-negative phenotype. RHD PCR confirmed the presence of RHD gene. In Sanger sequencing, a homozygous c.1227+2T>C variant was detected in intron 9. The allele frequency of this site has not been reported in any population cohort in the public database (GnomAD v4.0.0) or the Korean allele frequency database (KRGDB 1722). In-silico prediction showed a high probability of splicing change, with splice AI Δ scores of 0.86 for acceptor loss type and 0.90 for donor loss type. This result was predicted to skip exon 9 due

to the loss of the donor splicing site, eventually representing a similar effect to the Asian-type DEL.

Summary / Conclusions: This study reveals a novel c.1227+2T>C variant in RHD gene in a Korean individual, characterized by a D-negative phenotype due to a previously unreported homozygous splicing site variant in intron 9 of the RHD gene.

P588 | c.137-8C>T GYPB mutation found in a donor without the GYPB(P2) allele

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Background: Two alleles of the GYPB gene, encoding S, s and U antigens, are responsible for the S-s-U+^{var} phenotype: P2 and NY alleles. All GYPB(P2) and GYPB(NY) alleles analyzed so far carry the GYPB*S polymorphism. Arnaud et al. (2013) described a novel GYPB mutation that induces a genotyping error or failure for GYPB*S with the BioArray HEA v1.2 BeadChipTM kit: the discovery of two patients genotyped as heterozygous for GYPB(P2) and homozygous for GYPB*s with that kit led to the identification of a novel c.137-8C>T mutation - associated with the P2 allele - in their DNA.

Aims: To present a case of c.137-8C>T mutation in a GYPB*S allele, not associated to the GYPB(P2) allele.

Methods: Immucor BioArray HEA v1.2 BeadChipTM kit. Grifols fully automated serological test (DG Gel) performed for MNSs phenotyping. Bidirectional Sanger sequencing of the GYPB gene, performed on a 3730xl DNA analyzer using BigDye Terminators v.3.1 (ThermoFisher-Applied Biosystems, Foster City, CA).

Results: During a blood donor routine genotyping, we repeatedly had an indeterminate call (IC) for GYPB*S with the BioArray HEA v1.2 BeadChip kit. By sequencing DNA, the molecular presence of cytosine and thymine was found at the polymorphic site rs7683365 (c.143T/C), predictive of S(+) and s(+) antigens. Moreover, a heterozygous polymorphism c.137-8C/T was detected, which is located in the HEA probe binding region and is known to induce a failure in genotyping with the BeadChipTM technology. No evidence was found for c.270+5G/T polymorphism, which is characteristic of P2 allele. Serological testing for S and s with Grifols sera and automatic system showed a weak expression of S (+/+++++) and a normal expression of s (+++/+++++).

Summary / Conclusions: c.137-8C>T GYPB mutation is known to affect the detection of the S antigen (GYPB c.143T) by the BioArray HEA v1.2 BeadChip kit, resulting in the IC calls we have observed. Of note, this has allowed to discover the first case where this gene alteration is not associated with the presence of a P2 allele. Based on our serological data, this mutation causes a weak expression of S antigen when it's not associated to other GYPB polymorphisms.

P589 | Development of a multiplex PCR-SNaPshot assay to enhance RHD variants detectionE S Rodrigues^{1,2}, C M Ribeiro^{1,2}, L B Moreira^{1,2}, F L Santos¹, A R Zanelli¹, T B Cutter¹, R R Calado^{1,2}, D T Covas^{1,2}, S Kashima^{1,2}¹Hemocentro de Ribeirão Preto, ²Faculdade de Medicina de Ribeirão Preto (FMRP), Universidade de São Paulo (USP), Ribeirão Preto, Brazil

Background: Despite limitations, hemagglutination tests are widely used for immunohematological Rh blood group system diagnosis to ensure transfusion-safe medicine. Detection of weak and partial RhD variants is a common concern in blood bank services, as hemagglutination reveals the presence of an RhD variant but does not specify which variant is present. This challenge has been addressed with molecular assays, which enable the accurate characterization of samples with RhD variants. However, the methodology for RhD genotyping is either laborious or requires a substantial financial investment.

Aims: This work aimed to develop a molecular assay for the simultaneous detection of the weak RHD and partial RHD variants most prevalent in the Southeast Brazilian population.

Methods: A multiplex PCR reaction was standardized for specific amplification of exons 1, 4, 6, 7, 8, and 9 of the RHD gene. The amplified DNA product was then evaluated using the SNaPshot fragment analysis technology. Assay development involved careful PCR standardization steps, capillary electrophoresis extension, calibration, and fluorescence analysis to ensure accuracy in identifying RhD variants.

Results: Our results demonstrated specific amplification of the RHD gene, with an established multiplex PCR reaction that allows the efficient amplification of exons 4, 6, 7, 8, and exons 1 and 9 under the same cycling conditions. The SNaPshot reaction allowed the identification of samples with the variants RHD* weak type 1 (c.809 T>G), RHD* weak type 2 (c.1154 G>C), RHD* weak type 3 (c.8C>G), and weak/partial RHD* type 4 (c.602 C>G). Furthermore, the SNaPshot reaction allowed the simultaneous identification of weak RHD* type 15 (c.845 G>A) and other single nucleotide variants (SNVs) responsible for the classification of partial RhD variants, such as c.819 G>A characteristic of the DIII and DAR variants, c.1025 T>C present in DAR, DIV, DBT and RHD* weak type 29 and c.1136 C>T found in the DAU variant. Validation of the assay corroborated with previous results genotyped by PCR-Allele Specific and facilitated the complete classification of RhD variants.

Summary / Conclusions: SNaPshot assay developed in this study emerges as a valuable tool for the precise elucidation of RHD gene variants and contributes to the advancements in hematological and immunohematological research.

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P590 | In doubt, genotype—a case reportI Moreira¹, L Costa¹, S Silva¹, D Gonçalves¹, Santos², A Matos¹, B Alves¹, Soares¹, J Baldaque¹, H Sousa¹, T Ventura¹, J Martins¹, C Monteiro¹, A M Leite¹, M Carvalho¹, C Koch¹¹Immunohemotherapy, Unidade Local de Saúde de São João, Porto,²Immunohemotherapy, Unidade Local de Saúde Trás-os-Montes e Alto Douro, Vila Real, Portugal

Background: Neonatal red blood cells are immature and present a weak ABO expression that sometimes results in a mixed field agglutination pattern. Accurate ABO system identification is crucial for blood transfusion and transplantation procedures, namely in newborns (NB) where ABO discrepancy is warranted. Severe anemia after birth can result from a wide variety of causes, such as fetomaternal hemorrhage, hemolytic disease of the NB, sepsis or marrow failure.

Aims: Our aim was to investigate a case of an ABO mixed field agglutination pattern in a premature NB, with fetal anemia who needed an emergency-release blood transfusion at birth.

Methods: We reviewed the clinical records of a premature NB, who eventually received multiple red blood cell transfusions.

Results: Preterm female NB, eutocic delivery at 25+6 weeks' gestation (Apgar score: 4/6/9 after neonatal resuscitation). NB started surfactant therapy (133 mg/kg) and prophylactic ampicillin+gentamicin+fluconazole due to infectious risk (intraamniotic maternal infection plus maternal fever). Transfontanelar ultrasound revealed grade II intraventricular hemorrhage and reversed diastolic flow. She developed hypotension, needing dopaminergic support, anemia (hemoglobin [Hb]: 11.2 g/dL) and neonatal jaundice phototherapy-treated. At day 4 of life (D4), NB was diagnosed with a large patent ductus arteriosus and dilation of the left heart. Due to anemia of prematurity (minimum Hb value: 10.6 g/dL), NB was transfused with 4 packed red blood cells units (pRBC) (10 mL/kg) (Table 1) at D1, D11, D21 and D44. At the time of the first transfusion, mother's pretransfusion testing (PTT) revealed blood group A Rh(D)+ and negative antibody screening (IAT). However, NB's PTT confirmed the presence of a double population with antiserum-A. Considering the emergency of the initial transfusion, due to severe anemia, suspicion of fetomaternal hemorrhage along with the uncertainties in the PTT regarding the NB blood group, transfusion with group O Rh(D) + irradiated pRBC was performed. All pRBC transfusions were compatible with mother and daughter's samples and took place without adverse reactions. In a further NB's sample, the amount of cells that reacted with anti-A serum appeared to be smaller, thus it was hypothesized to be a B Rh(D)+ blood group. The expected discrepancy between serum and cellular levels in NB and subsequent transfusions with group O Rh(D)+ made it impossible to determine the blood group with certainty. Therefore, a sample to genotyping was obtained to clarify. Genotype determined by PCR-sequence-specific primer revealed: ABO*B.01/ABO*A2.01; RHD*01(DD); RHCE*01 (RHCE*ce); RHCE*02 (RHCE*Ce). Inferred phenotype was: A2B; D+; C+; c+; E-; e+. NB was discharged at D49.

Summary / Conclusions: Timely recognition of group A subtypes allows better transfusion outcomes, avoiding unnecessary exposure to A1 cells. In our report, considering the dissonant results in PTT and concerns about the safety and efficacy of blood transfusions for this fragile young patient, ABO genotyping proved to be a highly valuable source complementing evidence from serologic testing, thus confirming the suspicion of a weak subgroup.

P590 - Table 1: Phenotype of transfused packed red blood cell units.

Admission day	Pre-transfusion Hb (g/dL)	pRBC blood group	pRBC phenotype
D1	11.2	O Rh(D)+	Ccee;K-;M+;N-;S-;s+;Fya-;Fyb+;Jka+;Jkb+
D11	10.6	O Rh(D)+	Ccee;K-;M+;N-;S-;s+;Fya-;Fyb+;Jka+;Jkb+
D21	9.9	O Rh(D)+	CCee;K-;M+;N-;S-;s+;Fya-;Fyb+;Jka-;Jkb+
D44	8.3	O Rh(D)-	C-;c+;E-;e+;K-;Cw-

P591 | The frequency of RHD variants in a population of blood donors in North Eastern Algeria

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Background: The RH system is the most polymorphic of the human erythrocyte blood group systems. This system is of major clinical importance due to the high degree of immunogenicity of its antigens. In the field of blood transfusion, the correct determination of the D phenotype is of paramount importance to avoid the risk of alloimmunization.

Aims: Due to the increase in inter-population genetic variability of the RhD gene, it is of fundamental importance to establish the molecular polymorphism of the *RHD* locus in individuals from our region with a view to implementing an immunohematological approach based on molecular studies.

Methods: Molecular analysis of low and partial RHD variants carried out on a sample of 102 donors who showed a difference in reactivity with anti D. DNA was extracted by the QuickGene-Mini80 kit S (DB-S) system. For the molecular analysis by PCR-SSP we used the Kit (RBC-Ready CDE - SSP Inno-train) for the partial RHD variants and the Kit (RBC-Ready WEAK -D- SSP Inno-train) for Weak RHD variants. All samples were analyzed by both kits.

Results: In our population survey of blood donors from North East Algeria, we identified a total of 10 weak D alleles; type 4.0 SNP (602C/G and 819G/A), type 4.2 SNP (602C/G, 1025T/C and 957G/A), type 1 SNP (809T/G), type 2 (1154 G/C), type 3 SNP (8C/G), type 5 SNP (446 C/A) and 2 partial alleles; type DAU SNP (1136C/T) and type VII SNP (329T/C). Among the weak and partial D alleles found in our study, type 4 was the most common, with a frequency of 78.26%, followed by type 2 (7.09%), type 1 and 3 (4.87%) and the partial D type DAU (2.43%).

Summary / Conclusions: The variant RHD alleles identified in our study potentially encode altered or partial RhD antigens which could pose difficulties during routine serological typing, hence the interest of an essential molecular analysis to assess the potential risk of alloimmunization.

P592 | Gender-inclusive blood group sequencing—a novel pseudoautosomal and X-chromosome ploidy estimation approach for sex chromosome blood group prediction with a next generation sequencing enrichment panel

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Background: Sex chromosome ploidy, often determined by a donor's gender, or in some cases, the sex assigned to them at birth (SAAB), is often used as part of sample identity check in high-throughput genomic assays, and is critical for precise interpretation of phenotypes encoded in the X and Y chromosomes. For blood typing applications, this includes the XG and XK blood group systems, in addition to the GATA1 transcription factor, which are encoded in the human X chromosome and/or pseudoautosomal region. However, gender and SAAB are not only inexact proxies for sex chromosome ploidy, but their use raises important inclusivity considerations. Asking SAAB to gender-diverse donors is not recommended unless necessary because of the potential for psychological harm. Best practice recommends asking only the information needed for clinical decision-making. Since no blood group antigens are currently known to be encoded by the Y chromosome, we designed a new Next Generation Sequencing (NGS) blood group enrichment assay for future donor and patient typing whereby probe targets were limited to the relevant X chromosome and pseudoautosomal regions. To improve existing data QC and support gender-inclusive practices, we developed an objective analytic strategy that is independent of self-declared gender/SAAB information.

Aims: To develop an accurate pseudoautosomal and X-chromosome quantification tool, coupled with a custom NGS blood group enrichment panel, for XG blood group prediction in a gender-diverse population.

Methods: A sequencing enrichment 80mer DNA probe panel was designed to capture all blood group relevant genomic regions, including all exons, flanking sequences, and non-coding regulatory sites for two XG blood group genes: *CD99* encoded in the pseudoautosomal region, and *XG*, which straddles the pseudoautosomal/X chromosome boundary.

P592 - Table 1: *Needs phasing, CN = Copy Number, PA = PseudoAutosomal, hom = homozygous, het = heterozygous, var = variant, ref = reference

samples count	CN chrX non-PA PA CN	rs311103; Predicted CD99	Predicted Xga	Known Xga phenotype
1	~2 2	het;high	+	+
2	~1 2	het;high	*	+
1	~2 2	het;high	+	-
2	~1 2	hom var;low	-	-
3	~1 2	hom ref; high	+	+
2	~1 2	het; high	*	-
1	~2 2	hom ref; high	+	+
2	~2 2	hom var; low	-	-

Genomic DNA of 18 calibration samples (with known diverse sex chromosome aneuploidies) and 14 well-characterized test samples (Table 1) with known Xg^a phenotype, were subjected to enrichment library preparation and short-read NGS. The calibration samples included known XX, XY, XO, XXXXY, XXXX, XXY, XYY, and XYY chromosome ploidies. Genome Analysis ToolKit and mosdepth were used for read alignment, variant calling, and coverage estimation. A linear regression model was derived from the coverage data of the aneuploid set and their observed ploidy to produce a sex-chromosome ploidy estimator.

Results: Leave-one-out cross validation of our ploidy estimation tool provided an $R^2 = 0.992$ and a mean standard error = 0.07 between observed and predicted values. No major alterations were identified in the XG, CD99, XK and GATA1 genes in this sample set. We predicted Xg(a+) Xg(a-), CD99-high and CD99-low phenotypes in the 14 testing samples, using our sex chromosome ploidy estimation and the GATA-binding regulatory site (rs311103) zygosity (Table 1). Xg^a phenotype prediction accuracy and precision were 0.9 and 0.75 respectively. Four samples were appropriately flagged for phasing ambiguity. One discordance was further investigated with Whole Genome Sequencing.

Summary / Conclusions: We present an analytic tool for precise quantification of blood-group related sex chromosome regions in a custom NGS enrichment panel, and its application for XG blood group phenotype prediction. This novel approach supports gender inclusive practices for donors and patients.

P593 | ABO system—from phenotype to genotype

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Background: The main application of the ABO system is blood transfusion, and the development of molecular biology techniques has made it possible to remove ambiguities of interpretation in certain situations where serological techniques have their limits, and to use ABO alleles as genetic markers in the study and understanding of human genetic diversity.

Aims: Determine the allelic and genotypic frequencies of the ABO system in the North-East Algerian population, enabling its anthropogenetic characterization, and estimate the genetic distances with other populations.

Methods: The phenotypic study involved 10208 blood donors using plate and gel card techniques. ABO genotyping was carried out both to remove certain ambiguities of interpretation with serological techniques and to determine the ABO genotypes present in our population. SPSS V.20 and R software were used for statistical comparison with other populations.

Results: the order of ABO allele frequencies is O>A>B with respectively 0.6820, 0.2112, 0.1068. Whereas the order of frequency of ABO genotypes determined by molecular biology is: OO > AO > BO > AA > AB > BB with respectively: 44.44% > 23.08% > 15.38% > 10.26% > 5.13% > 1.71%. The frequencies of the ABO, A1, A2, B, O1 and O2 alleles are respectively: 16.67%, 6.41%, 11.97%, 58.12%, 5.55%. Estimation of genetic distances using Principal Component Analysis and hierarchical ascending classification revealed that they are not always well correlated with geographical distances, but influenced by other factors generating more mixing with certain populations than with others.

Summary / Conclusions: ABO blood grouping is one of the easiest tests to perform in medical biology, but its interpretation is sometimes very tricky. Molecular biology has enabled us to better understand the basis of genetic polymorphism in this system.

P594 | Molecular genetic analysis of two novel A allele to cause Ax phenotype in Chinese

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Background: Mutations of ABO gene may cause the dysfunction of ABO glycosyltransferase (GT) that can result in weak ABO phenotypes.

Aims: Here, we identified two novel weak ABO subgroup alleles and explored the mechanism that caused Ax phenotype.

Methods: The ABO phenotyping and genotyping were performed by serological studies and direct DNA sequencing of ABO gene. The role of the mutations was evaluated by 3D model, predicting protein structure changes, and *in vitro* expression assay. The total glycosyltransferase transfer capacity in supernatant of transfected cells was examined.

Results: The results of serological showed the subject RJ23 and RJ52 both were Ax phenotypes. The novel A alleles, *Avar-1* and *Avar-2* were identified according to the gene analysis. Both *Avar-1* and *Avar-2* harbored recombinant heterozygous alleles, specifically A2.05 and O.01.02. These alleles showcased substitutions at positions c.106G>T, c.189C>T, c.220C>T, and c.1009A>G in their respective exons. It is worth noting that the crossing-over regions of these two alleles differed from each other. *In vitro* expression study showed that GTA mutant impaired H to A antigen conversion, and the mutant did not affect the production of GTA though the Western bolt. *In silico* analysis showed that GTA mutant may change the local conformation and the stability of GT.

Summary / Conclusions: The *Avar-1* and *Avar-2* alleles were identified, which could cause the Ax phenotype through changing the local conformation and reducing stability of the GTA.

P595 | How genotyping may help in ABO discrepancies

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Background: ABO discrepancies can be challenging, especially in patients with conditions that modify antibody production. Such difficulties appear in cases with a history of transfusions where accurate ABO grouping data is unavailable. Moreover, monoclonal antibody therapies can modify the surface of red blood cells (RBCs), making serological tests for ABO determination more complicated.

Aims: RBC genotyping for ABO antigens could be a valuable tool in certain scenarios. It can assist in accurate characterization of a patient's blood group, enabling the selection of the most appropriate ABO match for transfusion. We demonstrate a case where the genotyping was crucial for the choose of the right blood to transfuse.

Methods: 66 years old, male patient, with Acute Erythroleukemia, Refractory Anemia with Excess Blasts (AREB). The ABO testing in the Hospital showed a discrepancy between the direct and reverse tests. The patient was transfused the week before with two O Rh negative RBC concentrates. The patient's sample was sent to Lisbon Immunohematology Reference Laboratory for further studies. The ABO testing was performed by tube (Lorne and Immucor reagents), with RBCs treated with modified bromelain (ID-diluent 1 Biorad), and also by the Biorad card and the Ortho BioVue card. Genotyping was carried out by SSP-PCR (Sequence-Specific Primer Polymerase Chain Reaction) using the kit ABO Type from BAG Diagnostics GmbH.

Results:

P595 - Table: Results.

Anti-A	Anti-B	Anti-AB	Anti-A1	Anti-H
MF*	0	3/4	0	1/2
Direct test				
Auto control	Donor RBC A1	Donor RBC B	Donor RBC 0	
0	0	w	0	
Reverse test				

Table 1) and 2) ABO Grouping in tube by direct and reverse tests.

*Mixed field

The results for the genotyping were: genotype **ABO*O.01.01/A1.01** and the predicted phenotype: **O01(O1)/A1; A1.**

Mixed field (double population) with Anti-A direct test suggests that there are two distinct populations of red blood cells (RBCs) in the sample, one reacting to the Anti-A antibody. This could be due to either a recent transfusion with a different ABO blood group, or with a sub-group of A (like A3, which might not react strongly with some Anti-A reagents). Also, a slight reaction on Anti-B Antibody in reverse typing was observed indicating the presence of Anti-B antibodies in the patient's serum. The use of bromelain didn't provide different results.

Summary / Conclusions: Mixed-field agglutination in the anti-A direct test and weak anti-B reaction in the reverse test was solved by the use of ABO genotyping at Lisbon Immunohematology Reference Laboratory (LIRL). It was identified the A1 subgroup, correctly determining the patient's ABO group. A better Patient Blood Management (PBM) was attained allowing the use of further O Rh negative RBC for the right patient on the right moment.

P596 | Sickle cell disease and GATA promoter region mutation—blood banks challenges after population diversification

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Background: Red blood cell transfusion (RBCt) is challenging in sickle cell disease (SCD). The Fy (a-b-) phenotype, mostly found in African ancestry, is rare in European ancestry. It occurs due to a GATA promoter region mutation (GPRM) upstream of the FY allele.

Aims: Our aim was to describe 2 cases of GPRM carriers.

Methods: We reviewed the clinical records of 2 patients with SCD who presented GPRM.

Results:

Case #1:

A 2-year-old Black African male referred to our hospital in 2021 due to SCD; already transfused at 7-months-old (M). He had had malaria.

In 2022, the patient presented hemoglobin (Hb) S: 51%, baseline Hb 7-8 g/dL and no need of RBCt. Pretransfusion testing (PTT) revealed blood group O Rh(D) positive ccee K negative, negative antibody screening (IAT) and negative direct antiglobulin test (DAT). He was transfused with 3 packed red blood cell unit (pRBC) at our hospital (10mL/kg)-Table 1. Every RBCt underwent without adverse reactions. In 2023 his blood group genotype was determined by SSP-PCR: RHD*01 (DD); RHCE*01 (RHCE*ce); RHCE*01 (RHCE*ce); FY*02N.02 (GATA box promoter: M/M); inferred phenotype: D+; C-; c+; E-; e+; Fya-; Fyb-.

Case #2:

A 24-year-old Black African male, with compound heterozygosity for HbS and alpha-thalassemia (baseline Hb: 6-7 g/dL) complicated by chronic kidney disease (stage 5d) and interrelated comorbidities. He was recurrently admitted to our hospital due to severe vaso-occlusive SCD crisis. PTT revealed blood group O Rh(D) positive ccee K negative, negative IAT; first DAT positive, but negative in later determinations. To date, patient was transfused with 182 pRBC (Table 2) and no adverse reactions, maintaining negative IAT. In 2016 the following genotype was determined by PCR-SSP: RHD: DAU 0,1,2,3; RHCE: RHc/RHc; RHe/RHe; KEL2/KEL2; JKA/JKB; FYnull01; promotor Duffy (GATA box promoter: M/M); MNS2; MNS4; inferred phenotype: C-; c+; E-; e+; K-; k+; JKa+; Jkb+; Fya-; Fyb-; M-; N+; S-; s+.

Summary / Conclusions: SCD patients are expected to immunize lifelong due to multiple transfusions and blood group mismatching between donors of European descent and patients of African descent represents a major risk. Despite GPRM mutation prevents expression of the gene in RBC, anti-Fyb development rarely occurs, due to Fyb's antigen expression on non-RBC. This explains why these patients were transfused with Fyb+ pRBC with no adverse outcome. Due to stock limitations in European blood banks, this is also comforting data. These cases reflect that sometimes, due to RBC phenotype limitations and urgent need of RBCt, blood banks must make timely wise decisions and genotyping informs about a larger number of blood groups that can be inaccessible by serology.

P596 - Table 1: Characterization of patient 1 transfusion episodes. Caption: URI: upper respiratory tract infection.

Age	Reason for admission	Pre-transfusion Hb (g/dL)	Transfused pRBC phenotype
13 M (2022)	COVID-19 infection	4.3	ORh(D)+;Ccee;K-;Cw-
20 M (2023)	URI	4.7	ORh(D)-;ccee;K-;Cw-; N-;Fyb-;s-;M+; Fya+;Jkb+; JKa+; S+
24 M (2023)	URI	5.6	ORh(D)-;ccee;K-;Cw-; Jkb+;Fyb+;s-;M+; S+;Jka+;N+;Fya-

P596 - Table 2: Phenotype of some of the transfused packed red blood cell unit.

Transfusion's date	Exemples of transfused pRBC phenotype
Before 2015	CcEe;K-;M+;N+;S-;s+;Fya+;Fyb+;Jka-;Jkb+ Ccee;K-;M+;N+;S-;s+;Fya+;Fyb-;Jka-;Jkb+ Ccee;K-;S-;s+;Fya+;Fyb-;Jka-;Jkb+
After 2016	ccee;K+;Jka+;Jkb+;Fya-;Fyb+;M-;N+;S-;s+ ccee;K+;Jka+;Jkb+;Fya-;Fyb+;M+;N+;S-;s+ ccee K- Jka+ Jkb+ Fya+ Fyb+ M- N+ S- s+ ccee K- Jka+ Jkb+ Fya+ Fyb- M- N+ S- s+

P597 | The identification methods for antigen D

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Background: The complex treatment of patients with surgical, hematological, oncological, and obstetric profiles involves transfusions of blood components. The RH antigen system is one of the most significant after the ABO system. In order to avoid alloimmune conflicts, it is crucial to correctly identify antigen D in donor's and recipient's blood. Antigen D can be well identified by salt agglutination, but there are some variants of antigen D that are difficult to identify serologically. To ensure the safety of transfusions, additional research methods are necessary.

Aims: Evaluate the efficiency of using serological methods and genotyping to determine antigen D.

Methods: Material for the study was 3,039 samples obtained from primary donors of the Federal State Budget Institution "Russian Research Institute of Hematology and Transfusion of the Federal Medical and Biological Agency". Typing of D, C, c, E, e antigens were carried out using a gel agglutination test using the analyser IH-1000 BioRad (USA). The estimated range of erythrocyte agglutination was between 1+ and 4+. Weak agglutination (from 1+ to 3+) in the indirect antiglobulin test with the ID-DiaClon anti-DD-IgG reagent (BioRad, USA) were the criteria for genotyping. The RHD genotype was determined using SSP-PCR RBC - FluoGene D-weak (Inno-train Diagnostik). This determination was compared to those of RhD- and RhCcEe-phenotyping.

Results: In serological determination of antigen D, ambiguous results were received from 19 donors. Two donors showed weak agglutination in DiaClon ABO/D ID cards (reaction strength 2+), while 17 did not show any agglutination. All 19 samples in indirect antiglobulin test demonstrated agglutination with a reaction strength range of 1+ to 3+. In five specimens, the allele-specific PCR method identified D weak type 1 with Ccee (n = 3), CCee (n = 1), and ccee (n = 1) phenotypes. In one case, D weak type 2 with phenotype ccEe was established. In 11 specimens, D weak type 3 with Ccee (n = 8), CCee (n = 2), and ccee (n = 1) phenotypes were established. A normal antigen D allele has been identified in two specimens with the CCee

phenotype. Transfusions and pregnancy do not cause the production of anti-D-alloantibodies in individuals with D-weak types 1, 2 and 3. In our study, D-weak was present in 0.56 percent of the cases, with the majority of them being D-weak type 3. RHD*D weak 4, 4.0, 4.1, 4.2 (DAR), 5, 11, 14, 15, 17 and D partial DII, DIII, DIV, DV, DVI, DFR, DAR, DBT, etc. have not been identified.

Summary / Conclusions: The study revealed only three D weak types: D weak type 1, 2, and 3. The occurrence rate was 0.56 percent. There was a high prevalence of the D weak type 3 antigen. Genotyping techniques are advisable for complex cases of antigen D identification, as demonstrated by the results.

P598 | Detection of spontaneous Rh type changes as a sign of loss of heterozygosity in acute myeloid leukaemia may anticipate a clinical diagnosis—a case report

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Background: Mixed-field agglutination in serological Rh typing may be noted in iatrogenic chimerism (hematopoietic stem cell transplantation) or in non-iatrogenic settings, mainly regarding the D antigen. Acquired Rh antigen loss is mostly observed in patients with clonal myeloid diseases. Loss of heterozygosity (LOH) is a form of allelic imbalance by which a heterozygous somatic cell becomes homozygous because one of the two alleles is lost. LOH is frequent in acute myeloid leukemia (AML). Recurrent regions of LOH includes 1p (short arm of chromosome 1), where RHD/RHCE loci are located. Few reports have been published on weak expression of D antigen in serological RhD typing due to LOH in AML and even less on weak expression of other Rh antigens.

Aims: To present a very rare case of D and E antigens weakness due to LOH, preceding and suggesting the diagnosis of AML.

Methods: GM, caucasian female, 76 years old, was observed in San Donà di Piave Hospital (Venice district) because of asthenia due to previously unknown severe anemia. Blood units for transfusion were requested. In our laboratory was detected inconsistent results in serological RhD and RhE typing with different anti-D and anti-E reagents. The patient was provisionally considered as D variant and continued to be transfused as an outpatient, since no certain causes of the anemia were recognized. Further investigations were performed by molecular biology tests to detect any variants of these antigens. Immucor BioArray RHD/RHCE BeadChipTM Technology for D and E variants. RHD DNA testing performed for RHD genotyping: PCR-multiplex analysis for RHD exons 4 and 7, and the inactive RHD pseudo gene. Zygosity determination by hybrid Rhesus box detection. PCR-RFLP for RHD exon 8 c.1136C>T. Sequencing of exons 1 to 10. PCR for RHD exon 3 duplication.

Results: No D variants were detected. Subsequent investigation found the E typing was weak/variable as well as reduced signal for E marker. RBCs also phenotyped C-c+e+. Genotype for RHD was

requested. PCR-multiplex: RHD exons 4 and 7 were present, but with reduced intensity; negative for the inactive RHD pseudo gene; positive for the hybrid Rhesus box associated with deletion of RHD; negative for the RHD c.1136C>T change, slight reduction in intensity of PCR product was noted. Sequence results: No changes in RHD exons 1 to 10. Negative for RHD exon 3 duplication. Therefore the conclusions were: RHD hemizygote, with no RHD changes.

Summary / Conclusions: DNA testing performed found a reduced signal for RHD which suggests a mixed population with only a portion of cells harboring RHD. These results may be consistent with a history of bone marrow transplantation, possible clonal loss of RHD or with a somatic cell or natural chimera. Patient denied having a twin or a bone marrow transplantation, so we reported the results to her doctors who, in the meantime, had performed a bone biopsy. This confirmed the diagnosis of AML with normal karyotype and somatic cell chimera.

P599 | Detection of ABO*AW.06 ABO*O.01 blood group in routine blood transfusion compatibility tests

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Background: The clinical significance of blood group antigens relates to their ability to stimulate immune antibodies that can cause hemolysis and transfusion reactions. The most important antigens for safe transfusion are ABO and D (Rh), and typing of these antigens is routinely performed for patients awaiting transfusion and blood donors. Molecular typing of blood group genes in diagnostics allows the resolution of clinical problems especially weak expressed blood antigens that cannot be detected by hemagglutination. Determining the variant blood groups of individuals using molecular methods before transfusion is important for safe blood transfusion.

Aims: Herein we report a patient whose blood typing wasn't determined by serological method but determined ABO*AW.06 ABO*O.01 by molecular typing. In this study, we wanted to emphasize the importance of molecular typing in pre-transfusion tests.

Methods: To detect the presence of weak A antigen, thermal elution was performed on the patient's RBC. The eluate was tested with group A and O cells and was shown to have weak A antigen on erythrocytes. Secretory status was determined using the patient's saliva to detect the presence of soluble substances. The patient was found to be a non-secretor with detectable substances A and H in saliva. As a result of the results obtained in these serological reactions, it was thought that the patient's blood type might be A variant or a blood group similar to this serology. In order to determine this exactly, in addition to the studies, DNA was obtained from the patient and end-point fluorescence-based blood grouping test was performed with PCR-SSP using the molecular typing method.

Results: In this study, ABO forward-reverse incompatibility was observed in the first blood grouping of the patient, who we

determined to have a weak variant of blood group A. When a positive reaction was observed with anti-H lectin and a negative reaction with anti-A1 lectin, it was thought to be a variant of A, and further serological examinations including adsorption-elution tests and saliva tests were performed. In adsorption elution tests, the presence of A antigen was detected on the surface of erythrocytes. The patient was not a secretary. The presence of anti-A1 was observed in serum. The identified weak A phenotype was defined as the weak A genotype (ABO*AW.06 ABO*O.01), serologically similar to the A3 phenotype, but in molecular typing.

Summary / Conclusions: If ABO differences are found between forward and reverse typing in any donor or recipient, this should be reported to both laboratory personnel and physicians due to the high probability of poor expression of blood group antigens. Additional specific procedures such as molecular testing for mutations may be performed to differentiate weak subgroups. Blood groups determined using this method give the safest and most accurate results in safe transfusion.

P600 | **In silico investigations are employed to examine the theoretical structures of RhD and RhD mutation, which leads to weak D**

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Background: In silico analyses examine the theoretical protein structure and detect the distinctions between the RhD wildtype (WT) and the RhD mutation responsible for weak D. The objective of our study was to determine the theoretical structural distinctions among the three amino acids (top 3 mutations for weak D) of RhD, and to investigate how RhD mutations lead to weak D utilizing in silico analyses.

Aims: The objective of our study was to determine the theoretical structural distinctions among the three amino acids (top 3 mutations for weak D) of RhD, and to investigate how RhD mutations lead to weak D utilizing in silico analyses.

Methods: The term "secondary structure" refers to the local folding patterns of a protein or nucleic acid molecule. The physicochemical profile was assessed using the measurement of hydrophobicity and electrostatic potential. The structures underwent evaluation and validation using Ramachandran plot and ERRAT, ProSA-web in a sequential manner.

Results: The surface area of all proteins does not significantly vary due to differences in hydrophobicity and electrostatic potential. The hydrophobic region of RhD WT had a percentage of amino acids of 53.64%, while RhD Weak D had a percentage of 58.42%. The primary cause of RhD weak D is a mutation at position 270 in the sequence, resulting in a substitution of valine with glycine. The amino acid position of this mutation is located in the transmembrane region. In the case of weak D, this specific mutation has the highest frequency in terms of phenotype. The structures were assessed and confirmed by

Ramachandran plot and ERRAT, ProSA-web consecutively, with a respective accuracy of 99.16% and 99.52%, and a Z score of -6.75 for RhD MT and -6.59 for RhD Mutation causing weak D.

Summary / Conclusions: The physicochemical characteristics of Rh D wild-type and mutant proteins exhibited little differences. Through in silico investigations, we identify the specific mutation sites in the RhD protein located in the transmembrane region that result in a weakened D antigen. This weakened D antigen poses challenges in its detection using anti-D antibodies. To overcome this, we create precise structural models that closely resemble those acquired through experimental methods. A change in the amino acid sequence leads to a modification in the folding of the tertiary structure and the way it interacts with other proteins.

P601 | **CRISPR-CAS9 driven antigen conversion of clinically relevant blood group systems**

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Background: The common practice of blood transfusion entirely relies on blood donations from the population. Ensuring blood group compatibility between a donor and a recipient is paramount to prevent critical adverse reactions. Finding compatible blood can be challenging given the high diversity of blood group antigens, especially for chronically transfused patients at higher risk of alloimmunization owing to repeated exposures to foreign RBCs. In addition, due to the immunogenicity of the ABO blood group and the highly polymorphic nature of the Rhesus (Rh) system, they both remain of prime importance in transfusion medicine. Cultured red blood cells (cRBCs) may eventually provide an alternative for blood donations, at least in some circumstances. Combining cRBCs culture from stem cells with gene editing of relevant blood group systems could broaden transfusion accessibility by making antigen expression compatible with rare phenotypes, thus meeting the needs of more patients.

Aims: We aimed at evaluating the potential of using CRISPR-Cas9-mediated gene ablation to produce (1) regulatory-type Rh_{null} erythroid cells starting from Rh⁺ hematopoietic stem and progenitor cells (HSPCs), and (2) group O erythroid cells starting from group A HSPCs.

Methods: Starting from mobilized, erythroid-primed HSPCs, we used virus- and selection-free, CRISPR-Cas9-mediated knockouts to produce erythroid cells devoid of AB and Rh antigen.

Results: The approach yielded almost complete conversion of cRBCs (i.e., up to 96%) to O- and RhNull phenotypes, as determined by standard hemagglutination and flow cytometry analyses.

Summary / Conclusions: Combined with robust cRBC production protocols from HSPCs, these clinically relevant phenotypic changes could eventually expand the accessibility of blood transfusion for specific and unmet clinical needs.

P601-A | Biochemical characterization of blood type-related variants of ABCG2, antigen in the JR blood group system: R147W and S572R variants are deficient in the plasma membrane localization

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Background: Accumulating evidence suggests genetic associations between Jr^a-negative [Jr(a-)] blood type and mutations in human ATP-binding cassette transporter G2 (ABCG2, also called as *breast cancer resistance protein*; BCRP) gene. Whereas this transporter protein has been known as a membrane machinery that regulates the pharmacokinetics of its substrate drugs, ABCG2 reportedly serves as an antigen in the JR blood group system (ISBT number 032).

Aims: Previous studies reported some nonsynonymous single nucleotide variations in ABCG2 including c.439C>T (p.R147W, rs372192400) and c.1714A>C (p.S572R, rs200894058) in the context of association with Jr(a-) blood type. Based on this information, we herein focused on these two amino acid substitutions and biochemically investigated, using a polarized cell line suitable for the evaluation of the membrane-sorting activity of ABCG2, whether each ABCG2 mutation can disrupt plasma membrane localization of ABCG2, which would result in a cause for the Jr(a-) blood type.

Methods: Each variant of ABCG2 tagged with EGFP was transiently expressed in Madin-Darby canine kidney (MDCKII) cells. To examine the cellular expression and localization of transduced ABCG2 protein, immunoblotting and confocal microscopy were conducted, respectively. Also, we addressed a cryo-electron microscopy structure of human ABCG2 (previously published in PDB ID: 5Nj3) for further biochemical considerations.

Results: While ABCG2 wild-type was expressed as a matured N-linked glycoprotein, p.R147W and p.S572R variants were hardly matured and their protein levels were extremely lower compared with wild-type. In contrast to the wild-type, these two variants were not localized on the apical membrane of the MDCKII cells. The structural information of ABCG2 protein indicated that the R147 and S572 residues are localized on the edge of an intracellular helix and the kinked region between two embedded helices in the membrane bilayer, respectively. Also, there are ethnic differences in the allele frequency of these two ABCG2 variants although they are rare variants according to the latest information from dbSNP (accessed in February, 2024).

Summary / Conclusions: Our results demonstrated that p.R147W and p.S572R disrupted protein stability and plasma membrane localization of ABCG2. Given structural insights, these mutations may destabilize

protein structure via affecting helix structure in which they are contained. These findings will contribute to deeper understanding of JR blood group alleles.

Immunohaematology—rare donors

P602 | Identification of high-frequency antigen-negative blood donors using the Universal Blood Donor Typing and UK Biobank genotyping arrays

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Background: A High-Frequency Antigen (HFA) is defined as an antigen absent from the red blood cells of less than 1 in 1000 donors. Most HFA have limited antigenicity, but on rare occasions, a HFA-negative patient may form an alloantibody after transfusion or in pregnancy. To be able to supply compatible units, blood services make immense efforts to identify HFA-negative donors. Genotyping for clinically relevant blood groups will significantly simplify the identification of rare donors. The BGC has developed the Axiom Universal Blood Donor Typing (UBDT_PC1) and Axiom UK Biobank (UKBB_v2.2) arrays to genotype donors for human erythrocyte, platelet and leukocyte antigens (HEA, HPA and HLA, respectively).

Aims: To identify HFA-negative donors in the five main ancestry groups in an international multi-centre validation study using the UBDT_PC1 array and in the STRIDES NIHR BioResource (SNBR) study using the UKBB_v2.2 genotyping array.

Methods: DNA samples of 6946 donors were provided by blood services from AU, CA, GB, FI, NL, SA, and US, and were genotyped with the UBDT_PC1 array on a GeneTitan-MC instrument (Thermo Fisher) at the New York Blood Center and SANQUIN. A set of challenge samples enriched for HEA/HPA phenotypes lacking in the 6,946 samples was also selected and genotyped ($n = 333$), including 131 HFA-negative donors. The inferred phenotypes of 53 HEAs (MNS, RH, LU, KEL, FY, JK, DI, YT, SC, DO, CO, LW, CROM, KN and VEL) by the bloodTyper module of the AxiomTM Total Blood Typing Solution were compared to HEA types retrieved from electronic donor records. Finally, 82,000 DNA samples from NHS Blood and Transplant (NHSBT) donors, enrolled in the SNBR study, were genotyped with the UKBB_v2.2 array by the NHSBT.

Results: The dense genotyping results allowed for automated quality control of the samples, and 6,867/6,946 (98.9%) passed. Genotypes were used to infer ancestry, showing that 34.8% of the samples were from non-European (EUR) donors. 156 HFA-negative donors (4 U-, 8 Lu(b-), 36 k-, 1 Kp(b-), 8 Js(b-), 1 Di(b-), 10 Yt(a-), 2 Hy-, 8 Jo(a-), 17 Co(a-), 4 Kn(a-), 54 McC(a-), 3 Vel-) were identified in the 6,867 samples, and 99/156 were newly identified. The 156 HFA-negative donors were from African (AFR, $n = 72$), Admixed American (AMR,

$n = 10$), East Asian (EAS, $n = 1$), South Asian (SAS, $n = 3$), and EUR ($n = 70$) ancestry. No Wr(b-), Lw(a-) and Cr(a-) donors were identified, which is in keeping with these types being exceptionally rare. In the set of selected samples known to be HFA-negative, 126/131 were identified by the array, with the remainder being untyped for their respective HFA-negative antigen. Inspection of genotyping call plots for these five samples revealed that the issue could be rectified by an algorithmic adjustment. Ancestry inference showed that 3,817 of the first 21,444 SNBR samples were from non-EUR donors, with 3.0%, 3.1%, 1.6%, 9.1% being of AFR, AMR, EAS and SAS ancestry. 160 HFA-negative donors (24 Lu(b-), 27 k-, 3 Js(b-), 24 Yt(a-), 33 Co(a-), 8 Kn(a-), 40 McC(a-), 1 Vel-) were identified.

Summary / Conclusions: The Axiom™ Total Blood Typing Solution is capable of identifying HFA-negative donors, represented in the five major ancestry groups. To determine the specificity of the array, the newly identified HFA-negative donors are presently confirmed by an accredited test. This study illustrates how high-throughput genotyping brings benefits for identifying donors with extremely rare HEA types, including those who are negative for HFA types.

P603 | Abstract withdrawn

P604 | Rare donor programs and IgA deficiency defining the landscape

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Background: Immunoglobulin A deficiency (IgA def) is a common immunodeficiency. Absolute IgA def is generally defined as IgA levels $<0.05\text{mg/dL}$ and its incidence among populations ranges between 1:500 in Caucasians to 1:18,000 in Japanese. Such individuals, especially those with anti-IgA, have the potential for life-threatening anaphylactic reactions following transfusion. Providing algA components is a challenge for transfusion services. Rare donor program testing for algA def, the need and quantity of IgA def products provided is unknown.

Aims: To determine how many rare donor programs include IgA def as a rare phenotype, what method is used for screening and how many

IgA def donors are registered. To understand how IgA def is defined, how often IgA def products are requested and filled and what alternative products may be recommended for these patients.

Methods: The ISBT central office emailed a survey to 39 ISBT Working Party on Rare Donor members from 28 countries in January 2024. Responses were compiled and analyzed.

Results: Responses were received from 15 members from 12 countries. Eight programs in 7 countries reported that IgA def is considered a rare phenotype; 4 defined IgA def as $<0.05\text{mg/dL}$ while each of the other centers had criteria ranging from $<0.09 - 10\text{ mg/dL}$. Four programs are actively screening for IgA def, whilst another program had screened in the past and two programs recruit IgA def patients and/or their family members as blood donors. Screening methods include turbidimetric, nephelometric, ELISA and magnetic bead-based assays. Six programs maintain a frozen inventory of IgA def plasma, with 424 units (range 5-250 units) across all programs. Only 1 of these 6 programs classifies IgA def as $<0.05\text{mg/dL}$; this program has 250 frozen plasma units. In all, 380 donors are classified as IgA def according to the different program criteria. Only three centers reported screening for anti-IgA with one reporting that they do not distribute products as IgA def from donors with anti-IgA. In 2023, five centers provided 49 plasma, 42 platelet and 1 red cell unit from IgA def donors. In response to requests for IgA def platelets, 4 programs reported primarily supplying IgA def platelets while the others provided washed platelets. In response to requests for IgA def red cell units, while other programs reported the use of washed red cell units, 1 center's policy is to supply IgA def red cells.

Summary / Conclusions: Of the 15 survey respondents, 8 programs consider IgA def a rare phenotype. Only 4 programs are currently screening for IgA def; methodologies and criteria differ. Ninety-two IgA def blood products were issued across all programs in 2023. Programs that do not have IgA def donors or products reported providing washed products to patients with a history of anaphylaxis presumed to be due to IgA def; several require detailed patient history and clinical consultation. Two programs expressed interest in establishing a program to identify IgA def donors. Responses from additional programs surveyed are expected and when received, the data will be re-analyzed to include those results. Despite the limitations of this survey, the relatively low demand for IgA def products raises questions concerning the prevalence of algA def in different populations as well as the engagement of rare donor programs around transfusion support of patients with algA def.

P605 | McLeod phenotype and chronic granulomatous disease - the importance of transfusion management in these patients with high risk of alloimmunization

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P605 - Table 1: Results.

	P1	P2	P3
Age at McLeod phenotype diagnosis (years)	8	14	20
RBC alloantibodies	No	No	No
Autologous donations	No	Yes	No
RBC transfusion during Allo-HSCT	16	4	X
Platelet transfusion during Allo-HSCT	15	4	X
ABO and RhD	A positive	O positive	B positive
Antibody screening	Negative	Negative	Negative
Phenotype and Genotype	Kell (K- k-), Kp (a- b-) kk. KPA/KPB. JSB/JSB	Kell (K- k+w), Kp (a- b+) kk. KPB/KPB. JSB/JSB	Kell (K- k-), Kp (a- b-) K/k. KPB/KPB. JSB/JSB
Genetic studies	Deletion with absence of XK and CYBB genes	Deletion with absence of 9 genes, among them CYBB and XK	Deletion with absence of 4 genes: LANCL3, XK, CYBB and DYNLT3

Background: McLeod blood group phenotype is caused by the XK gene deletion on the short arm of the X chromosome (Xp21.1). This mutation results in the absence of Kx antigen, the low expression of Kell antigens and, consequently, the lack of Km antigen. In addition, these patients may develop muscular, cardiac and neurological late clinical manifestations, which are commonly known as McLeod neuroacanthocytosis syndrome. Some patients have larger deletions on the short arm of the X chromosome that affect not only the XK gene but also the CYBB gene, a phenomenon known as "contiguous gene syndrome". These patients with CYBB gene mutation suffer from chronic granulomatous disease (CGD). Many CGD patients need treatment due to the seriousness of their infectious and inflammatory complications. Allogeneic hematopoietic stem cell transplantation (allo-HSCT) is the only curative treatment for CGD. However, allo-HSCT is incredibly challenging in patients with CGD and McLeod phenotype due to the risk of alloimmunization and the complexity of finding compatible blood components.

Aims: To describe a rare clinical situation that requires multidisciplinary management and to determine the importance of agreed transfusion strategies during HSCT follow-up to avoid alloimmunization.

Methods: Three patients with McLeod phenotype and CGD are presented (P1, P2 and P3). Immunohematology techniques and genetic studies are performed to collect and analyze results.

Results: The following table shows the results of two patients with McLeod phenotype and CGD who received an allo-HSCT (P1 and P2). P1 died during the course of transplantation and P2 presented complete chimerism on the day +80. P3 has not received allo-HSCT and is currently being followed up in another center.

Summary / Conclusions: Allo-HSCT in CGD and McLeod phenotype patients is recommended at an early age and requires the coordination and participation of several medical specialties, particularly the Transfusion Service. The high transfusion requirement during allo-HSCT carries the risk of developing clinically significant alloantibodies (anti-Kx and anti-Km). None of our patients developed these antibodies, which can be attributed to their immunosuppressed state. It is important to highlight that the probability of finding blood components with

McLeod phenotype is low and there are currently no available donors or compatible components in Spain. Therefore, autologous donations should be considered. Moreover, even though allo-HSCT is a curative treatment for CGD and McLeod phenotype, there is a risk of developing neuropsychiatric, cardiological and muscular manifestations afterwards. In conclusion, international collaboration between Transfusion Services and Transfusion Centers is essential to address the transfusion requirements of these patients and to share action protocols.

P606 | A high-throughput one-step method for the screening of IgA deficiency in blood donors

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Background: IgA deficiency is the most common immunodeficiency. Some IgA-deficient individuals have developed anti-IgA which may cause severe allergic transfusion reactions and should thus be transfused with IgA-deficient blood products. Although red cell concentrates and platelets can be washed to remove IgA, IgA-deficient plasma can only be obtained from IgA-deficient donors. Therefore, blood services must maintain a registry of IgA-deficient donors to provide this product upon request. Several screening methods to identify IgA-deficient donors mostly based on ELISA exist but, while being very reliable, they are also time-consuming. Typically, sample preparation and loading in microplates take about 7 hours for 500-700 samples respectively, for one technician. The ELISA technique itself requires about 4 hours to be completed.

Aims: Improve the efficiency of screening.

Methods: We set up a fluorescent-based competition assay requiring only one incubation step. The workload for sample distribution in microplates is considerably reduced by using automated dispenser commonly

used in blood services or hospital testing laboratories to distribute plasma samples (50 ml) from the leftover tubes used for ABO/Rh testing, in IgA-precoated and dried 96-well microplates. The microplates can then be frozen to prevent plasma spillage (if shipment to another facility is required) or used immediately. For testing, anti-human IgA-fluorescent conjugate is added directly to each well. The plates are incubated for one hour at 37°C, washed, emptied, dried and read on a fluorometer.

Results: The anti-human IgA-fluorescent conjugate is neutralized by the IgA contained in normal plasma, resulting in background fluorescence. In contrast, plasma from IgA-deficient donors does not interfere with the binding of anti-human IgA to IgA-coated wells, yielding high fluorescence and allowing the easy identification of IgA deficient donors. The procedure is completed within 2 hours, compared to more than 12 hours using a standard manual ELISA procedure for the same number of samples. Using this method, we tested 105 335 samples (9 months period) and identified 140 IgA-deficient donors (frequency of 1/752). These samples were retested using our in-house cytometry-based bead assay for preliminary confirmation of the IgA deficiency status. All samples were IgA-negative, showing the specificity of the screening method.

Summary / Conclusions: We have developed a cost-effective and efficient method for the screening of thousands of blood donors for IgA deficiency.

P607 | Prevalence and specificity of alloantibodies against high frequency red cell antigens in Oman—a national reference laboratory experience

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Background: Diagnosis and management of patients with antibodies against high-frequency antigens (HFAs) pose significant challenges in transfusion medicine, particularly when established rare donor registries and frozen blood inventories are lacking.

Aims: The study was conducted in Oman aiming to estimate the prevalence and specificity of antibodies against HFAs, analyze patients demographics and clinical profiles, and study the phenotypes of rare donors.

Methods: This is a retrospective descriptive study conducted in the central blood bank, which also has the main reference laboratory in the country. All referred cases from the 1st of January 2017 to 31st of December 2023 were manually screened and cases of antibodies against HFAs are collected.

Results: We identified 31 out of 6016 referred patients (0.51%) with antibodies against 14 different HFAs or variants of Rh antigens necessitating rare blood. The median age of these patients was 33.5 years, with females comprising 90% of cases. More than half of the patients were pregnant, and 13% had sickle cell disease. The most commonly encountered antibodies were anti-U and anti-k, constituting 39% of cases. Six

pregnant patients had anti-U or Glycoprotein B-related antibodies with anti-U-like specificity, and no cases of hemolytic disease of fetus and newborn (HDFN) were observed. Six other patients had anti-k antibodies. One patient was initially referred with a delayed hemolytic transfusion reaction after receiving k positive red cells and her next pregnancy was complicated by mild HDFN. Another patient with declining anti-k antibody titer required emergency transfusion using k-positive blood under IVIG coverage with no hemolytic reactions observed. Four patients from the same governorate with three being from the same tribe had the rare P1K phenotype with anti-P antibody and two -whom are second degree relatives- had additional anti-PX2 antibody. Interestingly, both of the two pregnant patients with P1k phenotype had successful pregnancies. A patient with anti-Jsb had a successful pregnancy outcome after multiple intra uterine fetal deaths due to HDFN. This was achieved by IVIG, steroids and multiple intrauterine transfusions using maternal blood and imported foreign blood. Patient's plasma was used for random cross matching with more than 2000 units including her family members. However, no compatible blood was found. Herein we also report the pregnancy outcome of the first and only reported case of antibody against the novel Luom antigen who had uneventful pregnancy with no clinical or serological evidence of HDFN. Through mass donor screening and affected patients' families screening, 89 rare donors were identified including Bombay (Oh), r'r', and r''r'' phenotypes and antigen negative for k, Kpb, Jk(a-b-), Lub, Yta and (S-,s-). Yet, no Rh null or P1K phenotype and no Lan or Jsb negative donors are found.

Summary / Conclusions: In Oman, numerous cases of antibodies against 14 different HFAs are observed. Despite our geographical proximity to neighboring populations, our population has a distinct profile of antibodies against HFAs. Realizing such finding is essential in order to have a personalized and efficient healthcare planning. Timely access to compatible rare blood is crucial to achieve good patient outcomes and this can be attained through a well-established national rare donor registry and a local frozen blood inventory. Undoubtedly, collaboration with regional and international rare donor panels is indispensable.

P608 | Managing a frozen rare blood inventory—a survey of American rare donor program member facilities

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Background: The American Rare Donor Program (ARDP) is made up of 90 member facilities, of which 76 are blood collectors. Many manage an inventory of rare red blood cell (RBC) units. In 2023, a vendor of an instrument used widely for deglycerolizing thawed red cell units announced their plan to retire the instrument. The plan for a replacement for this instrument is unclear. The ARDP gathered data on the inventory of rare red cell units in the inventory of US members and the instrumentation and capabilities of these facilities to manage this inventory.

Aims: This study aimed to determine the number of rare RBC units stored in ARDP member facilities and what methods are in place to thaw and deglycerolize these units.

Methods: The ARDP emailed a survey to US member facilities on November 2nd, 2023. The survey instructed recipients to complete a spreadsheet with questions regarding frozen red cell inventory and methods used to wash blood products. Members submitted their responses through December 31st, 2023. The data were compiled and analyzed. American Red Cross member facilities were counted as one blood collector for purposes of data analysis.

Results: Responses were received from 31 facilities from 22 blood collectors including 10 American Red Cross facilities. The respondents reported a total of 11,783 frozen rare RBC units, with nearly half (47%) of these units being from two large multisite blood collectors. Of the 22 blood collectors, 17 (77%) utilize only the Terumo COBE 2991 cell processor. Four centers (18%) reported having an alternate procedure and dual inventory. Two centers reported having Haemonetics ACP 215 automated cell processors but had not validated or were unable to validate for deglycerolizing frozen RBC units. Eighty-one percent of the frozen rare RBC units across all facilities would require COBE 2991 for processing. Two centers (9%) reported having a manual procedure that could be used as an alternative to process frozen RBC units. Additionally, members noted the use of the cell processor to provide washed platelets for patients with IgA deficiency and history of anaphylaxis.

Summary / Conclusions: The US has a substantial inventory of frozen rare RBC units. Data gathered from 31 facilities associated with 22 blood collectors demonstrate that more than 80% of rare RBC units in inventory at these centers were collected to be processed on the COBE 2991 instrument. Most facilities do not have an alternative procedure for processing their frozen units. ARDP members expressed concern about the future state of the frozen rare red blood cell inventory when the COBE 2991 cell processors become unavailable. Additionally, the instrument is used by many centers to wash platelets to remove plasma components. Based on these findings, there is a need to explore alternatives not only for future processing of frozen red cells but for processing of the existing inventory of rare red cell units and for providing washed platelets.

P609 | Expanding rare blood donor identification—a cytometric screening approach

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Background: Rare blood donors are challenging to identify. Besides, the regulation of some countries prevent asking donors about

ethnic origin, making difficult to settle an effective algorithm to optimize the probability of finding a donor lacking certain high prevalence antigen. Frequencies for uncommon phenotypes vary from 0.2% to 0.01% or even more scarce. This rarity makes the analysis of individual samples inefficient. Only some high-tech molecular platforms have a real high throughput capability to study large number of samples for many rare alleles at the same time (Gassner, Transfus Med Rev, 2013). More traditional methods imply large volumes of difficult to obtain antisera and long working hours. This highlights the need of finding novel and more scalable ways to deal with this problem. Flow-cytometry can be used as an efficient way to detect characteristics within large populations of cells. Although unusual, it is feasible to use for routine donor phenotyping (Liwski, Vox Sang, 2023).

Aims: Test if flow cytometry could be used to detect rare blood groups in a mixed population of red blood cells (RBC) from 32 different donors. Validate the screening method to identify rare phenotypes in pooled samples. Ratify this methodology in real life and establish it in our routine labwork.

Methods: The uncommon (0.2%) Cellano negative (k-) and rare (0.02%) Rautenberg negative (Kpb-) phenotypes were chosen as candidate antigens to test our approach. Individual sample analysis would set a cut-off for the pool validation with known phenotype RBC. The data was presented as the median fluorescence intensity (MFI). A population $\approx 3.12\%$ below the cut-off MFI should be found in those pools bearing a negative sample among the other 31. The pooling was done mixing RBC from 32 ABO isotype blood donors per pool. A dilution from each pool was incubated with either anti-k (Ortho Clinical Diagnostics) or anti-Kpb (Bio-Rad). The staining process used a second antibody FITC-conjugated with anti-human IgG specificity (Jackson ImmunoResearch), and the acquisition by a FACSLytic flow cytometer (BD Biosciences). We would ratify the method in routine work during January 2024 using never phenotyped donors. Any pool with cells below cut-off, would be break down into 4 subpools of 8 samples

P609 - Table 1

Antigen of interest	Pooling Validation	% cells below cut-off
k	No k- (8 pools)	0.07%
	1 k- (8 pools)	3.15%
Kpb	No Kpb- (3 pools)	0.26%
	1 Kpb- (3 pools)	3.41%

P609 - Table 2

	Pools (total samples)	Expected rare donors	Rare donors identified	% cells below Cut-off
anti-k	49 (1.568)	2-3	3	3.2%
anti-Kpb	84 (2.688)	0-1	1	2.47%

searching the one of interest. Those 8 final samples were routine phenotyped. Any negative or weak expression sample found, would be genotype confirmed.

Results: The validation process was successful, with a cut-off: 260 MFI for k- and 109 MFI for Kpb-. Slight overlap was found on the Kpb validation pools but the number of cells falling below the cut-off were enough to identify a pool with a negative sample.

Routine lab work:

The 3 K/K and the KPA/KPA new blood donors where pheno and genotype confirmed.

Summary / Conclusions: It is feasible to analyze pooled samples to identify blood donors with rare phenotypes. A cytometric screening approach like ours, is a scalable method for a more efficient rare blood donor identification. This procedure has the huge advantage of being easily applicable in most labs, as it uses standard tools and techniques commonly available. It has already been included in our routine and other antigens will be explored soon.

P610 | Multi-rare: A case of an H-deficient Co(a-) negative patient and a review of multi-rare donors at Canadian Blood Services

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Background: The management of rare blood phenotypes can be complex and require significant coordination with blood suppliers to identify appropriate rare units and donors. These situations can be compounded by the presence of multiple unexpected rare phenotypes (i.e. not within the same blood group system or otherwise linked) in an individual patient. There is limited literature published on multi-rare phenotypes.

Aims: We report an extremely rare case of an H-deficient pediatric patient with anti-H (Bombay phenotype) who has the additional rare phenotype of Co(a-). We also aim to review the number of multi-rare donors within the Canadian Blood Services' (CBS) Rare Blood Program (RBP).

Methods: For the case workup, hospital-based ABO typing, antibody screen and panel investigations are performed in solid-phase (Capture-R). Additional methods include saline tube antibody panels. National Immunohematology Reference Lab (NIRL)-based testing includes saline tube and gel (MTS gel) antibody panels with and without ficin and DTT treatment. Red cell (RBC) genotyping performed using ID CORE XT. To quantify multi-rare donors in CBS' RBP, a search was performed in our donor database assessing all donors for multiple rare types. The following were excluded: 1) null types affecting antigens within the same blood group, 2) linked rare types (e.g., InLu and AnWj-, En(a-) and Wr(b-)).

Results: A pediatric patient from Syria presented to hospital with a bowel perforation. A history of RBC transfusion as a newborn

was obtained. Initial testing indicated an O RhD positive blood group with a positive antibody screen. A 14-cell panel in solid-phase was panreactive with 3+/-4+ reactivity accompanied by a negative autocontrol and DAT. Samples were sent to NIRL for further workup. RBC genotyping indicated CO*B homozygosity predicting a Co(a-) negative phenotype that was confirmed serologically using unlicensed anti-sera. As the rest of the serologic workup was pending at NIRL and patient required urgent surgery, a liquid Co(a-) unit was issued to the hospital with assumption that the patient may have anti-Co(a). The unit was 4+ incompatible when tested at the hospital. Additional serologic testing at NIRL demonstrated 3+ reactivity with Co(a-) negative cells at all three phases suggesting another high incidence antigen responsible for the panreactivity; 4+ reactivity was seen with ficin and DTT treatment. Testing with anti-H was non-reactive suggesting an H-deficient phenotype. Additional testing with multiple H-negative cells was non-reactive confirming the presence of anti-H. Patient typed as Le(a+b-). Crossmatch compatible H- K- red cells were recommended for transfusion. Patient blood management was implemented at the hospital and no transfusion was required following intravenous iron and erythropoietin. The patient and family were provided education and connected with CBS' RBP to identify potential rare donors. In our donor database search for multi-rare CBS donors, four donors were identified to be multi-rare: (1) S-s-U- Js(b-), (2) S-s-Uvar Js(b-), (3) S-s-Uvar Jo(a-), (4) k- Co(a-).

Summary / Conclusions: To our knowledge, this is the first reported case of an H-deficient Co(a-) patient. This case emphasizes the importance of correlating genotyping with serology, and the existence of multi-rare patients and donors. It also highlights the importance of a multidisciplinary approach in the management of rare blood patients involving transfusion medicine physicians and technologists from the hospital and blood supplier.

P611 | Creation of a register of Chilean blood donors with rare phenotypes from the Santa Maria Clinic Blood Bank

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Background: Donors with uncommon or rare phenotypes have a frequency close to 0.1%. These donors are necessary for transfusion therapy of patients with rare phenotypes, who do not have an alternative to transfusion. In Latin America there are only registries in Brazil, and an Ibero-American registry is being built up by the Grupo Colaborativo Iberoamericano de Medicina Transfusional (GCIAMT). In Chile, the first and only registry that exists to date is the one presented in this report.

Aims: To create a register of Chilean donors with rare blood group phenotypes.

Methods: Samples from 5107 blood donors were studied by serological and/or molecular methods. All donors agreed to participate in the

P611 - Table 1: Results.

Screening method (1st sample)	Phenotype found	No. of Donors	Confirmation method (2nd sample)	Confirmation by NGS
IDCORE XT	Yt ^a -	1	IDCORE XT	X
Serológico	k-	1	HeaBeadChip	✓
Serológico	Di ^b -	2	HeaBeadChip	X
Serológico	Di ^b -	1	HeaBeadChip	✓
Hea BeadChip	Fy null	1	HeaBeadChip	X
Hea BeadChip	Fy null	1	HeaBeadChip	✓
Hea BeadChip	Lu ^b -	1	HeaBeadChip	✓

study. All analyses were performed in the laboratories of immunohe-matology and blood group molecular biology of the Clínica Santa María blood bank.

1. Screening

Serological methods: The NeoGalileo equipment (IMMUCOR) with antisera (anti-K,-k, -Dia) and plasma with anti-Dib was used. K antigen was studied in 4200 samples, K+ samples were studied with anti-K; and in 825 samples Dia antigen was also studied, Dia+ donors were analyzed with anti-Dib.

Molecular methods: 907 samples were genotyped, the platforms employed to genotype the samples were: (i) HEA Beadchip (IMMUCOR), 38 blood group antigens from 11 blood group systems were studied in 720 samples; and (ii) IDCORE XT (Grifols), 37 erythrocyte antigens from 10 blood group systems were analyzed in 187 samples. Donors with an uncommon phenotype were contacted and a second sample was requested to confirm the phenotype.

2. Confirmation: HeaBeadChip and IDCORE Xt platforms were used. In addition, 4 samples were studied by Next generation sequencing (NGS) (iSeq™ 100, illumine) previously confirmed genotyped, 2 panels were used to analyze 47 genes from 44 blood systems (001 to 044). Sequence analysis was performed by a bioinformatic engineer. Donors with a confirmed rare phenotype were contacted to inform them of the result and to invite them to participate in the registry.

Results: Eight donors with rare phenotypes were found and confirmed, the phenotypes detected are shown in Table 1:

All donors agreed to participate in the registry of donors with rare phenotypes of Clínica Santa María and in turn of the GCIAMT registry.

Summary / Conclusions: (1). The strategies implemented allowed the formation of a registry of donors with uncommon phenotypes, being the only one in Chile. (2). Both serological and molecular strategies have a high value in the search. (3). NGS allows the study of genes from all blood systems, which is superior to genotyping platforms; however, it demands more time and training. (4). It is necessary to increase efforts to search for donors in other centers in Chile and to be able to respond to the transfusion needs of patients with these phenotypes.

P612 | Rapid flow cytometry beads assays for the confirmation of IgA-deficiency and the detection of anti-IgA in IgA-deficient individuals

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Background: IgA-deficient blood products are sometimes requested to treat patients with a known IgA-deficiency (< 500 ng/ml) and a history of allergic transfusion reaction and/or with the presence of anti-IgA in their blood. Although severe allergic transfusion reactions due to IgA/anti-IgA interaction are very uncommon, blood services must be able to provide adequate blood components at the request of hospitals. Various screening methods can be used to identify IgA-deficient donors; however, IgA deficiency must be confirmed before these donors can be included in an IgA-deficient donor registry. The ability to detect the presence of anti-IgA in IgA-deficient recipients is also important to guide the selection of appropriate blood components for transfusion.

Aims: Develop rapid flow cytometry bead assays to confirm IgA deficiency and detect the presence of anti-IgA in IgA-deficient individuals.

Methods: For the confirmation of IgA deficiency, polystyrene beads are coated with a mouse anti-human IgA monoclonal antibody. Samples are diluted (1/50) and a volume of 40 µl is mixed with 2.5×10^4 beads. Human IgA subclasses standard Serum (500 ng/ml) is used to determine the threshold signal for IgA deficiency. After incubation (20 min) and washing, anti-human IgA-FITC is added and incubated (20 min) before washing. The beads are then analyzed by flow cytometry. For anti-IgA detection, the beads are coated with purified human IgA. The rest of the procedure is similar except for the dilution of the sample (1/100) and the use of anti-human IgG-FITC is used to detect the presence of anti-IgA on the beads. Appropriate positive and negative controls are included in the assays.

Results: Testing of 9 normal plasmas and 13 confirmed IgA-deficient plasmas with the rapid bead assay enabled all samples to be correctly identified. Testing of 7 IgA-deficient plasmas already characterized for the presence ($n = 3$) or absence of anti-IgA ($n = 4$) with the rapid bead assay also showed the correct detection of anti-IgA in all positive samples, and its absence in negative samples. The rapid bead

assay takes 90 min to perform instead of 4.5-5 h for a standard ELISA (about 3 times faster).

Summary / Conclusions: The two rapid flow cytometry bead assays described are very useful for the rapid confirmation of suspected IgA-deficiency or the presence of anti-IgA. They have been shown to be less expensive than a standard ELISA but equally accurate, and better suited for the analysis of a small number of samples. With regard to the level of anti-IgA detected, we arbitrarily set a threshold of 12.5 U/ml based on a commercial anti-IgA ELISA, given that clinically relevant anti-IgA levels are unknown. This threshold could be revised if samples from patients with transfusion reactions involving IgA/anti-IgA were available, to more accurately reflect clinically significant levels of anti-IgA.

P613 | Update on the prevalence of IgA-deficient blood donors at the Oporto Blood and Transplantation Center (CSTP)

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Background: Selective Immunoglobulin A Deficiency (SIgAD) is the most common primary immunodeficiency in the Western world, and most affected individuals are asymptomatic (75-90%). This condition is defined by the European Society for Immunodeficiencies (ESID) as a serum IgA <0.07 g/L, in individuals aged ≥ 4 years, with normal IgG and IgM levels and excluding secondary causes of hypogammaglobulinemia. The American Rare Donor Program (ARDP) defines SIgAD as an IgA level <0.05 mg/dL when tested on two separate occasions, while the clinical definition of IgA deficiency is <7 mg/dL. Anti-IgA antibodies can be directed against IgA1 and IgA2 and can be detected in individuals with IgA deficiency. Although most anti-IgA antibodies are IgG1 in nature, they can be of the IgE isotype, which can cause type 1 hypersensitivity and anaphylactic reactions during a blood transfusion. The prevalence of SIgAD varies according to ethnicity and global location, and is approximately 1:700 individuals worldwide. In the Caucasian population, SIgAD ranges from 1:500 to 1:3000, while in the Asian population this prevalence is significantly lower (1:4000 in China to 1:18000 in Japan). Since 2006, the Laboratory of Leukoplatelet Immunology of the Oporto Blood and Transplantation Center (CSTP) has had a panel of IgA-deficient donors in order to respond safely to requests from hospitals.

Aims: Increase the SIgAD donor panel as more donors are screened and update the prevalence data in the CSTP.

Methods: Between 2006 and 2023, 28,561 male donors of all blood groups in the ABO system were screened for IgA and donated either in mobile brigades or at the CSTP's fixed post. IgA was quantified using the nephelometry method on the BN ProSpec System (Siemens Healthineers®, Germany). Samples with levels <26.5 mg/dL were subsequently tested with a protein precipitation reagent with a detection limit of 6.97 mg/dL. Samples with values <6.97 mg/dL were tested with a highly sensitive technique (IgA Latex) that allows the detection of IgA concentrations <0.0233 mg/dL.

Results: A total of 87 donors were found to have an IgA deficit, with values <0.0233 mg/dL. The calculated frequency between 2006 and 2023 was 0.30% (87 deficits out of 28,561 donors), which translates into a prevalence of 1:328.

Summary / Conclusions: Despite the increase in the donor panel, the prevalence remained close to that calculated between 2006 and 2021, which was 1:363 donors. Based on this panel, the CSTP responds to blood requests from hospitals in the following ways: calling donors to donate or thawing cryopreserved components, in the case of reserve requests and washing components for urgent requests.

P614 | Rare Donor Registry of India (RDRI)—creating a database of rare and regular blood donors profiled for all blood group antigens

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Background: Most blood banks in India do not test for minor blood group (BG) antigens and issue only ABO and RhD matched units. Obtaining compatible donors is challenging when patients are alloimmunized against foreign minor BG antigens. Many units have to be crossmatched and even then, they may receive least incompatible units as a national database of BG typed donors is not available. This suggests a need for a national donor registry.

Aims: The study aims to develop a database of regular Indian blood donors typed for all the red cell antigens and establish Rare donor registry of India.

Methods: 4000 'O' group regular donors from four different regions were enrolled and tested for 22 clinically important antigens (8 BG systems) using Gel card technology. For predicting the remaining BG antigens, targeted Next generation sequencing (NGS) was used and 51 genes of 43 BG systems were genotyped.

Results: The donors from north and south regions had higher frequency of RhD negatives with lowest in East India. The frequency of Fy^a, Fy^b Jk^a, Jk^b, M, N, S, s and P₁ was almost similar in all the four groups. 50 donors with rare Rh phenotypes were identified. Only one K homozygous donor was identified. Frequency of SS homozygous was highest (13.2%) in West India. A total of 2808 samples were tested by NGS. The genotyping results for common BG antigens of Rh, MNS, Kell, Kidd, Duffy, Lutheran and P (where commercial antisera were available) were concordant with serotyping. A total of 41 donors were found to be positive for low incidence antigens and 21 were predicted to have weaker expression (Table 1). Three donors were found to be high frequency antigen negative namely: YTLI-

P614 - Table 1: Unique observations of predicted blood group phenotype based on genotyping in Indian donors

BG system	Genotype	Predicted Phenotype	n
A Low Incidence Antigen positive donors			
MNS	GYP* <i>Mur</i>	Mi(a+)	1
KEL	KEL*02.10/KEL*02	UJ ^a	3
	KEL*02.21/KEL*02	Kp(a-b-c+)	1
Diego	DI*01/DI*02	Di(a+)	5
Cartwright	YT*02/ YT*02	Yt(a-b+)	11
Colton	CO*01.-04/CO*01	CO: - 1, -2, 3, -4	1
Indian	IN*01/IN*02	In(a+)	25
KANNO	KANNO*01.-01/ KANNO*01.-01	KANNO (1-)	8
B Predicted weak expressing antigens			
Rh	RHD*08.01/ RHD*08.01	Partial D (DFV)	3
	RHD*18/ RHD*18		
Duffy	FY*02W.01/FY*01	Fy ^x leading to weak Fy ^b expression	15
Cromer	CROM*01.-04/CROM*01.04	Weak expression of Tc(c+)	3
VEL	VEL*01N.01/VEL*01	Weak expression of Vel	7
C Novel variants			167

(n = 2) and KANNO (1-) (n = 1). Based on the data generated, a total of 515 rare donors, negative for a combination of clinically important common blood group antigens were identified. Out of these, 79.03% donors were negative for a combination of two common antigens, 15.92% for three, and 4.85% for four antigens. These have now been registered as part of RDRI. Also, 150 very rare donors (including Bombay, Para Bombay, D-, p null etc.) were also recruited and registered as part of the registry.

Summary / Conclusions: We have established the RDRI, an electronic database of red cell antigen typed regular voluntary blood donors at national level for the first time. We have also defined rare donors negative for combination of common antigens and identified novel, rare and common variants prevalent in the Indian population. Lastly, we have determined the frequency of all the BG antigens in Indian donors.

P615 | Rare donors in Portugal—a project for growth

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Background: Blood donors with rare phenotypes are found in less than 0.1% of the population and include individuals with specific phenotypic combinations or the absence of high-prevalence antigens. Resolving antibody specificity(ies) in immunised patients is usually easy for any immunohematology laboratory. However, in patients with sickle cell anaemia, thalassaemia, rare phenotypes

related to ethnic populations (hereditary characteristics of blood group antigens), multiple antibodies against high-frequency antigens, screening for haemolytic disease in newborns and pregnant women, global collaboration is essential to provide rare blood. Many countries have established rare blood programs to aid patients with complex serologic requirements, and international programs exist to coordinate efforts across national boundaries.

Aims: How to proceed in order to have a Rare Donor Panel with an increase in new donors every year.

Methods: Depending on the country's economic availability, there are always some efforts to be made in order to make it possible to add new rare donors at any time. Different methods can be used to determine a donor's RBC (red blood cell) phenotype, including the use of serological phenotyping antisera or genotyping. We present automated phenotyping for around 5-10% of all donors per year and genotyping for around 2%, with a selection of various phenotypes according to the needs of our Portuguese hospitals: phenotypes such as R1R1, R2R2, Ror, r'r', r''r'', Fy(a-b-), Fy(a-b-) with Jka or Jkb, Fy(a-b-) with S or s, Fy(a-b-) with ccDee, p (PP1Pk antibody), Kp(b-), Jr(a-), Co(a-b-), Jk(a-b-), S-s-U-, Yt(a-); Increase the Selective Immunoglobulin A Deficiency (SigAD) donor panel as more donors are screened using the nephelometer method on the BN ProSpec System (Siemens Healthineers®, Germany). Continue to add units to our Cryobiology portal, which consists of homologous and autologous RBC units and IgA-deficient units (FFP and RBC units) frozen by automatic glycerolization (ACP 215 Haemonetics) using the high glycerol concentration method.

Results: In the last two years we have been able to phenotype 10369 of our donors in Lisbon and in Porto. Some of these donors have been genotyped with Innotrains Kits for SSP-PCR, Germany. The number of rare donors in our ASIS® Computer System is around 192 and in the

Cryobiology Portal 197. With improved automation and extended phenotyping, we hope to look for more rarities and genotype even more of our donors.

Summary / Conclusions: The importance of having further studied donors allows us to have the right blood when the need comes, and to manage our reserve units for our patients. It is important to have good policies for the retainance of these donors. As global migration becomes more prevalent, the need for rare blood will increase. In Portugal, it will be necessary to develop better coordination and organizational with hospitals at a national level, to increase the racial diversity of our donor base and to create the National Rare Donor Panel/Registry. This program ensures the availability of various blood components for patients who have unique requirements and whose needs are not easily met by the general donor population. In these scenarios, good communication, realistic expectations, and collaboration with all parties will ensure favourable patient care and outcome.

P616 | The creation and development of the Irish rare donor programme

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Background: Ireland's population is diversifying as a nation, and this is evidenced by the detection of rare red cell phenotypes in both our donor and patient cohorts. To reduce our reliance on international rare donor banks the Irish Blood Transfusion Service (IBTS) is establishing an Irish Rare Donor Programme (RDP). Malarial antibody testing was introduced by the IBTS in May 2023, in conjunction with targeted ethnicity testing. This has opened potential donor recruitment outside the White Irish donor pool and has facilitated targeted phenotyping of different ethnic cohorts.

Aims: The aims of the RDP are to identify, engage, and retain donors within the Irish donor pool who have a rare phenotype.

Methods: The IBTS began recording donor ethnicity on 22nd May 2023, and introduced target antigen phenotyping by donor ethnicity to maximise antigen negative types from donor screening, and identify rare donor types. Kp^b and Lu^b phenotyping of primarily O RhD negative donors commenced on the Beckman Coulter PK7400 in May 2023 and January 2024 respectively. Quotient Anti-Lu^b was tested at a 1/50 dilution, Lorne anti-Kp^b was tested at a 1/2 dilution for mass screening on the PK7400.

Any suspected antigen negative phenotypes were confirmed by the IBTS Red Cell Immunohaematology department. KEL2 (k) typing was carried out on a select number of K+ donors. Current donors with a rare type were individually contacted and notified of their rare phenotype. These donors were asked to either donate on a regular basis, or as requested, depending on the need for their phenotype.

Results:

P616 - Table 1: Number of donations tested vs. success rates

Antigen phenotype	No. of donations screened	No. of antigen negative phenotypes	% confirmed antigen negative types
Jk(a-b-)	144	0	0%
Jr(a-)	1	0*	0%
Fy(a-b-)	64	23	35.9%
U-	64	1**	1.56%
Lub-	1283	10	0.63%
Kpb-	9828	2	0.02%
k-	684 K+ donors	93***	13.6%

* 1 Crossmatch compatible donation identified in 2020 was subsequently identified as Jr(a+^w) by the IBGRL.

** 1 additional S-s- donation identified, however donor was Malaria antibody positive and was not referred for U phenotype confirmation.

*** K+ donors account for 9% of the Irish donor pool.

Summary / Conclusions: The IBTS are in the process of developing the RDP further. Our next steps are as follows: (1). Develop and maintain communication with donors by appointing a designated medical officer to inform donors of their rare phenotype, and regularly check in with them. (2). Scope out the feasibility of a frozen blood bank. Currently, most of the rare blood is imported for specific patient need, with an associated time delay. (3). High-throughput molecular genotyping of blood donors will also be scoped out in 2024. Molecular genotyping offers an advantage over serological phenotyping when identifying rare donors, as more antigen types can be identified in one test. (4). The IBTS will contribute to the international rare donor panel.

P617 | Rare group blood—the dilemma between blood units cryo-preservation and donors register

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Background: Selecting units of rare blood for transfusion to patients with complex immunisation is one of the most critical processes in Transfusion Medicine. In January 2018 in the Transfusion Medicine of the Venice District (North-East Italy) a project was

P617 - Table I: Activity of our rare group cryopreserved blood bank

Year	Genotypin in Blood Donors	Rare Blood Units in Storage	Ex novo frozen Units	Transfused Rare Blood Units
2018	1322	273	5	29
2018	1023	249	5	45
2020	1150	209	9	21
2021	1159	197	20	44
2022	1464	172	29	14
2023	1372	188	37	11

established with the following goals: (1) identifying, using both phenotyping and genotyping, rare blood donors; (2) creating a registry of rare donors; (3) organising a bank of liquid and frozen rare blood units.

Aims: In this paper we report our experience in selection of rare group donors, phenotyping and genotyping procedures adopted, the strategies in managing the inventory of rare group donors and the database of cryopreserved rare group red blood cells.

Methods: Our extended serum typing profiles involves the characterization of the following erythrocyte blood group antigens: ABO, C, E, c, e, K, k, Kpa,b, Fya,b, Jka,b, M, N, S, s, Lua,b, P1, Xga, Lea,b using a adopted a gel test method supplied by Grifols. For genotyping, we adopted the microarray-based method "Bead-Chip" supplied by Immucor. Our extended genotyping serum profile involves the genetic characterization of the following erythrocyte blood group antigens: C, E, c, e, K, k, Kpa,b, Jsa,b, Fya,b, Jka,b, M, N, S, s, Lua,b, Dia,b, Doa,b, Hy, Joa, Coa,b, Sc1,2, LWa,b. For cryopreservation our choice fell on a "high glycerol" system and storage in an electric ultra-freezer at -80°C using a Haemonetics ACP 215 equipment.

Results: As reported in table I from 2018 to 2023 we observed a progressive decrease in the number of cryopreserved blood units which dropped from 273 to 188. As reported in table II genotyping activity is settled between 1023 and 1472 donors / year.

Summary / Conclusions: Blood establishment involved in rare blood programs usually perform mass screening of the donated red cell products, using automated method to study phenotype ad/or genotype. Usually, these screening are performed following some pre-established criteria that involves testing for some specific antigens in large batches; donors' selection considering ABO and Rh phenotype, and in western Countries, extensive typing of blood donors of not Caucasian origin. A relevant matter of discussion is whether it is preferable to have a supply of cryopreserved rare group blood units or whether it is preferable to have a database of rare group donors to be called upon in case of need; each of these approaches has advantages and disadvantages. To create our bank of rare blood we adopted synergistically both these approaches combining a large database of genotyped donors with a small supply of cryopreserved extremely rare group red blood cells.

P618 | A comprehensive study of a positive direct antiglobulin test in a newborn resulted in the recruitment of three rare blood donors

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Background: The Diego blood group system consists of 23 antigens carried on a multipass membrane glycoprotein called Band 3 but it is the presence or absence of Di^a and Di^b that is of importance in determining a person's Diego blood type. Di^b is universally expressed in most populations while Di^a is a low frequency antigen, found in individuals with Mongolian or Amerindian ancestry. Anti-Di^b has been reported to be a clinically significant antibody responsible for mild Haemolytic Disease of the Newborn (HDN) and none to moderate haemolytic transfusion reactions.

Aims: The aims of this study were to report the absence of HDN due to anti-Di^b in a baby and the recruitment of three Di^b negative blood donors for the registry of rare blood donors.

Methods: Case report.

Results: We report the case of a O+ newborn with a positive direct antiglobulin test (DAT) without signs and symptoms of HDN. The mother (proband) was a 32-year-old woman, gravida 3, para 3 with current medically uncontrolled pregnancy. She was O+ with a positive indirect antiglobulin test. Antibody identification studies performed in the maternal serum as well as in the eluate of the newborn red blood cells showed a uniform pan-reactivity with all commercial panels used. Direct antiglobulin test and autocontrol were negative. The pan-reactivity was also observed after enzyme (papain) and DTT treatment of panel RBCs. The results obtained suggested the presence of an alloantibody directed against a high frequency antigen. Allogeneic adsorption studies were also performed to analyze a possible mixture of alloantibodies giving negative results. Considering the occurrence of Di^a in our admixed population, maternal RBCs were tested with a polyclonal anti-Di^a showing a positive result, however, no anti-Di^b were available in our laboratory. Diego genotyping performed with a SSP-PCR method indicated that the mother was Di^aA/Di^aA while the newborn was Di^aA/Di^bB. Later, first-order blood relatives were summoned, all of them being O+. The crossmatch between the proband's serum and the RBCs of her relatives were incompatible with their parents and compatible with the two sisters. Finally, the Diego genotype of each relative showed that the parents of the proband were Di^aA/Di^bB and her sisters Di^aA/Di^aA. These findings suggested that the alloantibody found was anti-Di^b.

Summary / Conclusions: A comprehensive study of a positive direct antiglobulin test in a newborn allowed the identification of an anti-Di^b alloantibody in a childbearing age woman that, in the case reported, did not cause HDN. In addition, the family studies carried out allowed to form a group of compatible donors (the three sisters) for potential transfusions. Furthermore, the contact with the patient and her relatives made it possible to raise awareness about the importance of integrating the database of donors with rare phenotypes.

P619 | Application of successful PBM measures to manage a patient with a rare anti-Cra antibodyC George¹, K Perera¹, E Davies¹, H Davies¹, C Harris¹¹Clinical Services, Welsh Blood Service, Pontyclun, United Kingdom

Background: The Cr^a antigen is present on >99% of blood donor's red cells, making the provision of serologically compatible blood difficult / impossible in those with an anti-Cr(a) antibody. Due to the rarity of the antibody, there is little evidence in the literature about its clinical significance, although isolated case studies suggest that it is unlikely to cause an acute transfusion reaction but may result in accelerated destruction of transfused cells after transfusion.

Aims: To provide clinical advice on the management of patients with the rare antibody anti-Cr^a

Methods: In March 2021 the Red Cell and Immunohaematology (RCI) Department received a routine sample referral for an antenatal patient. Upon investigation, a pan-reactive antibody was detected (+5 reactions against all donor panel cells). The patient serum was tested against a panel of rare high-frequency negative cells and anti-Cr^a was detected. The baby was not affected and no blood was required for the mother at delivery. In November 2022, the patient was due to undergo elective gynaecological surgery requiring two units of blood on standby. Investigations confirmed the anti-Cr^a was still present. Allo-adsorptions were performed to remove the anti-Cr^a and excluded the presence of underlying additional allo-antibodies hidden from detection by the strong high-frequency antibody. There were no Cr(a-) donors or frozen blood units available in Wales or the rest of the UK for the patient.

Results: Patient Blood Management (PBM) Advice (1). Optimise the patients Hb and haematinics. (2). Minimise blood loss during surgery: optimum peri-surgical anticoagulation/anti-platelet drugs, use tranexamic acid and intra-operative cell salvage. (3). Transfusion advice: ABO, D and K compatible blood should be given for urgent transfusions, with particular vigilance for delayed haemolytic transfusion reactions. (4). Post surgery: Assess haemoglobin threshold tolerance based on patient's clinical state, using restrictive transfusion triggers and consider iron supplementation for management of post-operative anaemia.

Outcome: Surgery completed as planned, no blood was required for this patient during or after the procedure, and patient had an uneventful recovery.

Summary / Conclusions: Summary/Conclusion: Effective use of PBM measures and collaborative multi-disciplinary team working across organisations minimises the need to transfuse in patients with complex antibodies where compatible blood cannot be found.

P620 | The rarity of the anti-Kpb antibody—overcoming challengesD H Cardoso¹, C Peixoto², M Guz¹¹Imunohemotherapy, Hospital Espírito Santo de Évora, Évora,²Imunohemotherapy, Santa Maria Hospital, ULS, Lisbon, Portugal

Background: A Kp^b negative phenotype is a rare occurrence in the population related to a reduced antigen expression and with the possible production of antibody anti-Kp^b. Anti-Kp^b is reported to cause haemolytic transfusion reactions and hemolytic disease of the newborn. As such, when a patient in need of transfusion is classified as Kp^b negative, finding a compatible blood unit can be challenging.

Aims: Report of a rare case of anti-Kp^b.

Methods: We retrospectively analyzed the course of a patient with a rare anti-Kp^b antibody.

Results: A 77-year-old female was admitted due to a post-traumatic periprosthetic fracture of the right femur requiring surgical treatment. The patient's medical history included chronic lymphocytic leukemia (CLL) with history of previous transfusions. A pre-transfusional test was conducted to secure a blood unit for the operation room. The results revealed a positive indirect antiglobulin test, indicating the presence of an anti-Kp^b antibody. Given the surgery's elevated risk of bleeding, along with the patient's existing health conditions and a baseline hemoglobin level of 10.8 g/dL, the decision was made to postpone the surgery until a compatible blood unit could be ensured. In the interim, hematological optimization commenced with intravenous iron supplementation, and efforts were taken to coordinate with the Portuguese Institute of Blood and Transfusions (IPST) to secure an appropriate blood unit: two cryopreserved blood bags had already been allocated for this patient during a prior study conducted at the time of the CLL diagnosis. The patient's known phenotype was O ccddee kk, Kp (a+b-), MNSs,Fy (a+b+), P1+, Le (a-b+), LU (a-b+). Unfortunately, during thawing the units were damaged, rendering its use unfeasible. A search for a compatible blood donor was started, resulting in the identification of two potential donors: one located in Portugal and the other in Spain. While both donors were contacted, only the Spanish donor answered the summons. Before surgery, a compatibility test confirmed the absence of reactivity with the obtained blood unit. The patient underwent surgery without requiring a transfusion, and the blood unit was subsequently cryopreserved.

Summary / Conclusions: The Kp^b negative phenotype is exceedingly uncommon. These antibodies have been associated with acute or delayed hemolytic transfusion reactions, underscoring the critical importance of conducting compatible blood transfusions. Transfusing with a receptor of this phenotype poses a challenge, especially when antibodies have developed. Locating a suitable donor presents a considerable obstacle. This case highlights the vital importance of international cooperation, showcasing its essential role in providing treatment while prioritizing patient safety.

P621 | Assistential diagnosis of red cell transfusions with antigen matching in patients with hemoglobinopathies as a basis for the development of a computer system enabling the management of rare blood

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Background: The advancement of personalized medicine and the use of precise matching to a patient with specific antigen profile, particularly for chronically transfused populations, such as those with sickle cell disease and thalassemia, has increased the request for rare blood.

Aims: The objective of this study is to present data aimed at diagnosing patients and donors with rare blood types as a basis for the development of a computer system enabling risk management related to rare blood.

Methods: A cross-sectional study analyzing data from 3,647 patients with hemoglobinopathies at the Rio de Janeiro Blood Center, Brazil, was conducted as a partial diagnosis to identify patients with rare phenotypes. The institution's rare donor registry was also accessed to assess blood availability for these patients.

Results: The absence of multiple common antigens and other rare phenotypes such as Kp(b-), Jk(a-b-), Lu(b-), K+k-, U-, Js(b-), Hy-, Uvar Jo(a-) were identified in these patients. Rare phenotypes resulting from RH variants in sickle cell patients were also identified, such as DAR, DIIla, DAU5, hrB-. In twenty (32.3%) patients, 28 antibodies were identified. The mean red blood cell transfusion was 14 units. One hundred sixteen donors with phenotypic profiles corresponding to these patients were identified. However, we found that there are more patients with rare phenotypes than blood donors and that the number of compatible donors is insufficient to meet the transfusion need of the patients.

Summary / Conclusions: Based on the assistential diagnosis of compatible transfusions conducted in these patients with hemoglobinopathies, which demonstrates the shortage of donors with rare phenotypes for performing transfusions with precise matching, we propose a modeling for the development of a computer system that allows for better identification and monitoring of patients and donors with rare phenotypes, preventing associated complications, and enhancing blood transfusion processes in the most optimized way that best suits the reality of blood centers.

P622 | The importance of having a panel of rare blood donors - the k negative phenotype

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Background: In 2021 the Immunohematology Laboratory from Lisbon Blood and Transplantation Centre (CSTL), Portuguese Blood and Transplantation Institute (IPST,IP) studied the k antigen in all donors with K+ phenotype, in the attempt of finding a panel of k- donors. The Kell system, KEL (006) according to ISBT terminology, contains 38 described antigens, numbered from 1 to 38, with k or KEL:2 being the antigen antithetical to K (KEL:1). The k antigen is found in 99.8% of Caucasians and 100% of Africans. It is therefore a high-frequency antigen, making the KEL:-2 negative phenotype a rare one. Antibodies against the antigens of the Kell system are usually IgG and may cause severe Transfusion Hemolytic Reaction (THR) and/or Hemolytic Disease of the Newborn (NRHD). The presence of alloanti-k, makes the obtainance of blood for alloimmunized patients difficult. The study carried out in 2021 enabled CSTL to present a panel of 27 donors with the desired phenotype, k-negative (KEL:1,-2 or K+k-), who can readily be called upon when there is a need for transfusion in patients.

Aims: We present the procedure used in CSTL to find the K+k- donors suitable for patients with aloanti-k in a quick way.

Methods: During the last month of 2023 we had two requests of K+k- red blood cell concentrates for aloanti-k patients. 13/12/2023 - JPVS, male, 68 years old, with O CcDEe, K+k- phenotype. Request from one Hospital of 3 O Rh+ K+k- red blood cell (RBC) concentrates for a hip replacement surgery. The patient had previously been studied in our Center in 2022. The study for the IAI (irregular antibody identification) with the Panocell-10, and Panocell-20 from Immucor (U.S.A.) showed the existence of anti-k. 17/12/2023 - MLPB, female, 89 years old, with O ccDEe, K+k- phenotype. Request from another Hospital of 2 O Rh+ K+k- RBC concentrates for peri-implant fracture surgery of the left proximal femur. The study for the IAI with the Panocell-10, and Panocell-20 from Immucor (U.S.A.) in our Laboratory showed the existence of anti-k. In total 5 O Rh+, K+k- RBC concentrates were requested in a short period of time. The previously made list of K+k- donors allowed us to quickly identify and contact possible candidates for blood donation since there was no availability of K+k- units in our Blood Bank. The Immunohemotherapy Doctors of CSTL proceeded with the donor contacts from the list. Four were available to donate blood in the next days. On the 22nd of December 2023 we had four K+k- RBC units available. The first Hospital, with the male patient, performed the

surgery on the 12nd of January 2024 and there was an emergent request for two more RBC concentrates of the same phenotype. Two more units were available from donors who had been contacted in December. The second Hospital, with the female patient, performed the surgery as the patient's hematological parameters allowed it to be carried out and there was no need for transfusion support.

Results: The existence of the listed donors provided from the 2021 study allowed us to quickly search for the intended phenotype.

Summary / Conclusions: The creation and maintenance of an up-to-date list of donors with the k- phenotype is an important resource for the management of the transfusion needs in these patients. The Immunohematology Laboratory from CSTL is keeping the search for K +k- phenotype in donors and the list is continuously being updated.

Immunohaematology—platelet immunology

P623 | Kupffer cells and red pulp macrophages wipe out allogeneic platelets from the bloodstream during HLA class I-alloimmune refractoriness

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Background: Transfusing platelets into patients already alloimmunized against the HLA class I antigen can be challenging. Such alloantibodies can result in rapid elimination of the transfused platelets from the bloodstream, leading to transfusion failure known as platelet transfusion refractoriness (PTR). This removal process of allogeneic platelets from the bloodstream during refractoriness is not fully understood. Both the liver and the spleen are candidates for harboring cells responsible for the sharp clearance of allogeneic platelets in refractoriness. Liver macrophages excel at internalization of desialylated platelets, while the splenic counterpart are highly effective at phagocytosis of opsonized platelets.

Aims: To identify the immune cells in the liver and spleen taking up allogeneic platelets in a murine model of HLA class I-alloimmune PTR.

Methods: A previously described murine model of platelet alloimmunization was used to mimic PTR. Fluorescent allogeneic platelets (H2b) were tracked by flow cytometry following transfusion into naïve or alloimmune mice (H2d). After 30 min, organs were enzymatically digested and cells stained with CD45, F4/80, and CD11b and analyzed by flow cytometry to identify Kupffer Cells (KC) in the liver and Red Pulp Macrophages (RPM) in the spleen. Precise allogeneic platelet localization was evaluated by confocal microscopy. The relative importance of spleen and liver were studied in vivo by evaluating PTR either in splenectomized/sham mice or in mice treated with asialofetuin/fetuin (8 mg i.v.). Data are presented as mean \pm SEM.

Results: In immunized mice, allogeneic platelets were removed from the circulation within 30 min. At this time point, we found (61.7 \pm 7.7)%

of KC and (18.7 \pm 3.0)% of RPM positive for allogeneic platelets as compared to (2.5 \pm 0.9)% and (1.5 \pm 0.1)% in naïve mice, respectively ($p \leq 0.05$, $n = 3$). In both organs, CD45⁺ F4/80⁻ cells were not positive for allogeneic platelets in neither naïve nor refractory mice at this same time point, which underscore KC and RPM as the main immune cells responsible for platelet uptake. Accordingly, z-stacks from confocal microscopy showed multiple allogeneic platelets inside KC and RPM in refractory but not in naïve mice. To evaluate the role of the spleen in platelet clearance during PTR, immunized mice underwent splenectomy prior to transfusion. Surprisingly, alloimmune mice remained in a refractory state similar to the sham control mice: (0.8 \pm 0.4)% vs (1.0 \pm 1.0)% of total platelets in circulation at 30 min, respectively ($n = 3$), indicating that the spleen is dispensable for platelet elimination. We treated mice with asialofetuin to block asialoglycoprotein receptors prior to transfusion. The treatment did not affect the platelet clearance at 30 minutes ((0.8 \pm 0.4)% vs (0.7 \pm 0.4)%; $n = 3$), indicating that the mechanism of platelet internalization is independent of desialylation.

Summary / Conclusions: Both KC in the liver and RPM in the spleen take up allogeneic platelets during refractoriness. Neither the sole absence of the spleen nor just the blockade of asialoglycoprotein receptors is sufficient to abrogate refractoriness in alloimmune mice, hinting at alternative mechanisms of platelet internalization mediated by anti-HLA class I antibodies during refractoriness.

P624 | Abstract withdrawn

P625 | Understanding the role of platelet desialylation in ITP physiopathology and treatment response

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Background: Immune thrombocytopenia (ITP) arises from the production of autoantibodies targeting platelet membrane proteins, predominantly GPIIb/IIIa (70%–80%) and GPIb α (20%–40%). Standard treatment protocols involve corticosteroids and intravenous immunoglobulin (IVIG), with splenectomy reserved for refractory cases. Traditionally, platelet clearance in ITP is attributed to the antibody-dependent (Fc) pathway within the splenic reticuloendothelial system. However, approximately 15%–25% of patients exhibit resistance to conventional therapies and splenectomy without clear explanation. Recent studies have highlighted the role of platelet desialylation, a process involving the removal of sialic acid from the platelet membrane. This exposes β -galactose residues, which are recognized by the Ashwell-Morell receptor (AMR) in Kupffer cells within the liver. Consequently, platelets may undergo phagocytosis in the liver instead of the spleen. Our hypothesis suggests that the presence of platelet desialylation may serve as a predictor of treatment failure with classic ITP therapies.

Aims: (1) Investigate the presence of platelet desialylation in patients diagnosed with ITP and correlate it with the presence of platelet autoantibodies; (2) Explore the correlation between the presence of

platelet desialylation and the response to corticosteroid therapy in ITP patients.

Methods: A prospective study was conducted, including all clinically diagnosed patients with ITP, GT, and BSS in a quaternary service over a period of 2 years. All enrolled patients were investigated for the presence of anti-platelet antibodies through the following methods: Platelet Immunofluorescence Test (PIFT), Monoclonal Antibody Immobilization of Platelet Antigens Test (MAIPA), and the platelet desialylation test. The latter method involves labeling platelets with Ricinus Communis Agglutinin I (RCA-1) and reading the results through flow cytometry. The Chi-square test was applied for group comparison.

Results: A total of 97 (78.2%) patients with primary ITP and 27 (21.8%) with secondary ITP were included. In the primary ITP group, 88.7% (86/97) had a positive desialylation test, while 11.3% (11/97) tested negative. In the secondary ITP group, 63% (17/27) showed positive desialylation. There was no correlation in both primary and secondary ITP between desialylation positivity and the presence of anti-platelet antibodies by PIFT or MAIPA ($p > 0.5$ for all analyses). Primary ITP patients with positive desialylation had a higher proportion of treatment failure with corticosteroids ($n = 33/86$ patients, 38.3%) compared to those with negative desialylation results ($n = 1/11$, 10%) ($p = 0.55$). This difference was not replicated in the secondary ITP group ($p = 0.452$), possibly due to the smaller sample size.

Summary / Conclusions: The frequency of platelet desialylation in patients with ITP (primary or secondary) is high and represents an important (and novel) pathophysiological step in platelet removal. Platelet desialylation has shown potential as a marker for therapeutic failure to corticosteroids in primary ITP, suggesting the need for an alternative immunomodulatory treatment approach.

P626 | Differences of platelet desialylation in immune purpuras and congenital thrombocitopathies

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Background: Congenital thrombocytopathies such as Glanzmann Thrombasthenia (GT) and Bernard-Soulier Syndrome (BSS) are rare hereditary disorders stemming from quantitative or qualitative alterations in the platelet membrane glycoprotein complex GPIIb/IIIa and GPIb α , respectively. Consequently, the treatment may involve platelet transfusion, thus raising the chances of developing alloantibodies against platelet antigens. Immune thrombocytopenia (ITP) is characterized by the production of autoantibodies targeting platelet membrane proteins, primarily GPIIb/IIIa and GPIb α . Therefore, management includes immunosuppressants, immunomodulators, and, if necessary, splenectomy, with transfusional support rarely required. The presence of alloantibodies and autoantibodies is mediated through an Fc-dependent pathway, leading to phagocytosis by the reticuloendothelial system. Another clearance mechanism arises from the presence of antibodies capable of platelet activation, releasing neuraminidase (NEU)

that removes sialic acid, resulting in cellular recognition by Kupffer cells and hepatic phagocytosis, a process known as platelet desialylation.

Aims: To investigate the presence of platelet desialylation in patients with ITP, GT, and BSS, and correlate it with the presence of alloantibodies or autoantibodies against platelets. Additionally, to evaluate whether the presence of platelet desialylation correlates with other platelet laboratory techniques, considered the gold standard.

Methods: A prospective study was conducted, including all clinically diagnosed patients with ITP, GT, and BSS in a quaternary service over a period of 2 years. All enrolled patients were investigated for the presence of anti-platelet antibodies through the following methods: Platelet Immunofluorescence Test (PIFT), Monoclonal Antibody Immobilization of Platelet Antigens Test (MAIPA), and the platelet desialylation test. The latter method involves labeling platelets with Ricinus Communis Agglutinin I (RCA-1) and reading the results through flow cytometry.

Results: A total of 97 (70.2%) patients with primary ITP, 27 (19.6%) with secondary ITP, 10 (7.2%) with GT, and 4 (3%) with BSS were included in the study. In the primary ITP group, 88.7% (86/97) had a positive desialylation test, while in secondary ITP, it was 63% (17/27). There was no correlation in both primary and secondary ITP between desialylation positivity and the presence of anti-platelet antibodies by PIFT or MAIPA ($p > 0.5$ for all analyses). In the congenital thrombocytopathies group (GT and BSS), 71.4% (10 out of 14) tested positive for desialylation. When correlating the positive desialylation test with the presence of anti-platelet antibodies by PIFT or MAIPA, the rate was 70% (7/10). There is evidence of a correlation between test positivity and the presence of platelet alloantibodies in this patient group.

Summary / Conclusions: The platelet desialylation test has proven to be a potential laboratory biomarker, demonstrating that when antibodies are present, hepatic clearance also occurs. This finding justifies the refractoriness of 15%–25% of patients to conventional therapies and splenectomy.

P627 | Development of iPS cell-derived specific HPA panel cell lines using genome editing technology—specific detection of HPA-3a antibody

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Background: There are 35 human platelet antigens (HPAs) involved in thrombocytopenia caused by antigen-antibody reactions. Many of these are present on GPIIb/GPIIIa, including HPA-3, one of the most clinically significant HPAs. We previously developed a gene transfer system that allows for antibody testing and reported transgenic cells that are applicable for antibody detection; however, their low GPIIb/GPIIIa expression is a drawback. It is now possible to mass-produce

induced pluripotent stem (iPS) cell-derived platelets from the expandable immortalized megakaryocyte progenitor lines, imMKCLs, which express GPIIb/GPIIIa.

Aims: In this study, we produced an imMKCL expressing specific HPAs using genome editing technology. In addition, we estimate imMKCLs were evaluated to detect HPA-3a antibody with specificity by indirect fluorescent antibody test (IFT).

Methods: HPA-3-edited imMKCL generation: gRNA and ssODN were generated and introduced into iPS cells that were reprogrammed from an imMKCL and Cas9 using an electroporator and cloned to obtain HPA-3a and HPA-3b homozygous cells. Then, the cells were cultured for differentiation into megakaryocytes and subjected to flow cytometric analysis using CD41 and CD42 antibodies, and the resulting cells (imMKCLs) were passaged once or twice a week.

Antibody testing: WHO standard sera (anti-HPA-1a, anti-HPA-3a), specific anti-HPA-3a serum, or healthy human serum were incubated with 30,000 imMKCLs. The cells were then washed, incubated with FITC-labeled CD42b and PE-labeled anti-human IgG, and subjected to flow cytometry.

Results: The gene-edited imMKCLs continued to grow for more than 12 months. The mean doubling time of these cells was approximately 24 h. The anti-HPA-1a serum was positive for HPA-3a and HPA-3b imMKCLs. The anti-HPA-3a sera were only positive for HPA-3a imMKCL and standard HPA-3a serum could be detected at up to $32 \times$ dilution (this serum was positive up to $8 \times$ or $16 \times$ dilution with IFT using platelet).

Summary / Conclusions: Our findings show the possibility that our imMKCLs could replace platelets for antibody testing. In addition, the small number of cells used in the test and the proliferation of the cells over a long period make testing convenient. This technique can likely be applied to other HPA antibody tests on GPIIb/GPIIIa, and will alleviate the difficulty of preparing platelets of low frequency antigens in the future.

P628 | Haplotypes analysis reveals the genetic basis of CD36 deficiency in Chinese population

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Background: CD36, glycoprotein IV, can be divided into two subgroups based on the expression of CD36 on monocytes. Previous studies have primarily focused on mutations discovered through Sanger sequencing. Few studies have investigated the CD36 haplotypes.

Aims: A long-read sequencing approach was used to directly phase CD36 haplotypes and analyze their phylogeny associated with type I/FNAIT disease.

Methods: All subjects were of Chinese origin and provided informed consent. The Guangzhou Blood Center CD36 Deficiency Bank provided 9 CD36 I and 13 CD36 II deficiency blood samples from healthy donors (76.19% men and 23.81% women, 34.19 ± 7.13 years). 7 blood

samples were collected from healthy donors at the Guangzhou Blood Center 71.43% men and 28.57% women, 35.57 ± 7.51 years. 4 blood samples were collected randomly from FNAIT patients or PTR CD36 type I deficiency patients with anti-CD36 antibodies between 2015 and 2022. The Coombs test was negative results. A primer panel was designed to amplify regions from exon2 to 3'utr of CD36 genes, including sequences from intronic regions. Seven sets of primers were designed to amplify amplicons with more than 1 kb overlap between each of them. The single-molecule real-time (SMRT) libraries were created using a one-step method according to the manufacturer's instructions. The sequencing enzymes and primers were bound to the final library using the Sequel Binding Kit 2.2 (Pacific Biosciences) and Internal Control Kit 1.0 (Pacific Biosciences). The output data was primarily analyzed using SMRTLink v10.1.0 software (Pacific Biosciences). Linkage disequilibrium analysis was performed on CD36, followed by construction of the phylogenetic tree and analysis of variants association. The output data were compiled and individual haplotypes, including gene crossover region haplotypes, were generated using an in-house software module.

Results: In this study, a total of 180 variants were identified spanning from 5UTR to 3UTR in the 37 samples. 12 of these mutations change the amino acid sequence. Four (c.220 C>T; c.329-330delAC; c.430-1 G>C; c.1006+2 T>G) can be considered as premature termination mutations. 7 of the variants are the non-synonymous mutations. One variant is 12 basepair (TATTGGTCAAGC) deletion from position 1228 to 1239 in exon 12 which lead to the deletion of Leu-leu-Val-Lys at amino acid position 391-394. One variations (nt-602A>C) were detected upstream of the start codon, which have been reported to reduce protein expression of CD36. All variants found in the haplotype sequences were used to identify haplotype blocks of CD36. 4 cases of FNAIT combined with hydatiform fetuses were observed, all of which contained the c.1-602-C - c. and EXON 5 329-330AC del a haplotype. Two cases showed one normal strand and one strand containing the c.1-602-C - c. and EXON 5 329-330AC del haplotype. It is important to note the haplotype of c.1-602-C - c. and EXON 5 329-330AC del in the development of FNAIT with hydatidiform fetuses. Fisher's exact test calculated the association between the variants and the phenotypes. A greater significance is observed between the 5' block and the Type I deficiency. Most haplotypes from Type I individuals cluster with the lineage carrying the nt-602 and other amino acid change mutations, such as the c.329-330delAC.

Summary / Conclusions: This study employed triple sequencing technology and genomic DNA as templates. The data show that the c.1-602-C - c. 329-330 del AC haplotype directly causes CD36 I deficiency.

P629 | Establishment and application of NGS method for the genotyping of HLA-I and HPA in the single tube based on multiplex PCR technique

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Background: Platelet transfusion refractoriness (PTR) has become one of the difficult problems in the clinical blood transfusion. Immune PTR is mainly related to HLA-I antibodies and HPA antibodies. At present, the most common method to solve the immune PTR is to screen the serological matched platelets by random cross-matching test. Another approach is to select donor with compatible genotype of HLA-I and/or HPA. This method requires a platelet donor bank with known data about the genotyping of HLA-I and HPA. It is necessary to establish a fast and reliable method for the genotyping of HLA-I and HPA, which could be used for building up a platelet donor bank. Currently, there are few methods for simultaneous detection of HLA-I and HPA. This study will provide a novel NGS method for the detection of HLA-I (HLA-A, -B and -C) and HPA (HPA-1 through HPA-6, HPA-15 and HPA-21) based on multiplex PCR technique, which could realize the detection of HLA-I and HPA genes in the same tube.

Aims: To establish a novel method for the genotyping of HLA-I and HPA simultaneously, which could help to build up a platelet donor bank.

Methods: First, ten pairs of specific primers for HLA-A, -B, -C, HPA-1 through HPA-6, HPA-15 and HPA-21 were designed and synthesized. The appropriate fragments were amplified in a single multiplex reaction by optimizing the concentration of different primers and the parameters of the PCR reaction. After a cleanup step using paramagnetic beads, the amplicon-library was prepared by TruePrep Flexible DNA Library Prep kit for Illumina. The quality of amplicon-library was detected by electrophoresis with Aligent 4200 and sequenced by Illumina Miseq platform. Finally, the genotyping of HLA-A, -B and -C was analyzed by TypeStream Visual Software version 3.0(TSV 3.0). The genotyping of HPA-1 through HPA-6, HPA-15 and HPA-21 was performed using CLC Main Workbench 23.0 software(CLC). To validate the accuracy of the developed method, commercial NGS kits for the genotyping of HLA-A, -B and -C and TaqMan real-time PCR methods in-house for the genotyping of HPA-1 through HPA-6, HPA-15 and HPA-21 were used to detect all the samples in parallel.

Results: A novel NGS method based on multiplex PCR was developed for genotyping HLA-I and HPA simultaneously in a single tube. The total of 386 samples were detected and the results of the genotyping for HLA-A, -B, -C, HPA-1 through HPA-6, HPA-15 and HPA-21 were obtained simultaneously, which is 100% consistent with the commercial NGS method for the genotyping of HLA-A, -B, -C and the TaqMan real-time PCR method in-house for the genotyping of HPA. In addition, four new HLA alleles including two HLA-A alleles and two HLA-B alleles in this study were also reconfirmed, indicating the developed method in our study is reliable and accurate. However, this method had less DNA input, reduced the testing time and saved the cost of testing when compared with the original method. Among 386 samples, 29 alleles for HLA-A, 60 alleles for HLA-B and 31 alleles

for HLA-C were detected. Moreover, HPA-1 through HPA-6, HPA-15 and HPA-21 are polymorphic observed in our study.

Summary / Conclusions: In our study, a novel NGS method based on multiplex PCR was established to detect HLA-I and HPA in a single reaction. This method is high-throughput, rapid and accurate, which could be applied to build up the platelet donor bank in a large scale.

P630 | The GPIa (p.Ala319Thr) variant investigated as a potential cause of fetal and neonatal alloimmune thrombocytopenia: A case study

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Background: Most cases of fetal and neonatal alloimmune thrombocytopenia (FNAIT) are caused by maternal production of alloimmune antibodies directed against fetal human platelet antigens (HPA) of paternal origin. Newborns with FNAIT present with abnormally low platelet counts ($<150 \times 10^9$ plt/L), which can be severe ($<50 \times 10^9$ plt/L) and lead to complications, including intracranial hemorrhage (ICH) and intrauterine fetal death (IUFD). Most HPAs are generated by single nucleotide polymorphisms (SNPs) affecting platelet surface glycoproteins. To this day, up to 41 such HPAs have been reported, but many cases of neonatal thrombocytopenia remain unexplained.

Aims: Héma-Québec's platelet Reference immunology laboratory identifies alloimmune HPA antibodies in patients throughout Québec (Canada). The anti-HPA antibody identification pipeline uses platelet antigen bead array (PABA) assays, monoclonal antibody immobilisation of platelet antigen (MAIPA) assays and directed genotyping for the most common HPA mismatches known to cause FNAIT. Additional analyses are conducted to elucidate cases of unexplained newborn thrombocytopenia referred to our laboratory.

Methods: MAIPA is a sensitive and versatile technique that detects anti-HPA antibodies by challenging the patient's serum with platelets of known genotypes. In cases of FNAIT, the mother's serum can also be challenged with paternal platelet glycoproteins. Sequence-specific oligonucleotide (SSO) and sequence-specific primer (SSP) techniques were used for directed genotyping of HPA SPN. Sanger-based typing (SBT) was used as a non-targeted approach.

Results: Our laboratory investigated a case of severe thrombocytopenia ($<40 \times 10^9$ plt/L) in a newborn from a mother known for multiple failed pregnancies (Gravida [G] 6, Para 4, Abortus 2, G5 IUFD at 36 weeks). Anti-HPA-5b antibodies were detected in the mother's serum using PABA and MAIPA. Moreover, the mother's serum weakly reacted with the GPIa-IIa complex of the father's platelets, as noted by MAIPA. No antibodies directed against the GPIIb-IIIa complex (anti-HPA-1a)—the most common antibody causing severe FNAIT—were detected by PABA or MAIPA. No mismatch was identified between

both parents for HPA-1 through 11 and HPA-15, as found by SSO. The HPA-5a/5a genotype was confirmed by SSP and SBT for both parents and the newborn. Furthermore, both parents' phenotype was HPA-5a-positive and HPA-5b-negative, as tested by MAIPA. No mismatch between the parents was identified for other known HPAs found on the GPIa (i.e., both were homozygous "aa" for HPA-13, 18 and 25). Given the positive reaction of the mother's serum against the father's GPIa-IIa glycoprotein complex, the exons of *GPIa* and *GPIIa* were sequenced for the mother, father, and newborn (after obtaining informed consent). This led to the identification of a heterozygous mutation (ITGA2 NM_002203: c.955G>A, p.Ala319Thr) within the father and newborn GPIa coding sequence. The mutation is located 215 amino acids away toward the N-terminus from the HPA-5 position and is not known to be associated with FNAIT.

Summary / Conclusions: This is the first report describing the occurrence of the mutation c.955G>A in a newborn with thrombocytopenia. More work is needed to establish whether this mutation is a new HPA. The identification of new HPAs and searching for maternal anti-HPA antibodies is crucial to identify high-risk pregnancies and initiate early mitigative treatments against FNAIT complications.

P631 | Selection of appropriate capture antibody and detergent is crucial for detection of anti-Nak(a) (anti-CD36) antibodies in Immunocomplex Capture Fluorescence Analysis (ICFA)

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Background: Monoclonal antibody-specific immobilization of platelet antigens (MAIPA) and immunocomplex capture fluorescence analysis (ICFA) are commonly used for detection of anti-Nak(a) antibodies (Abs). However, the assays are plagued by false-negative reactions due to competitive inhibition between epitopes of anti-Nak(a) Abs in some patients and commonly used CD36 monoclonal Abs (mAbs).

Aims: This study aimed to explore appropriate CD36 mAb selection and optimal solubilization conditions in ICFA.

Methods: We performed ICFA using six mAbs against CD36 (FA6-152, VM58, 5-271, 185-1G2, 4-62, and 1E8) and two detergents, TritonX-100 (TX-100) and n-octyl- β -D-thiogluconide (OTG). Panel platelets (plts) were isolated from healthy volunteers. S1 serum that had tested positive for anti-Nak(a) Ab in MAIPA, and S2 serum that had tested negative in MAIPA but positive in PIFT-FCM and MPHA (false negative in MAIPA) were assessed by ICFA. Plts were incubated with each serum, washed, and solubilized with TX-100 or OTG. Anti-Nak(a) Ab and CD36 antigen complexes were captured with each of the six CD36 mAb-conjugated Luminex beads. The captured complexes were detected with PE-labelled anti-human IgG, and their fluorescence intensities were measured using a Luminex system. Index values were calculated as the ratio of the adjusted fluorescence intensity of the sample serum to that of background beads. An index

value of ≥ 2.0 was considered positive. To evaluate epitope competition, plts were treated with FA6-152 or 1E8, and the reactivity of S2 serum against Nak(a) was measured by flow cytometry. To assess the effect of detergents, plts were incubated with S2 serum, and PE-labeled anti-human IgG was added. Plts were solubilized using TX-100 or OTG. Fluorescence intensity was measured after capturing the immunocomplexes using 1E8-conjugated beads.

Results: S1 serum tested positive in all assays with any combination of CD36 mAb-conjugated beads with TX-100 or OTG. The index values ranged from 27 to 157 with a mean of 81 in assays with TX-100, and from 228 to 544 with a mean of 417 in OTG assays. S2 serum tested positive only in assays using FA6-157 or 1E8-conjugated beads with low index values (3.4 ± 2.8 for FA6-157 and 2.6 ± 1.4 for 1E8) when using TX-100. However, when using OTG, S2 serum tested positive in 1E8 or 4-62-conjugated bead assays with high index values (294 ± 112 and 31 ± 15 for 1E8 and 4-62, respectively). In the epitope competition assay, the S2 serum reaction against Nak(a) was inhibited by $88 \pm 7\%$ with FA6-152, with no inhibition when using 1E8 ($2 \pm 25\%$) ($p < 0.01$). In the assay to assess the effect of detergents, significantly higher fluorescence intensities (4792 ± 1424) were observed when using OTG compared to TX-100 (245 ± 197) ($p < 0.01$).

Summary / Conclusions: 1E8 appeared to be more appropriate for capturing the immunocomplexes of CD36 and Abs in S2 serum than FA6-152, because 1E8 recognizes a different epitope on CD36 from the epitopes of Abs in S2. S2 tested positive in ICFA using 1E8-conjugated beads with higher index values when using OTG rather than TX-100. Further, S1 also tested positive with higher index values when using OTG rather than TX-100. These results suggested that 1E8 can capture the CD36-antibody complex without altering the steric structure of CD36 when using OTG. In summary, capture assays, such as ICFA, might be effective for screening anti-Nak(a) Abs and potentially for identifying new platelet antigens/abs, when appropriate capture mAbs and the optimal detergent are selected.

P632 | Development of reference material for platelet flow cytometry to support the quality and safety of transfused platelets

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Background: Platelet biology is notoriously difficult to standardise due to a high susceptibility to misinterpretation and differences in platelet responses from person to person. Flow cytometry is commonly used in transfusion medicine to analyse platelets. They are identified based upon their size, granularity and the presence of surface receptors. However, the small platelet size, sensitivity to activation and subsequent shape changes can lead to difficulties separating platelets from cellular debris and subjective gating techniques between labs and users. Additionally, due to the demand for platelets in transfusion medicine continuing to rise, there has been an increased research effort for the

development of novel platelet products. The emerging technology presents added challenges to the assessment of the quality and safety of the platelets (e.g. the presence of nuclear contamination in platelets generated from human pluripotent stem cells (hPSCs)). Therefore, there is an urgent need for physical platelet standards to ensure the safety and functionality of emerging products and improve consistency of data. A globally recognised reference material will aid researchers and the clinical community to characterise platelet populations and provide meaningful comparable results between laboratories and equipment.

Aims: Develop a WHO International Standard for Platelet Flow Cytometry. The main parameters to be evaluated by flow cytometry are size and granularity, viability, platelet expression markers and contamination with nucleated cells.

Methods: Pooled platelet donations were stained with a viability dye, fixed with formaldehyde, and combined with fixed *in vitro* produced megakaryocytes. Following trehalose loading, 1 mL of the material was filled into ampoules and lyophilised using a 4-day freeze-drying cycle. After lyophilisation, ampoules were resuspended in PBS, stained with anti-CD41a, anti-CD61, and anti-CD42b antibodies and assessed by flow cytometry. Nuclear 'contaminating' cells were observed by DAPI staining. An accelerated degradation study (ACD) was performed with ampoules stored at -70, -20, +4, +37, +45 and +56°C for six months. Additionally, stability after reconstitution of the material was performed with ampoules stored +4°C for up to one month.

Results: Flow cytometry analysis showed no loss in percentage of viable cells following lyophilisation (98.6%) in comparison to pre-lyophilisation (98.2%). Forward and side scatter consistent with platelets and expression of CD41a/CD61 and CD42b was observed. DNA 'contaminating' cells could be identified post-lyophilisation with DAPI staining. The 6-month ACD study showed minimal loss of viable cells. Additionally, stability after reconstitution showed minimal changes of the candidate reference material for up to 1 week with storage at +4°C.

Summary / Conclusions: We have produced the first candidate WHO reference standard for platelet flow cytometry, intended to provide quality control of platelets by measuring receptor expression, viability/membrane integrity, and nuclear contamination from hPSCs that will be present in *in-vitro* generated platelets. Standardisation is important to reduce inter- and intra- lab variation and is vital part in the measurement of product safety. This study demonstrates a lyophilised preparation suitable as a candidate standard for platelet flow cytometry, which will provide the calibration of instruments used for comparing samples using the markers described above.

P633 | Why platelet crossmatch is so important? Third report of a Caucasian mother with anti-HPA-4b alloantibodies discovered in a severe neonatal thrombocytopenia context

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Background: Fetal and Neonatal Alloimmune Thrombocytopenia (FNAIT) results from maternal platelet alloimmunization against paternal antigens inherited by the fetus, most often due to Human Platelet Antigen (HPA)-1 system in Caucasians.

Aims: We investigated in 2023, a 30-year-old Caucasian woman Gravida 2 Para 1 who gave birth at 35 weeks of gestation to a male (body weight 2210 g) without signs of bleeding. A severe thrombocytopenia (platelet count at 3 G/L) was discovered incidentally few hours after delivery in the context of the management of a respiratory distress. The neonate recovered after only one platelet concentrate transfusion and normalized his platelet count at Day 5. The neonate left the hospital at Day 15 with a corrected term of 37 WG + 1 D.

Methods: FNAIT investigation was performed according to guideline recommendations. Platelet genotyping carried out by multiplex PCR (BioArray Solutions, Immucor). Maternal serological investigation included Monoclonal Antibody-specific Immobilization of Platelet Antigens method (MAIPA, Complete MAIPA kit apDia) and Luminex technology (PakLx TM Assay, Immucor GTI Diagnostics).

Results: Direct MAIPA against the glycoprotein (GP) Ia-IIa, IIb-IIIa and Ib-IX complexes was negative. Maternal serum was negative for antibodies against GPIa-IIa, IIb-IIIa and Ib-IX complexes when screened by indirect MAIPA (positive results when OD > 0.200). Only Class I anti-HLA antibodies were detected (O.D = 3.932). Surprisingly, the crossmatch of maternal serum with paternal platelets revealed strong positive reactions against GPIIb-IIIa (OD = 3.928) suggesting the presence of a rare alloantigen on the paternal platelets. Parental and newborn genotyping pointed out an HPA-4 incompatibility between the mother and the newborn and the father. Luminex technology with Pak Lx assay identified an anti-HPA-4b alloantibody.

Summary / Conclusions: We described the third case of anti-HPA-4b alloantibody discovered in a Caucasian mother. This case strengthens the need for reference laboratory to genotype a panel of HPA alleles reflecting local genetic population diversity and to perform systematically a crossmatch with fresh paternal platelets in clinical suspected cases of neonatal alloimmune thrombocytopenia for the identification of rare or private human platelet antigens. An appropriate management for a new pregnancy could be proposed to parents involving a prenatal diagnosis to determine the fetal HPA status and an antenatal maternal treatment in case of fetal incompatibility.

P634 | Performing fetal platelet genotyping on maternal plasma is not necessary in all indications—first retrospective study of French practices and neonate characteristics

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Background: Fetal and/or neonatal thrombocytopenia (FNAIT) is due mainly to a feto-maternal incompatibility in four human platelet antigen -1, -3, -5 and -15 (HPA) systems in Caucasians. Determination of fetal platelet genotyping is essential to manage feto-maternal alloimmunization in case of incompatible fetuses for an early evaluation of the risk of pregnancy to introduce maternal antenatal treatment by intravenous immunoglobulin (IVIg).

Aims: From 10/2020 to 06/2022, a retrospective study of French practices of non-invasive prenatal testing (NIPT) for fetal platelet genotyping was conducted regarding to laboratory requirements and international recommendations to introduce maternal antenatal treatment by IVIg. We also investigated the neonate characteristics especially platelet count (PC) at birth according to the introduction or not of antenatal treatment.

Methods: Droplet digital PCR technology was used to determine fetal platelet genotyping in HPA-1, -3, -5 and -15 systems according to the parental incompatibilities. Clinical data were collected through a laboratory form associated to the samples and/or maternal and neonates hospitalization reports sent by clinicians. In 101 NIPT indications sent to the laboratory, 90 from 89 pregnant women were eligible for this study since fetuses/ neonates information were available.

Results: Eighty-nine women were included; 55.5% of them were alloimmunized (66% by anti-HPA-5b, 10% by anti-HPA-1a alloantibodies). FNAIT or suspected FNAIT history (53.3%) were the main NIPT, intracranial hemorrhage or cerebral abnormalities background or discovered during pregnancy represented 20%. One emerging indication consisted in the incidental finding of anti-HPA-5b alloantibodies during pregnancy (10%). Approximately half of the first NIPT determinations was performed after 22 weeks of gestation (WG) not allowing antenatal treatment (laboratory requirements from 12 WG). Platelet count was performed in 88.9%. Fifty-five fetuses with platelet count available were incompatible in one or more HPAs (65.5%). Thirteen neonates (16.2%) were thrombocytopenic at birth. Severe thrombocytopenia was only observed in one incompatible neonate in HPA-1 presenting petechiae (PC at 6 G/L) due to anti-HPA-1a maternal alloimmunization not treated during pregnancy. No difference was found in PC regarding fetal platelet genotyping indications concerning incidental discovery of anti-HPA-5b during pregnancy vs other indications, HPA fetal status and maternal antenatal treatment. Overall, the 55 incompatible fetuses were found in 38 immunized mothers, but only 23 were treated by IVIg (60.5%). Then, thirty incompatible fetuses did not benefit from antenatal treatment while 15 mothers were immunized by anti-HPA-1a, -1b, -5b or -15b alloantibodies.

Summary / Conclusions: This is the first retrospective study of French practices of NIPT. The relevance of the extension of the indications has to be evaluated. Laboratory requirements and international recommendations were not systematically followed in case of incompatible fetuses in immunized women including anti-HPA-1a alloimmunization, but also the introduction of antenatal treatment in anti-HPA-5b alloimmunization did not follow well-defined criteria. A prospective study is needed to evaluate fetal platelet genotyping indications according to the alloimmunization and the criteria of introduction of antenatal treatment for improving future guidelines since no consensus currently exists between the different expert centers.

P635 | Detect rare allele frequency of human platelet antigens in Taiwanese population through Taiwan Biobank dataset

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Background: Human platelet antigens (HPAs) are alloantigens due to the polymorphic amino acid sequences of membranous glycoproteins on platelets, and their frequencies vary between populations. Incompatibility in platelet antigens during blood transfusion or between mother and fetus during pregnancy may induce alloantibodies and cause thrombocytopenia and hemorrhagic disorders. There are 35 recognized HPAs located on 7 glycoproteins. Our previous study has shown the genotype frequencies of HPA 1 to 6 and 15 based on a cohort of 998 blood donors from Taipei blood center. Since the accuracy of rare variant calling in Taiwan Biobank (TWB) single nucleotide polymorphism (SNP) arrays is proved to be improved through quality-control procedure, the array data could be used to evaluate the allele frequencies and bring us more about the genetic variations of HPAs inside the population.

Aims: To determine whether using the TWB SNP arrays to detect rare variants (with minor allele frequency less than 1%) is adequate to understand the genetic background of HPAs in the Taiwanese population.

Methods: Genotyping data for 147,882 individuals from Taiwanese population (27,719 typed on the TWBv1 custom array and 120,163 on the TWBv2 array, with 1463 typed on both) was obtained from the Taiwan Biobank with the approval by the Ethics and Governance Council of Taiwan Biobank. The TWB SNP arrays dataset with the plink format was filtered with the 35 HPA variants and processed

P635 - Table 1.

HPA	TWB MAF	TBSF MAF	Global MAF†
HPA-1	0.002	0.005	0.089
HPA-2	0.037	0.035	0.091
HPA-3	0.432	0.442	0.382
HPA-4	0.002	0.003	0.003
HPA-5	0.016	0.015	0.104
HPA-15	0.472	0.463	0.511
HPA-21	0.007	NA	<0.001

† Global MAF data was referred from dbSNP database.

NA: Not available.

using the PLINK v1.9 software, then the genotypes and allele frequency of variants as well as individual gender were analyzed by Microsoft Excel and Power BI.

Results: Eighteen of the 35 HPA variants were found in the TWB SNP dataset. The minor allele frequencies (MAF) of HPA 1 to 5 and 15 were close to our previous data (TBSF), even the rare variants (Table 1). Moreover, HPA 21, the relatively common platelet antigen among Asians, had a MAF about 0.007 in the study population, which was also approximate to the publication (JA Peterson, Transfusion, 2012). And the MAF of the other 11 HPAs, HPA 10, 12, 16-19, 22-25, and 30, were equal to or less than 0.001. Furthermore, the homozygous type of minor allele among studied subjects found in HPA 2, 4, 5, and 21 represented that these rare types of platelet antigens were existent in Taiwanese population while the proportions across gender were similar.

Summary / Conclusions: This study showed an approach to evaluate the allele frequency of HPAs through SNP arrays. There were 18 variants relevant to the recognized HPA variants found in TWB SNP arrays, and the minor allele frequencies of 7 variants of them, even the variants with known frequency less than 1%, were close to our previous study using a cohort of 998 blood donors from Taipei blood center and the previous publication. Many of the HPA variants and rare types of platelet antigens in Taiwanese population shown in this study were first analyzed, and the genotype frequencies were different from those of the global populations. Therefore, this study provided insights into the difference of HPA genetic background among populations and gave us more about the profiling recommendation in Taiwanese population.

P636 | Detection of C3d sensitized platelets by a novel microcolumn gel based competitive immunoassay

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Background: Several immunological assays are required prior to clinical blood transfusion, including tests for red blood cell-related antibodies and complements, as well as platelet-related antibodies. However, assays concerning platelet-related complement, such as C3d, are not presently mandatory. C3d represents the final degradation product of complement component C3. Platelets bonded with C3d are more susceptible to phagocytosis by splenic macrophages, leading to adverse effects post-blood transfusion, including platelet transfusion refractoriness (PTR). To mitigate these adverse effects and conserve blood resources, a rapid and effective method for detecting C3d-sensitized platelets is urgently needed.

Aims: To establish a rapid and effective detection method for C3d-sensitized platelets using microcolumn gel-based competitive immunoassay, providing a reference basis for precision transfusion and treatment.

Methods: The microcolumn gel-based competitive immune assay is performed as follows: (1) Preparation of C3d-coated red blood cells (RBCs) by incubating whole blood with 9.24% sucrose solution and treating with trypsin. (2) Division of patients' blood samples into two parts: one for making C3d-coated platelets using similar methods as for C3d-coated RBCs, and the other left untreated for detection. (3) Incubation of C3d-sensitized or untreated platelets with monoclonal antibody against C3d, followed by harvesting of platelet-free supernatants after centrifugation. (4) Addition of C3d-sensitized RBCs and supernatants from previous steps to microcolumn gels, along with monoclonal antibody at the same dilution as negative control. (5) Observation of results by naked eye. Samples containing C3d-sensitized platelets exhibit reduced agglutination compared to the negative control group, while platelet-free groups with C3d sensitization show similar agglutination to the negative control group.

Results: This method was applied to 22 clinical samples, with 6 testing positive for C3d-sensitized platelets and 16 testing negative.

Summary / Conclusions: We have established a rapid and effective detection method for C3d-sensitized platelets using microcolumn gel-based competitive immunoassay, suitable for further clinical evaluation.

Acknowledgement: Thanks Mr. Yong Li, Suzhou Institute of Biomedical Engineering and Technology Chinese Academy of Sciences, for guiding this study.

P637 | Glycoprotein V is a new target for platelet autoantibodies in Immune Thrombocytopenia (ITP)—the experience of the Platelet Immunology Reference Laboratory of Rennes (France)

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Background: Anti-platelet autoantibodies against GPIbIX, Ialla and IIbIIIa play a major role in the destruction of platelets in Immune Thrombocytopenic Purpura (ITP).

Aims: Recently, Vollenberg et al. described the presence of anti-GPV autoantibodies among a cohort of patients (Glycoprotein V is a

P637 - Table 1.

	Direct MAIPA test (172)	Indirect MAIPA test (217)
Negative	137 (79.7%)	180 (82.9%)
Positive for GPIbIX and/or Ialla and/or IIbIIIa, and Positive for GPV	15 (8.7%)	6 (2.8%)
Positive for GPIbIX, Ialla and/or IIbIIIa but GPV Negative	4 (2.3%)	25 (11.5%)
Negative for GPIbIX, Ialla and IIbIIIa but GPV Positive	16 (9.3%)	6 (2.8%)

relevant immune target in patients with immune thrombocytopenia; Haematologica, 2019).

Methods: the MAIPA method was adapted to search for anti-GPV autoantibodies, then tested on a cohort of patients between January 2022 and May 2023.

Results: Anti-GPV autoantibody detection was negative in 180 patients (82.9%), and positive for 37 patients (17.1%). In 2.8% of patients, anti GPV autoantibody was the sole specificity detected. (2) Fixed autoantibodies were negative for 79.7% of patients. Interestingly, anti GPV autoantibody was the sole specificity identified for 9.3% of the patients.

Summary / Conclusions: the specificity of platelet autoantibody testing for ITP patients is improved by the new GPV target.

P638 | Abstract withdrawn

P639 | Virtual platelet cross-matching for patients with immune platelet transfusion refractoriness in a single blood service, China

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Background: Platelet transfusion is an important treatment method for the patients with thrombocytopenia or impaired function of platelets in clinical blood transfusion. However, multiple platelet transfusions may cause immune platelet transfusion refractoriness, which was mainly due to HLA alloimmunization. Choosing the donor platelets that crossmatch with the patients will help solve the problem of platelet transfusion refractoriness. But it is necessary to establish a HLA-typed platelet donor bank and provide the crossmatch strategy.

Aims: Establish a HLA-typed platelet donor bank and develop an information system for virtual platelet cross-matching based on cross reactive groups (CREGs) and /or avoidance donor specificity antibody (DSA) method in a single blood service, China, then applicate the HLA-typed platelet donor bank to provide crossmatch-compatible platelets for patients with platelet transfusion refractoriness.

Methods: HLA-A and HLA-B loci were genotyped using the PCR-sequence based typing or next generation sequencing methods, and HLA antibody was detected using Luminex technology. Blood donors were recruited to join the HLA-typed platelet donor bank in our blood center, and collected the specimens from blood donors for testing with informed consent. An information system for HLA-typed platelet donor bank were constructed, which can choose the suitable donors using the HLA antigen CREG or avoidance DSA model. The patient was done the HLA genotype, and also tested the HLA antibody as needed. If the HLA antibody is positive, it will use the avoidance DSA method with CREG together.

Results: A HLA-typed platelet donor bank with a capacity of over 25,000 blood donors was established. Among them, 8001 donors who has participated in platelet blood donation as before. An information system for HLA-typed platelet donor bank have been also constructed, including management for HLA-type data of the donor, patient information management, application for virtual platelet cross-matching,

CREG match and avoidance DSA model. Virtual platelet crossmatching for the patients can be automatically performed, based on patient information to select the most suitable platelets using the information system. The total crossmatch compatible platelets for the patients were 651 times in 2023. Retrospective analysis 55 times crossmatch platelets with 12 times for A and B1U level, 36 times for B1X and B2UX level and 7 times only avoiding HLA-I antibodies, it was showed that the 24-h corrected platelet increase index (CCI) is 9.7, confirming the effectiveness of the crossmatch-compatible platelets.

Summary / Conclusions: A HLA-typed platelet donor bank in single blood center in China was established. Virtual platelet cross-matching for patients to selection compatible platelets has been automatically operated using a specialized information system.

P640 | Analysis of donor-specific HLA antibodies profiles in platelet refractory patients and application to platelet donors selection

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Background: For immune-mediated Platelet transfusion refractoriness(iPTR), donor-specific HLA antibodies(DSA) represents a Highly sensitised condition associated with increased fatal complications and delayed therapeutic interventions. However, the specificities profiles of HLA antibodies among these patients is not fully known and that can be used as predictors for platelet donor selection are poorly reports.

Aims: To analyze the specificity of HLA class-I alloantibodies in the iPTR patients of Chinese. The transfusion outcome of acceptable HLA-mismatch strategy according to DSA specificity were also be evaluated.

Methods: All enrolled patients were iPTR with HLA alloantibodies positive (HPA and CD36 antibodies were not included in this study). The specificity of HLA-I antibodies were identified in the patient plasma by Luminex technique and analyzed with computer software(HLA Fusion). The strength of HLA antibody reactivity was divided according to mean fluorescence intensity(MFI).The HLA-A and -B genotyping of both patients and donors were obtained by PCR-SBT method with our previous reports.The outcome of transfusions was assessed by the 24 h corrected count increment (CCI), more than 4.5 was considered a successful response.

Results: A total of 203 iPTR patients received 1532 single-donor apheresis Platelet transfusion between October 2019 and May 2023. Of those HLA antibody-positive patients, the most common HLA class-I alloantibody phenotypes in iPTR patients were HLA-A*25:01 (52.2%),-A*32:01(51.2%), -A*68:01 (50.2%), -B*13:02 (64.0%), -B*45:01 (62.6%), -B*57:01(62.6%) and -C*17:01(45.8%), -C*15:02 (32.5%), -C*02:03(32.5%).The 643 random PLT units (in 89 of 203 iPTR patients)and 358 acceptable HLA-mismatched based on DSA PLT units(131 in of 203 iPTR patients) were administered with a mean 24hCCI of 6.13 ± 7.88 and 11.87 ± 8.72 ,respectively. The DSA matching resulted in significantly higher CCI compared to transfusions with random platelet units ($p < 0.001$).

Summary / Conclusions: platelet donor selection based primarily on the levels of donor-specific HLA antibodies should be taken into account. The effect of HLA-compatible PLTs with DSA-based matching strategies predicted preferable transfusion outcome.

P641 | Transfusion of HLA-compatible platelets in patients with immune platelet refractoriness—experience in a tertiary-care hospital

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Background: Platelet transfusion refractoriness (PTR) is a major concern in patients with severe thrombocytopenia and may lead to significant bleeding complications. Most cases are due to non-immune causes, but up to 20% are of immune origin, with antibodies against class I HLA antigens being the most frequent. In clinical settings, this situation is managed by selecting HLA-compatible platelets, HLA-matched platelets or cross-matched platelets.

Aims: The aim of this study is to compare transfusion yield in patients presenting with immune PTR who received HLA-compatible platelets versus standard platelet transfusion.

Methods: We conducted an observational and retrospective study on patients with immune PTR who received HLA-compatible platelets in Hospital Universitario La Fe between September 2023 and January 2024. We included information on age, sex, body surface, pre and post-transfusion platelet count and relevant clinical information. Immune PTR diagnosis was confirmed by the detection of anti-HLA antibodies using class I single antigen bead array assay by Luminex technology. HLA-compatible platelets were defined as platelets obtained from donors who lack HLA antigens against which the patient is alloimmunized. For each HLA-compatible platelet transfusion we selected a control unmatched platelet transfusion for the same patient during the same hospitalisation episode. Post-transfusion platelet count was performed up to 24 h after the transfusion. Statistical analysis was performed using SPSS software.

Results: A total of 71 units of HLA-compatible platelets were issued to our hospital in the study period for six different patients presenting immune PTR. All patients were female, and their median age was 59 (ranging from 18 to 65). All were hospitalised on the Haematology ward, being the reasons for admission the administration of chemotherapy for acute leukaemia, neutropenic fever and allogeneic stem cell graft. None of them presented significant causes of platelet hyperconsumption except for one patient who had persistent severe gastrointestinal bleeding (this patient alone accounted for 52% of all the transfusional episodes in our study). Of the initial 71 HLA-compatible units, 30 (42%) were not administered to their target patients (29 were administered to other patients and one passed its shelf life). Overall,

41 HLA-compatible platelet units were transfused to their target patients. We selected 39 unmatched platelet transfusion events as our control group corresponding to the same patients and during the same clinical episode. Statistical analysis revealed the mean increase in platelet count after HLA-compatible platelet transfusion was of $12.18 \times 10^3/\mu\text{L}$, whilst that of unmatched platelets was of $2.94 \times 10^3/\mu\text{L}$ ($p = 0.02$).

Summary / Conclusions: Platelet transfusion refractoriness of immune origin represents a challenge for the treatment of patients with severe thrombocytopenia. One strategy to overcome this difficulty is the selection of HLA-compatible platelets, which entails significant effort for blood centres. In this study we observe that yield of HLA-compatible platelets is significantly higher than that of standard platelets, thereby justifying their use in our clinical setting. However, the high percentage of HLA-compatible platelets transfused to other patients highlights the existing difficulty in scheduling this directed transfusion therapy.

P642 | Luminex based HPA-antibody detection and identification

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Background: Human platelet antibodies (HPA) have been involved in fetal/neonatal alloimmune thrombocytopenia (FNAIT), immune thrombocytopenia (ITP), platelet transfusion refractoriness (PTR) and post-transfusion purpura (PTP). In addition to HPA, antibodies to human leukocyte antigen class I (HLA) may play a role in immune-mediated platelet disorders, particularly those associated with transfusion. Luminex represents the most sensitive method in the detection and identification of HPA antibodies against antigens within the HPA-1, -3 and -4 systems (GPIIb/IIIa), HPA-2 (GPIb/IX) and HPA-5 (GPIa/IIa) systems along with the detection of antibodies against GPIV and HLA class I antigens.

Aims: To determine the frequency and specificity of HPA antibodies as well as the presence of HLA class I antibodies in patients with thrombocytopenia.

Methods: We conducted a retrospective analysis of HPA antibody testing results using the Luminex bead assay LIFECODES® Pak Lx™ Assay (Immucor GTI Diagnostics, Inc., USA), which were performed at the Clinical Hospital Center Rijeka, Croatia from February 2019 to December 2023.

Results: A total of 578 serum samples (566 samples from patients with suspected immune platelet disorders, 12 samples for FNAIT diagnostics) were tested with Luminex bead assay over a 5-year period. Of the 566 samples tested for immune-mediated thrombocytopenia, 77 (13.60%) were positive. In 49 (63.64%) samples, only HPA antibodies were detected, in 20 (25.97%) samples, only HLA I antibodies were present and 8 (10.39%) samples were positive for both, HLA and

HPA antibodies. The most common HPA antibody specificity was anti-GPIIb/IIIa (31; 54.39%), anti-GPIa/IIa (10; 17.54%), anti-5b (10; 17.54%), followed by anti-GPIb/IX (3; 5.26%), anti-HPA-1b (2; 3.51%) and anti-HPA-5a (1; 1.75%). Of the 12 samples tested for FNAIT, 7 (58.33%) were positive. HPA antibodies were detected in 2 (28.57%) samples (anti-HPA-1a specificity), HLA class I in 4 (57.14%) and HLA with HPA (anti-HPA-5b specificity) antibodies in 1 (14.29%) sample.

Summary / Conclusions: Experience in our center has shown that HPA and/or HLA antibodies are detected in a small proportion of patients with low platelet counts. In immunized adult patients, HPA antibodies were mainly directed against GPIIb/IIIa antigens. In the FNAIT diagnosis, the HPA antibodies detected were anti-HPA-1a and anti-HPA-5b specificities, which is in accordance with the literature data. In our study, HLA antibodies were detected in half of antibody-positive patients with FNAIT, although the role of HLA antibodies in the development of neonatal thrombocytopenia is still being investigated and additional studies are required. Luminex is a powerful tool that allows the simultaneous detection of HPA and HLA class I antibodies, making it an important method in the diagnosis and treatment of immune-mediated platelet disorders.

P643 | Establishment a quantitative real-time fluorescence PCR method for HPA 1-6 w, 15 and 21 w genotyping based on MGB probe and its application

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Background: Human platelet alloantigens (HPA) mismatch can induce the alloimmunization to produce antibody by transfusion or pregnancy pathway, and then cause the platelet transfusion refractoriness (PTR) and neonatal alloimmune thrombocytopenia (NAIT). Therefore, HPA genotyping and selection match platelet transfusion will be helpful for their diagnosis and treatment. Currently, HPA genotyping methods have been widely used in the establishment of HPA-typed platelet donor bank and the diagnosis of these diseases. HPA distribution is difference in the population, and previous studies have shown that HPA 1-5, 6w, 15 and 21w are polymorphic and other HPAs are nearly aa genotype in the Chinese population. Therefore, it is worth to establish a rapid and efficient method for genotyping the HPA 1-5, 6w, 15 and 21w.

Aims: Establishment a quantitative real-time fluorescence PCR (qRT-PCR) method for HPA 1-5, 6w, 15 and 21w genotyping based on the Minor Groove Binder (MGB) probe. It can achieve rapidly diagnosis the HPA genotypes and will be used for HPA-typed platelet donor bank which will be used for selection match platelet transfusion.

Methods: According to the genome sequence of the platelet glycoprotein genes, eight pairs of primers (each pair for HPA 1-5, 6w, 15 and 21w respectively) and 16 specific MGB probes (two probes for a and b alleles in one HPA respectively) were designed using a

software. One pair of primers and corresponding a probe (HPA-a or HPA-b alleles) were added to a well respectively. After optimizing the conditions, the same polymerase chain reaction (PCR) condition was used as below. 95°C for 3 min, 95°C for 20 s, 95°C for 30 s, and 50 cycles, then determine the genotypes based on the amplification curve using the software. 3871 specimens from platelet donors who have been take part in the HPA-typed platelet donor bank were collected and extract their genomic DNA for test. 200 specimens for them were also detected using the PCR-sequence based typing (PCR-SBT) as our previous established. The consistency of the results of the two methods was compared.

Results: The qRT-PCR detection can be completed within 1.5 h. The HPA 1-5, 6w, 15 and 21w genotypes in 200 specimens of the qRT-PCR are completely consistent with those of the PCR-SBT method. A HPA-typed platelet donor bank in our blood center was established with 3871 donors, and the allele frequencies distribution of HPA 1-5, 6w, 15 and 21w are as follows: HPA-1a 0.9954, HPA-1b 0.0046; HPA-2a 0.9510, HPA-2b 0.0490; HPA-3a 0.5790, HPA-3b 0.4210; HPA-4a 0.9987, HPA-4b 0.0013; HPA-5a 0.9855, HPA-5b 0.0145; HPA-6wa 0.9821, HPA-6wb 0.0179; HPA-15a 0.5420, HPA-15b 0.4580; HPA-21wa 0.9925, HPA-21wb 0.0075.

Summary / Conclusions: A real-time fluorescence PCR method for HPA 1-5, 6w, 15 and 21w genotyping based in MGB probe and a HPA-typed platelet donor bank in single blood center were successfully established, and HPA allele frequencies distribution in the Chinese population was also obtained.

P644 | Automated method for anti-blood group antibody titration in platelet concentrates

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Background: Isoagglutinin (anti-A and anti-B) titration can be necessary to perform in some clinical scenarios as ABO incompatible hematopoietic stem cell transplantation and solid organ transplantation. In case of blood components transfusion containing ABO incompatible plasma, as platelets (PLT), it is necessary to know the isoagglutinin titer in order to assess the safety of the procedure. There are different methods to determine the isoagglutinin titers as tube and column agglutination, often performed as a manual method. Automation of this test simplifies the procedure and improves reproducibility. We have recently validated an automated method to perform isoagglutinin titers when needed.

Aims: To assess the feasibility of isoagglutinin titration of platelet concentrates (PLT) using Ortho Vision autoanalyser.

Methods: Samples from platelet concentrates of different ABO groups were taken in closed system and introduced in Ortho Vision analyser to perform isoagglutinins titration, anti-A IgM and IgG, and anti-B IgM and IgG. We previously have validated the automated method by comparing to manual method. Agglutination was

P644 - Table 1. Isoagglutinin titration according to the ABO blood group.

ABO group	Anti-A IgM	Anti-B IgM	Anti-A IgG	Anti-B IgG
O (n = 62)	32 (2-512)	32 (2-2048)	64 (4-512)	64 (4-2048)
A (n = 72)		32 (2-2048)		4 (0-64)
B (n = 20)	6 (2-64)		4 (2-8)	
p	<0.001	<0.001	<0.001	<0.001

P644 - Table 2. Isoagglutinin titration according to the PLT product. *SDA: single donor apheresis.

	Anti-A IgM	Anti-B IgM	Anti-A IgG	Anti-B IgG
Pooled (n = 71)	16 (2-128)	8 (2-256)	16 (2-256)	8 (0-512)
SDA* (n = 53)	32 (2-512)	16 (2-2048)	64 (4-512)	8 (0-2048)
Inactivated (n = 31)	64 (8-28)	16 (2-128)	64 (8-128)	16 (2-256)
p	0.061	0.576	0.019	0.611

considered positive when agglutination was $\geq 0.5+$. Samples from different platelet products (apheresis, pooled from buffy coat, and pooled from buffy coat and inactivated) and different ABO blood groups were collected. All PLT products were suspended in additive solution.

Results: We collected samples from 155 platelet products: 71 pooled from buffy coat (23 O, 29 A, 19 B), 53 from single donor apheresis (SDA) (29 O, 24 A) and 31 pooled from buffy coat and inactivated (11 O and 19A). Results of isoagglutinin titration according to the ABO group and type of PLT product are shown in table 1 and 2, respectively. Results are shown as median and range. Group O PLT had the higher isoagglutinin titers as expected, some of them showing very high titers (2048). There were no differences in median of isoagglutinin titers for each group according to the PLT products.

Summary / Conclusions: The automation of isoagglutinin titer is feasible, simplifies the technique and reduces the staff work load. The availability of this test is highly recommended in cases of ABO incompatible HSCT, incompatible ABO solid organ transplantation and transfusion of ABO incompatible plasma containing blood products.

P645 | Abstract withdrawn

P646 | Case report—neonatal alloimmune thrombocytopenia and HLA class antibodies

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Background: Fetal-neonatal alloimmune thrombocytopenia (FNAIT) is determined by maternal alloantibodies against paternal antigens present in fetal platelets. Platelets express HLA class I and human platelet specific antigens (HPA), although antibodies to HLA class I are often

detectable in pregnant women, FNAIT is considered to be primarily associated with antibodies to HPA. Cases in which FNAIT is determined by HLA class I antibodies are rare and their role remains questionable.

Aims: In the present study we describe a case of FNAIT in a firstborn infant determined exclusively by anti-HLA class I antibodies.

Methods: The samples arrived from the Neonatal Intensive Care Unit (NICU) and were subjected to the detection of antiplatelet antibodies Direct Test (Capture P Ready Screen, Immucor), carried out in the mother and the newborn, and to the determination of antiplatelet antibodies Indirect Test with methodology Luminex Pak Lx xMPAP (Immucor). The child, born at term, was hospitalized after birth in the NICU due to perinatal asphyxia, underwent hypothermia treatment and mechanical ventilation for 72 h due to severe hypovolemia, presented with anemia for which he was transfused with 2 units of concentrated red blood cells filtered zero Rh negative and 1 unit of Fresh Frozen Plasma. After 48 h of life, a significant thrombocytopenia was detected with a reduction in the count (8×10^9 /L), the appearance of petechiae and the presence of intracranial hemorrhage. The mother did not have thrombocytopenia, the antenatal TORCH serology was normal.

Results: The tests confirmed the suspicion of FNAIT with the search for antiplatelet antibodies Direct Test positive in the newborn, and negative in the mother. The identification test of the anti HPA and anti HLA alloantibodies showed positivity for class I HLA antibodies, in the absence of anti HPA antibodies. Genotyping was performed both in the parents and in the child for diagnostic investigation. the child received intravenous immunoglobulin (1 g/kg) from day 3 to day 9, and was transfused with platelet pool, this approach resulted in an increase in platelet count equal to 138×10^9 /L. The child was followed up 3 months after hospital discharge.

Summary / Conclusions: The presence of anti-HLA class I antibodies should be considered a potential cause of FNAIT in the neonate with severe thrombocytopenia and intracranial haemorrhage comparable to FNAIT determined by antibodies against platelet antigens. Few cases of pathology determined only by anti-HLA class I antibodies are

reported in the literature, however the presence of antibodies and neonatal thrombocytopenia was associated with significant perinatal morbidity, and these data were particularly evident for firstborns, as in our case. Our studies have not been able to address the question of causality, and it is not clear whether the presence of HLA class I antibodies is an epiphenomenon in pregnancies in which the newborn develops thrombocytopenia, therefore this question should be addressed in one larger prospective study.

P647 | An assessment of the PF4-dependent P-selectin expression assay in comparison to the heparin-induced platelet activation assay for the laboratory diagnosis of heparin-induced thrombocytopenia

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Background: Heparin-induced thrombocytopenia (HIT) is a potentially fatal reaction which occurs during treatment with the anticoagulant heparin and is characterized by the formation of IgG antibodies against heparin/platelet factor 4 (PF4) complexes, causing thrombocytopenia and/or thrombosis. A prompt and correct diagnosis is crucial to ensure adequate alternative treatment. Heparin enzyme-linked immunosorbent assays (ELISA) are widely used but lack specificity and are usually combined with platelet activation assays, such as the heparin-induced platelet activation assay (HIPAA). Unfortunately, these platelet activation assays are labor-intensive and not widely available. The flowcytometry-based PF4-dependent P-selectin expression assay (PEA) has previously been proposed as a relatively easy to perform nonradioactive assay with high accuracy for diagnosing HIT.

Aims: We assessed if the PEA could replace the more laborious HIPAA in the laboratory diagnosis of HIT.

Methods: The PEA was conducted on samples of 23 patients positive in both the anti-PF4/heparin ELISA and HIPAA, and 26 patients negative in the anti-PF4/heparin ELISA. Subsequently, samples from 195 suspected HIT patients of whom the ELISA and HIPAA results were known were blindly tested in the PEA.

Results: The PEA showed a high ability to identify HIT patients (AUC:0.87 (95% CI 0.77 - 0.97, $p \leq 0.0001$)). Among 118 HIPAA-positive patients, 82 were PEA-positive. Additionally, 55 of 77 HIPAA-negative patients, were PEA-negative. Overall, 135 patients (70%) had consistent results between the PEA and HIPAA. Subsequently, for a group of 18 patients tests were repeated at three different days and demonstrated inconsistent results. Only three out of 18 (17%) patient samples showed the same result in the PEA (negative) on every testing day; for five samples an initial discrepancy

between the PEA and HIPAA was absent in repetitions; for the other 10 samples one or more repetitions were discrepant.

Summary / Conclusions: Despite a prior recommendation in favor of using the PEA for HIT laboratory diagnostic testing, in our hands, discrepancies were revealed by repeated testing of samples raising concerns about its reliability and robustness. Therefore, we were unable to compare the performance of the PEA to that of the HIPAA. Further laboratory validation studies with clinically well-defined patients samples are necessary before considering the PEA as a replacement for the HIPAA.

P648 | A novel gel Coombs inhibiting test on platelet antibody—as simple as testing RBC antibody with MGIA

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Background: Platelet antibody testing holds significant clinical importance, such as confirming the diagnosis of immune thrombocytopenia, routine testing prior to transfusion or platelet transfusion, and platelet cross-match testing in experiments such as foeto-maternal platelet hematological compatibility testing. Currently, several commonly used methods for platelet antibody detection exist, including the platelet adhesion immunofluorescence test (PAIFI), antigen capture enzyme-linked immunosorbent assay (MACE), solid-phase red blood cell adhesion assay (SPRCA), monoclonal antibody-specific platelet antigen immobilization (MAIPA), etc. However, existing detection methods are far from ideal; they entail complicated processes that are time-consuming and require skilled professional technicians operating in dedicated laboratories. Establishing a new, straightforward platelet antigen-antibody detection method is crucial to ensure blood transfusion safety.

Aims: We aimed to develop a competitive assay for detecting anti-platelet antibodies using the erythrocyte Microcolumn Gel Coombs Test (MGCT) as an indicator system, with the ABO MGCT chosen as an example during development. The novel Immunoassay in question has been established based on the Known platelet ABO blood antigens and serum ABO antibodies.

Methods: This ABO blood grouping system-based competitive assay is described as follows: Type O platelets were used as positive controls, and type A, B, and AB platelets as negative controls; type O plasma served as reference plasma. Platelets were incubated with bovine serum albumin to block any non-specific bindings. Platelets were then incubated with plasma samples. The mixture was spun down and the supernatant discarded. The sediment was resuspended, and anti-human immunoglobulin antibodies were added. The solution from step 5 was tested using ABO MGCT. A positive result in MGCT indicated the absence of platelet antibodies; a negative or weaker result indicated the presence of platelet antibodies in the samples.

Results: In total, 16 platelet samples (8 type O and 8 type A, B, and AB) were used in this new assay. The results were as expected: type O platelets showed positive results, while type A, B, and AB showed negative results in MGCT. These results are consistent with the presence of anti-A and anti-B antibodies in type O plasma.

Summary / Conclusions: This study successfully established a micro-column gel-based competitive assay for the detection of anti-platelet antibodies. The accuracy and feasibility were demonstrated using ABO MGCT.

Thanks Mr. Yong Li, Suzhou Institute of Biomedical Engineering and Technology Chinese Academy of Sciences, for guiding this study.

P649 | Abstract withdrawn

P650 | Abstract withdrawn

P651 | Prevalence and causative factors of neonatal thrombocytopenia and elucidation of NAIT through human platelet antigen typing

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Background: Neonatal Allo-immune Thrombocytopenia (NAIT) results due to the Human platelet antigen (HPA) incompatibility between the mother and the foetus, which in turn leads to maternal alloimmunization against fetal platelet antigens. The most implicated antigen is HPA-1a. Neonatal Alloimmune Thrombocytopenia (NAIT) is thought to be a relatively rare cause of thrombocytopenia in neonatal period in view of paucity of data but can lead to serious life-threatening complications. In view of lack of a defined protocol for its diagnosis and management, it is important to identify at risk patients with the help of new non-invasive methods and prevent fetal complications.

Aims: The aim of our study was to evaluate the suspected cases of Neonatal Alloimmune thrombocytopenia so as to determine its prevalence and formulate an appropriate algorithm for its diagnosis and management in our setup.

Methods: Thrombocytopenic Neonates from neonatal services were included in the study between May 15th 2018-January 26th 2019 and June 15th 2019-March 20th 2020, which is a period of 1 year and 7 months. The study was conducted in the department of Transfusion Medicine and Neonatology unit of PGIMER, Chandigarh. Thrombocytopenic neonates were enrolled based upon their recorded hemogram. The probable causes of thrombocytopenia was studied. An algorithm was designed to evaluate Suspected NAIT including maternal platelet antibody screening and crossmatching using Solid Phase system by Galileo from immunocor Norcross A, USA. Further genotyping of both baby's and mother's was done using Real time Polymerase Chain Reaction from Applied Biosystems.

Results: Our study enrolled 1154 neonates, categorized based on the severity of thrombocytopenia into Mild (38%) Moderate (27%) and severe (27.8%). Clinical bleed was found to be about 5.9%. Overall, sepsis was found in 29.3% of neonates, followed by Birth Asphyxia (23%), Prematurity (14.1%), IUGR (9.5%), Syndromic (4%), NAIT (1.1%) and Out of these 1154 neonates, 10 gave positive results for maternal screening for the presence of anti-HPA antibodies as well as for the incompatibility testing done by Solid Phase Red Cell Adherence assay using Immucor Norcross, USA. On genotyping, HPA-3b and HPA-9b was found to be the most prevalent antigen in the neonate, which most likely has led to maternal alloimmunization and passage of anti-HPA-3b and anti-HPA-9b antibodies 30% of the neonates showed alloimmunization due to HPA-3b, and HPA-9b. HPA-15b was also found to cause alloimmunization (20%). The presence of, HPA-5b, HPA-2b, HPA-3a, HPA-8a, HPA-11a was also observed in the neonates which lead to alloimmunization.

Summary / Conclusions: 1154 out of the total 6237 neonates were thrombocytopenic (18.5%) Thrombocytopenia was majorly Mild ($100-150 \times 10^3/\mu\text{L}$) that is, 38% in the study population. Clinical bleed was found to be 5.9% in the study population. Sepsis (29.4%) was the most common cause for thrombocytopenia followed by birth asphyxia (23%), Prematurity (14%) and Intra Uterine Growth Restriction (9.5%). NAIT was prevalent in 0.9% of the thrombocytopenic neonates, and was 0.1% in the total admission of the neonates. HPA-3b and HPA-9b were found to be the most prevalent (30%) antigen; likely to cause NAIT. Allo-immunization was also seen due to the presence of HPA-15b (20%), and about 10% also showed alloimmunization due to the presence of HPA-2b, HPA-3a, HPA-5b, HPA-8a and HPA-11a.

Immunohaematology—granulocyte immunology

P652 | Anti-HN3 antibodies and renal CL-2—association with second episode of acute rejection after kidney transplantation

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Background: The recent identification of Choline Transporter-Like Protein 2 (CTL-2) as a new blood group system stands out for its distribution not only in red blood cells, but also in various cells and tissues of the human body. Predominantly localized on neutrophils, microvascular endothelial cells of the lungs, and renal tubular cells, this protein carries the human neutrophil antigen-3 (HNA-3) in its first extramembrane loop. Anti-HNA-3 antibodies are mainly associated with severe cases of Transfusion-Related Acute Lung Injury (TRALI) and Neonatal Alloimmune Neutropenia (NAIN). In situations of Acute Rejection (AR) of kidney transplantation, where antibodies against

Human Leukocyte Antigen (HLA) are not detected, there is increasing interest to investigate if anti-HNA-3 antigens may play a significant role in this context.

Aims: To investigate the association between the presence of pre-formed anti-HNA-3 antibodies and the development of the second episode of AR after kidney transplantation.

Methods: This retrospective cohort study included 1256 patients who experienced the first episode of AR after kidney transplantation between 2011 and 2019. The identification of anti-HNA-3 antibodies was performed using serum samples obtained before kidney transplantation. The following serological techniques were employed: Granulocyte Agglutination Test (GAT), Granulocyte Immunofluorescence Test (Flow-GIFT) and the bead-based kit LABScreen Multi (One Lambda). HNA-3 genotyping of patients and their respective kidney donors was conducted by PCR-RFLP, only on samples with suspected presence of anti-HNA-3 antibodies. The control group included patients who also experienced the first episode of AR but did not have anti-HNA-3 antibodies before transplantation. The groups were matched based on the date of transplantation, age, sex, time in dialysis, donor type (living or deceased), total HLA mismatches, induction therapy and maintenance immunosuppression.

Results: Anti-HNA-3 antibodies were detected in 33 (2.6%) of the 1256 patients included in the study: 4 anti-HNA-3a and 29 anti-HNA-3b. Among these, 13 (39.4%) had donor specific anti-HNA-3 antibodies. The incidence of the second episode of AR was significantly higher in the group of patients with anti-HNA-3 antibodies (33.3 vs. 9.1%, $p = 0.033$). Furthermore, graft survival up to 12 months after the second episode of AR was lower in the group of patients with anti-HNA-3 antibodies compared to the control group (66.7% vs. 90.9%, $p = 0.014$, Log Rank).

Summary / Conclusions: The study highlights the importance of further investigating this new blood group system in different contexts to determine the relevance of the CTL-2 protein. The presence of anti-HNA-3 antibodies before kidney transplantation is associated with an increased risk of a second episode of AR and lower graft survival, reflecting similar mechanisms to those observed in AR caused by anti-HLA antibodies. These results have significant clinical implications, suggesting the inclusion of anti-HNA-3 antibody as a risk factor for AR in kidney transplantation and opening new perspectives for investigating the intrinsic mechanisms underlying this process.

P653 | Donor-specific HNA-1a antibodies after unrelated hematopoietic stem cell transplantation

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Background: Antibodies targeting Human Neutrophil Antigens (HNA) are commonly associated with autoimmune neutropenia, neonatal alloimmune neutropenia and transfusion-related acute lung injury (TRALI). Moreover, in recipients of hematopoietic stem cell transplantation (HSCT) these antibodies may lead to immune neutropenia, increasing infection risk and potentially contributing to graft failure. We report two cases wherein antibodies directed against the donor's HNA-1a antigen, absent in the recipient granulocytes, were identified post-HSCT.

Aims: To describe two infrequent cases of anti-HNA-1a immunization post-HSCT.

Methods: A retrospective analysis of clinical and laboratory patient records was conducted. Direct and indirect immunofluorescence tests, as well as the LABScreen Multi assay using Luminex technology, were employed to detect anti-granulocyte antibodies and identify the HNA-1a specificity. HNA-1 genotyping was carried out using allele-specific primer PCR, for both patients and unrelated donors. Chimerism status was determined by Short Tandem Repeat (STR) analysis.

Results: Patient 1, a 6-year-old boy with X-linked adrenoleukodystrophy, underwent a bone marrow transplant from a matched unrelated donor (MUD) with a myeloablative conditioning regimen. On day +28, platelets and reticulocytes engrafted, but neutrophil counts remained $<0.2 \times 10^9/L$. Direct immunofluorescence test confirmed the presence of IgM and IgG antibodies coating granulocytes, with an HNA-1a specificity identified in the serum using the LABScreen Multi assay. No HNA-antibodies were present in the pre-HSCT sample. HNA-1a genotyping showed an HNA-1a negative result for the patient but positive for the unrelated donor. Treatment with filgrastim, steroids, rituximab, high-dose intravenous immunoglobulin (IVIG) and sirolimus was initiated, resulting in successful neutrophil engraftment on day +54. However, the patient experienced severe pancytopenia and subsequent secondary graft failure, requiring a second transplant six months later, with an HNA-1a negative MUD. Patient 2, an 18-year-old girl with Dyskeratosis Congenita, underwent HSCT with peripheral blood CD34+ cells from a MUD, using a reduced-intensity conditioning regimen. Successfully engrafted on day +11. Two years after the HSCT, immune thrombocytopenia was diagnosed and managed with thrombopoietin receptor agonists. Furthermore, severe neutropenia and recurrent urinary infections were observed. A direct immunofluorescence test revealed the presence of IgG anti-granulocyte antibodies and the Luminex assay identified an anti-HNA-1a specificity. As in the case of patient 1, no HNA-antibodies were present in the pre-HSCT patient's sample. Her HNA-1 genotype indicated a negative HNA-1a typing whereas the donor was HNA-1a positive. Filgrastim, rituximab and high-dose IVIG were administered as an approach to maintain an acceptable neutrophil count.

Summary / Conclusions: The cases reported here illustrate the development of donor-specific HNA-1a antibodies, highlighting a rare condition that can lead to severe immune neutropenia, potentially complicating the post-HSCT course. Early recognition and targeted intervention are crucial in managing such complications effectively.

P654 | Analysis of maternal FcγRIIIb isoantibodies using immunomagnetic negative selected neutrophilsJ O Martins¹, E Moritz¹, S A Abbas¹, B Bayat², M Oliveira Barros¹, R de Marco³, R M Fantini³, J O Bordin¹¹UNIFESP, São Paulo, Brazil, ²Institute for Clinical Immunology, Transfusion Medicine, and Haemostasis, Giessen, Germany, ³Instituto de Imunogenética (IGEN), São Paulo, Brazil

Background: The isolation of neutrophils and subsequent detection of anti-human neutrophil antigens (HNA) antibodies are crucial in clinical medicine for diagnosing autoimmune neutropenia, neonatal alloimmune neutropenia (NAIN), and transfusion-related acute lung injury (TRALI).

Aims: This study reports two cases of maternal anti-Fc gamma receptor IIIb (FcγRIIIb) isoimmunization without NAIN symptoms and compares the efficiency of immunomagnetic negative selection (IMNS) with traditional Dextran/Ficoll for neutrophil isolation in serological assays.

Methods: Investigating two cases of maternal anti-FcγRIIIb isoimmunization, neutrophils from three donors were isolated using IMNS and Dextran/Ficoll. Serological assays included the granulocyte agglutination and immunofluorescence test (GAT and GIFT), monoclonal-antibody immobilization of granulocyte antigens (MAIGA), and the LABScreen Multi kit (One Lambda). PCR-SSP was performed to identify the *FCGR3B* alleles. Absolute neutrophil counts in newborns were performed after birth (case 2) and after 40 days (cases 1 and 2). IMNS

and Dextran/Ficoll were compared in terms of cell yield, viability, time, cost, and purity.

Results: Maternal anti-FcγRIIIb isoantibodies with *FCGR3B* gene deletion were detected in both cases. Newborns and fathers exhibited specific gene combinations: *FCGR3B*02/FCGR3B*02* (Case 1) and *FCGR3B*02/FCGR3B*03* (Case 2), confirming the fetomaternal incompatibility. No neutropenia or signs of infection related to low neutrophil counts were detected at birth or on the fortieth day of the newborn's life. IMNS outperformed Dextran/Ficoll, yielding four times more neutrophils (average neutrophil counts: $18.6 \times 10^3/\mu\text{L}$ vs. $4.5 \times 10^3/\mu\text{L}$), efficiently removing non-neutrophil cells without the need for the lysis step, and reducing processing time (30–40 min vs. 70–90 min), though it incurred a higher cost (2.7 times). A high percentage of viable cells was observed in both methods (average of 91% vs. 86%) (Table 1).

Summary / Conclusions: Two cases of maternal anti-FcγRIIIb isoantibodies, unrelated to NAIN, were identified. Although neutropenia has not been described in these cases, we emphasize the importance of identifying asymptomatic cases with the potential for severe neutropenia. Additionally, IMNS is introduced as a rapid, high-yield neutrophil isolation technique beneficial for serological assays detecting anti-HNA antibodies.

Immunohaematology—fetal-maternal immunology

P655 | Automated gel card anti-D (RH1) titration—determination of risk thresholds for severe hemolytic disease of the fetus and newborn from clinical outcomes

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Background: In case of anti-D (RH1) alloimmunization, during pregnancy, maternal antibodies can cross the placenta and cause Hemolytic Disease of the Fetus and Newborn (HDFN). In severe cases, fetal anemia can develop and results, if untreated, to hydrops and intrauterine fetal death. In the event of significant alloimmunization defined in France by an antibody titer greater than 16 (indirect antiglobulin test tube method) or an anti-D quantification greater than 5 UI/ml (continuous flow analysis (CFA)), fetal ultrasound monitoring with weekly measurement of the middle cerebral artery peak systolic velocity (MCA PSV) is undertaken from the 18th week of gestation. If ultrasound signs of severe fetal anemia are observed, Intrauterine Transfusion (IUT) can be realized. Anti-D titrations and quantifications also make it possible to assess the risk of severe Hemolytic Disease of the Newborn (neonatal anemia and hyperbilirubinemia) as well as to anticipate care for the newborn (phototherapy, transfusion,

P654 - Table 1. Characteristics of the neutrophil isolation techniques used to study anti-FcγRIIIb isoantibodies against the cells of 3 previous genotyped healthy subjects.

	Average results (Donors 1, 2 and 3)	
	Dextran/Ficoll	IMNS
Cell type [$\times 10^9/\text{L}$ (%)]		
WBCs	4.9	19.2
Neutrophils	4.5	18.6
Other Leucocytes	0.4	0.7
RBCs	0	0
Platelets	0	0
Viability	0.86	0.91
Number of tests ^a		
GAT (2 $\mu\text{L}/\text{test}$)	941	3840
Flow-GIFT (40 $\mu\text{L}/\text{test}$)	24	96
Cost		
Catalog number	31392-50G/17-1440-02	19666
Company	Sigma/GE healthcare	Stemcell
Price per sample (USD)	0.60/1.03 (1.63)	4.44
Time spent (minutes)	70-90	30-40

Abbreviation: IMNS, Immunomagnetic negative selection.

^a Number of tests that can be performed from 8 ml of whole blood and a final volume of 1 ml.

transfusion exchange...). But titration by tube method is known to have high intra- and interassay imprecisions and CFA is only performed in specialized laboratories. Recently, automated titration methods on different supports has been developed by several manufacturers.

Aims: Our laboratory has evaluated an automated titration method using LISS Coombs gel cards (IH-500 Bio-Rad) in 2018 and the anti-D titers were found on average 3 times greater than the reference tube method. However, for some tube titers, the respective gel titers were 2 to 6 times higher making direct extrapolation of the tube threshold of 16 to a new gel card threshold difficult. We also demonstrated a correlation between the titer score in gel card and the anti-D quantification. The cutoff value of 5 IU/ml corresponded to a titer score of approximately 75 in gel card. We wanted to determine new cutoffs for anti-D titer and titer score in gel card based on clinical data.

Methods: From 01/01/2021 to 30/05/2022, data (gel card titers and titer scores) were extracted from the Laboratory Information System used at the National Reference Center in Perinatal Hemobiology (CNRHP) for anti-D alloimmunized pregnant women followed at Trousseau hospital in Paris. The number of fetal and/or neonatal transfusions and hemoglobin and bilirubin values at birth were collected.

Results: 75 patients were included. 25 fetuses developed severe fetal anemia. The minimum gel anti-D titer associated with a severe fetal anemia was 128 with a titer score of 61. Among the newborns who did not develop fetal anemia ($n = 50$), 19 presented anemia at birth (cord hemoglobin $< 14\text{g/dl}$) and 15 had hyperbilirubinemia (cord bilirubin $> 80\text{ }\mu\text{mol/l}$). The minimum gel titers and titer scores associated with neonatal anemia and severe jaundiced were 128, 69 and 256, 87 respectively.

Summary / Conclusions: For high-risk pregnancies due to anti-D alloimmunization, based on clinical data, automated gel titer and titer score thresholds with IH-500 triggering ultrasound monitoring and anticipating severe hemolytic disease of the newborn have been set at 128 and 61, respectively. Titer results correlate with the average of 3 dilutions difference with the tube technique and for the titer score, results are slightly lower than the previous estimate based on biological correlation.

P656 | Anti-D (RH1) scores calculated by automated solid phase red cell adherence titration tests correlate with continuous flow analysis anti-D quantification

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Background: In France, for pregnancies complicated by anti-D (RH1) alloimmunization, the tests currently used to quantitate maternal antibodies are tube method titration and continuous flow analysis

(CFA) determination of the antibodies concentration. CFA is more precise and detect an increase in antibody levels 2-weeks earlier than tube titration. Thus, it allows to better anticipate fetal transfusions and neonatal care. But it requires expertise and it is only available in specialized laboratories. Recently, an automated assay was developed using a solid phase red cell adherence plate technology on Neo Iris analyzer (Werfen, Immucor).

Aims: We wanted to evaluate the scores, calculated from the agglutination patterns (Marsh scores) and /99 scores given by the Neo Iris automated platform in titration technique, as a quantitative data to appreciate the level of maternal antibodies.

Methods: Titrations of anti-D in patient samples have been performed using the semi-automated tube method (indirect antiglobulin test) realized since decades in our lab and the automated plate method on the Neo Iris. Scores were calculated manually in both cases. Antibodies concentrations were also determined for all samples by CFA on our auto-analyzer devices (Astoria WHS or Proxima AMS Alliance) (2-stage technique). We looked for a possible correlation between anti-D patterns and /99 scores and the corresponding CFA concentrations using the Spearman correlation test. The correlation coefficients found were compared with those determined for Marsh's titration scores in tube.

Results: 19 samples containing anti-D with a titer between 2 and 256 on microplate and tube techniques were tested. Microplate titration pattern scores and /99 scores were significantly correlated with the quantification assay ($p < 0.0005$, Spearman), with a r correlation coefficient of 0.77, higher than that obtained for Marsh scores in tube technique (0.63). It also seems possible to extrapolate critical threshold scores, correlated with the critical anti-D threshold concentration of 5 IU/ml established to trigger fetal ultrasound monitoring for indirect signs of fetal severe anemia. These threshold scores would be around 60 for the pattern score and 500 for the /99 score.

Summary / Conclusions: The calculation of pattern scores and /99 scores from microplate titration results by solid phase red cell adherence on the Neo Iris automaton provides added value compared with titer reading only. Complementary studies with standards and on a larger number of samples, supported by clinical data, should be considered.

P657 | Non-invasive fetal RHD genotyping in D negative (RH:-1) patients with RHD genomic sequences—retrospective study over 11 years

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Background: Non-invasive fetal RHD genotyping is a benefit in antenatal obstetric and biological monitoring of D negative pregnant women, whether immunized or not. At the CNRHP, the identification of RHD positive fetuses is carried out first by quantitative real-time

PCR on *RHD* exons 10, 7 and 5. This test is only conclusive if the D negative women have a deleted *RHD* gene. If they have *RHD* sequences, this results in early signal amplification (Ct<35).

Aims: We wanted to provide a picture of the causes of non-invasive fetal *RHD* genotyping results with early Ct in our laboratory and to look at how these results were interpreted.

Methods: In patients with early Cts, we routinely perform the maternal D phenotype and *RHD* genotype and compare them to the result of the non-invasive fetal *RHD* genotyping. An additional quantitative PCR for *RHD* exon 6 could be performed if necessary. All of these data were extracted over an 11-year period from the Laboratory Information System used at the CNRHP.

Results: Between 2011 and 2022, 7.5% of fetal *RHD* genotyping presented Ct<35. 8.6% of patients were D negative with a deleted *RHD* allele. In this case, the fetus was found to be positive. Cts<35 were often due to advanced gestational age or twin pregnancies. 77.6% were D negative with a silent *D^{psi}* or *D-CE(7-4)-D* *RHD* allele. The genotype of the fetus could generally be concluded thanks to our multiple exons amplification approach. 3% were phenotypically D positive with the reagents used in our laboratory, 3.9% expresses a weakened D phenotype (partial or weak D (variant)) and 6.9% were D negative with an uncharacterized silent *RHD* allele. In these 3 cases, the fetus was concluded indeterminate by maternal DNA interference.

Summary / Conclusions: In 11 years, 0.5% of requests for non-invasive fetal *RHD* genotyping concern D positive or D variant patients and 6.3% of women present a silent *RHD* allele. Given the risk of providing an erroneous result for the fetus (false positive) or inappropriate clinical-biological advice, it is essential to know how to detect and interpret the presence of *RHD* maternal sequences to be able to produce a correct result for the fetus.

P658 | Cost-effectiveness of cEK-preventive matched transfusion strategies for female transfusion recipients to prevent hemolytic disease of the fetus and newborn

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Background: In the Dutch population, anti-K, anti-c and anti-E are the most important antibodies in causing severe hemolytic disease of the fetus and newborn (HDFN), after anti-D. These antibodies can develop after an incompatible pregnancy or transfusion. Preventive matched transfusion strategies are implemented in many countries. In the Netherlands (18 million people and 165,000 pregnancies per year) the transfusion guideline prescribes matching for K for female transfusion recipients under the age of 45 since 2004, and for c and E since 2011.

Aims: To assess the cost-effectiveness of matched transfusion strategies for c, E, and/or K antigens to prevent HDFN, in comparison with the current matching strategy for cEK-antigens in the Netherlands.

Methods: A decision analysis model was developed to compare the current preventive cEK-matching strategy to three other strategies: (1). matching for c and K (cK matching), (2). matching for K only (K-matching), and (3). no matching. Model parameters consist of prevalences and probabilities based on laboratory data from alloimmunized pregnancies screened and followed in the period 2000-2022, clinical data of pregnancies complicated by HDFN, literature and expert opinions. Costs were based on recent laboratory data, treatment costs were based on actual cost prices. One-way sensitivity analyses (OWSA) and probabilistic sensitivity analyses were performed.

Results: Compared to the cEK matching strategy, calculated to have total yearly costs of €5,786,000, the total costs of cK-matching are €5,707,000 and thus 79,000 euro per year less for the Dutch situation, whereas it results in only a loss of 0.015 Quality-Adjusted Life Years (QALYs) per year. The ICER (Incremental Cost-Effectiveness Ratio) of cK-matching compared to cEK-matching was 5.2 million euro per QALY. The probability of cK-matching being more cost-effective is higher than 85% for all acceptable values of the willingness-to-pay per QALY. Matching for K only, or a strategy without matching, were dominated by cEK- and cK-matching, making these scenarios less suitable. OWSA showed that the most influential parameter for all matching strategies was the annual number of antigen typings for c or E of women prior to transfusion.

Summary / Conclusions: In this analysis a strategy with c and K matching, without matching for E, was a more cost-effective strategy than the current cEK-matching strategy. The costs incurred for typing women prior to every (planned) transfusion are of large influence on the outcome. An adaptation of the Dutch transfusion guideline to a strategy with matching for c and K, and without matching for E, should be considered. The results of this study will be taken into account when deciding on possible policy adjustments, and can provide new insights for decision makers on preventive matching for female transfusion recipients to prevent HDFN.

P659 | Advancing prenatal diagnosis—a comparative study of ddPCR and qPCR for non-invasive *RHD* genotyping in D-negative pregnancies

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Background: Hemolytic Disease of the Foetus and Newborn (HDFN) caused by anti-D antibodies is a significant concern globally. To

prevent this condition, D-negative pregnant women are routinely administered prenatal anti-D immunoprophylaxis, followed by an additional dose post-delivery of a D-positive newborn. Non-invasive fetal RHD genotyping allows an early risk assessment of this disease in pregnancies with anti-D alloantibodies and has the potential to avoid antenatal anti-D prophylaxis in non-sensitized D negative pregnant women carrying RHD negative fetuses. This approach requires sensitive methodologies capable of detecting cell-free fetal DNA (cffDNA) at early gestational ages.

Aims: this study aims to evaluate a novel Droplet Digital PCR (ddPCR) technique, developed in our laboratory, against the already validated Real-Time PCR (qPCR) method for prenatal RHD genotyping in D-negative pregnant women.

Methods: peripheral blood samples (10 ml) from 27 D-negative pregnant women (gestation weeks 12-38) were collected for prenatal RHD genotyping. Plasma aliquots (4 ml) were processed to extract total cell-free DNA using the QIAamp MinElute ccfDNA Midi Kit (Qiagen). The ddPCR assays were performed on a QX200 Droplet Digital PCR System (Bio-Rad), following the manufacturer's guidelines, while the Real-Time PCR reactions were run on a Step One Applied Biosystem thermocycler. In both strategies, two different regions of the RHD gene (exon 5 and exon 7) were analysed alongside a reference gene (GAPDH) and a sex-determining gene (SRY).

Results: the ddPCR technique successfully genotyped all 27 cffDNA samples, identifying 3 RHD-negative and 24 RHD-positive fetuses, including 11 males and 16 females. Conversely, the Real-Time PCR method failed to characterize three samples, likely due to a low cffDNA concentration as these samples were from early gestations of 11 ($n = 2$) and 12 ($n = 1$) weeks. The genotype and sex determinations for the remaining 24 cffDNA samples were consistent across both ddPCR and qPCR methods. Serological analysis of cord blood samples confirmed the molecular findings with 100% accuracy.

Summary / Conclusions: the ddPCR technique formulated in our laboratory proves to be a dependable and effective approach for accurately determining prenatal RHD genotype in all analyzed cffDNA samples. Given that cffDNA constitutes only 5%–15% of total cfDNA, a significant advantage of ddPCR over Real-Time PCR is its superior sensitivity, particularly beneficial for samples with a low fetal cfDNA proportion.

P660 | Performance of non-invasive fetal RHD genotyping methods in a daily laboratory routine

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Background: For several years, reliable and feasible methods have been available for the non-invasive fetal Rhesus Factor D (RHD) genotyping in maternal blood. According to the German guidelines, non-invasive fetal RHD analysis should be offered to every RhD-negative

pregnant woman in Germany to avoid unnecessary anti-D prophylaxis for RhD-negative pregnant women with an RHD-negative fetus.

Aims: The aim of the study was to evaluate the reliability and robustness of fetal RHD analysis methods used over a period of more than three years for over five thousand pregnant women in our laboratory.

Methods: Maternal blood was collected in 9 mL EDTA tubes and transported at room temperature. Extraction of cfDNA was carried out from 2 mL of plasma using the QIASymphony DSP Circulating DNA Kit (Qiagen), following the manufacturer's protocol, with an elution volume of 60 µL. Two different real-time PCR test systems were employed in parallel for fetal RHD detection: the commercial Free DNA Fetal Kit RhD - Duplex (Jacques Boy) and an in-house method for fetal RHD detection. Both methods utilized amplification of exon 5, 7, and 10 of the RHD gene on the CFX Opus Real-Time PCR System (Biorad). Additionally, an internal control (IC) maize DNA was used with the commercial kit to assess DNA extraction efficacy and absence of PCR inhibitors. Samples were classified as positive if the IC ($C_t < 37$) and all RHD exons were amplified (C_t 34.5 to 41). Samples were considered negative if no amplification was detected for RHD exon 5, 7, or 10, and the IC was amplified ($C_t < 37$). In cases of discordant amplification results, samples were reprocessed.

Results: All samples received at our laboratory from January 1st to December 31st, 2023, for fetal RHD genotyping in maternal blood were retrospectively evaluated, a total of 2008 samples. Four of these were rejected based on German guidelines; three were collected before the 12th gestational week, and the fourth was rejected due to twin pregnancy. Additionally, 13 samples turned out as serologically RhD-positive. Out of the remaining 1991 samples, genotyping results could not be obtained for 23 samples (1.16%), most likely due to genetic variations in maternal or fetal RHD genotype. Nine of these showed early amplification in all analyzed RHD exons but were serologically RhD-negative. Conflicting results in different RHD exons or unexpected results (e.g., early amplification for one or more exons) were observed in 185 (9.6%) samples in the first setup. All these samples were processed a second time. Only 27 samples (1.36%) remained inconclusive after the second setup and were reported as test failures. For samples that failed, the average transport time was double that of all samples (4.6 days vs. 2.3 days). Only one of the samples (0.05%) was initially reported as false positive. Negative fetal RHD genotype was found in 732 samples (37.7%).

Summary / Conclusions: For the past three years, our laboratory has been analyzing samples for non-invasive fetal RHD genotyping in maternal blood. The real-time PCR-based method is feasible and allows a clear fetal RHD genotype assignment in 97.5% of samples. Genetic variations in maternal or fetal RHD genotype or test failure, mainly due to the degradation of cell-free fetal DNA during prolonged sample transport, are the most frequent reasons for test failure. Assuming that the laboratory is contacted if postpartum fetal RhD serology does not match with genotyping, the proportion of false negative or false positive results is very low.

P661 | Non-invasive prenatal testing utilising massively parallel sequencing for blood group genotyping of alloimmunised pregnant women in a single test system

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Background: Pregnant woman with antibodies to red blood cell (RBC)/human platelet (HPA) antigens are at risk of carrying a fetus affected by haemolytic disease of the fetus and newborn or fetal and neonatal alloimmune thrombocytopenia. Non-invasive prenatal testing (NIPT) of circulating cell-free fetal DNA (cffDNA) in the maternal plasma provides a safe approach to predict the fetal antigen status and identify pregnancies at-risk. However, testing is currently restricted to a small panel of clinically significant antibodies, which may be limiting when applied to ethnically diverse patient populations, such as Australia.

Recently, we demonstrated the feasibility of using massively parallel sequencing (MPS) to predict the fetal phenotype for RBC/HPA antigens in a single test system (McGowan et al., Br J Hematol, 2023). In the previous cases studied, the MPS approach validated NIPT for antigens in Rh, Kell, Kidd, Duffy, Lutheran systems and for HPA. Further validation is required for more difficult targets, such as complex Rh and MNS variants and rarer targets in the panel for which an NIPT has yet to be developed.

Aims: The study aims to assess the suitability of an MPS approach in an extended patient cohort by correlating fetal MPS phenotype predictions with clinically reported outcomes.

Methods: A second custom QIASeq Targeted DNA MPS probe-panel was extended to cover 13 blood group (BG) systems, targeting 6 exons in the RH and MNS systems and 43 single nucleotide variant (SNV) regions associated with RBC/HPA antigens. Alloimmunised antenatal cases ($n = 45$, gestation age 11–34 weeks) were enrolled with informed consent. Variant calling and allele frequency analysis was performed using QIAGEN CLC Genomics Workbench. The allele frequency from gDNA was subtracted from that of the cffDNA to account for non-specific background reads, resulting in a subtractive allele frequency (SAF). The SAF threshold was set at $<1\%$ and $>2.5\%$ for negative and positive predictions respectively. Where available, MPS predictions were compared with droplet digital PCR and/or infant phenotyping results.

Results: For 45 maternal blood samples there were 12 cases with multiple antibodies (8×2 RBC antibodies, 1×2 HPA antibodies and 3×3 RBC antibodies). In total, there were 60 (20 negative, 34 positive and 6 inconclusive) predictions relating to alloimmunisation status requests. Confirmatory results were available for 20 (9 negative and 11 positive) predictions and showed 100% concordance. Concordant predictions included RHD exon detection as well as for SNVs in RHCE, KEL, FY and HPA (HPA-1 and HPA-3). Of 6 samples with inconclusive

outcomes (SAF $>1\%$ – $<2.5\%$), one provided a second sample showing SAF values increased from 2.4% at 12⁺5 weeks gestation to 3.3% 18⁺4 weeks gestation. The multiplexing capability of this system is highlighted in a case at 27⁺1 weeks gestation involving Fy^a, C and e antigens that predicted Fy(a–), C+ and e+, respectively, which was concordant with the cord blood and infant phenotyping result.

Summary / Conclusions: The panel is performing reliably in predicting fetal BG phenotypes by NIPT, including that for Rh. Ongoing assessment of NIPT fetal predictions in pregnancy cases with rarer antigens and those with multiple antibodies against RBC/HPA antigens showed that the comprehensiveness and multiplexing capacity of the MPS panel proved relevant to the diverse Australian patient population. Additional testing of samples shall continue to assess this NIPT MPS approach further for clinical translation.

P662 | Evaluation of presence of anti-D antibody in breast milk of RhD negative mothers

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Background: Secretory IgM and IgG isotypes have been reported in varying concentrations in human breast milk along with primary secretory IgA antibody. Breast milk IgG can transfer intact across the sub-mucosa into the blood and can lead to significant red cell binding and premature hemolysis of antigen-positive fetal red cells.

Aims: To identify the presence of anti-D antibody in breast milk of Rh D negative mothers. To compare the presence and titers of anti-D or any other alloantibody identified between maternal plasma and breast milk samples.

Methods: The present descriptive observational study was conducted over a period of 8 months at a tertiary care hospital after approval from Institute Ethics Committee. A total of 64 Rh D negative pregnant mothers with gestational age ≥ 34 weeks were enrolled. Two ml breast milk in a sterile container and 5 ml blood sample in an EDTA vacutainer were obtained from enrolled postpartum Rh D negative mothers within 7 days of delivery. ABO blood grouping and Rh D typing on maternal plasma and reverse blood grouping on whole breast milk samples was performed using tube technique. Indirect Antiglobulin test (IAT) was performed using microcolumn gel technique (BioRad, Switzerland) on both the samples. If IAT was positive, antibody screen, identification and doubling dilution titers of the antibody identified were performed using microcolumn gel technique on maternal plasma and breast milk and results compared. ABO blood grouping, Rh D typing and Direct Antiglobulin Test (DAT) were performed using micro-column gel technique on neonate sample within 24 hours of delivery as a part of hospital policy. Neonate was also followed up for requirement of phototherapy, double volume exchange transfusions (DVET) or for top up packed red cell transfusions (PRBC) during hospital stay.

Results: The mean age of enrolled mothers was 27.4 ± 5.1 years. Of 64 mothers, 23 (35.9%) were primigravida, 20 (31.3%) were gravida two, 18 (28.1%) gravida three, 2 (3.1%) gravida four and 1 (1.6%) was

gravida five. Maternal blood group distribution was as follows: 20 (31.3%) as O Negative, 18 (28.1%) as B Negative, 16 (25%) as A Negative and 10 (15.6%) as AB Negative. The presence of anti-A and anti-B antibodies in breast milk corresponded to the respective maternal blood groups in all the breast milk samples. Of 64 enrolled mothers, IAT was positive in 22 (34.4%) maternal plasma samples whereas in only 17 (26.6%) breast milk samples. Antibody identified was anti-D in 21 (95.5%) and anti-D along with anti-C antibody in 1 (4.5%) maternal plasma while only anti-D antibody (100%) was identified in 17 breast milk samples. Among 22 mothers with anti-D antibody in maternal plasma, 13 (59.1%) mothers had received Rh immunoprophylaxis and allo-anti-D antibody was seen in 9 (40.9%) cases. Titers of anti-D due to Rh immunoglobulin ranged from 1:1 to 1:2 in maternal plasma and negative to 1:1 in breast milk. Allo-anti-D antibody titers ranged from 1:4 to 1:32 in maternal plasma and 1:1 to 1:4 in breast milk. DAT was positive in 7 neonates with anti-D in 6 and anti-D along with anti-C antibody identified in eluate. Phototherapy was required in 4 (57.1%) neonates and phototherapy plus DVET in 2 (28.6%) neonates. Top up PRBC transfusions were not required in any of the neonate.

Summary / Conclusions: Our study demonstrated the presence of anti-D antibody in breast milk. No correlation was observed between titer of antibodies in maternal plasma and those identified in breast milk in our study.

P663 | Performance of an automated antibody titration method using solid phase red cell adherence

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Background: In France, the Immunohematology decree of May 15, 2018 paved the way for the use of other antibody titration techniques than the reference tube technique. The tube method is largely manual and is known to show great inter-laboratory results variability depending on multiple parameters. The new developed titration techniques could be automated, and better technical performance are expected.

Aims: We wanted to assess the performance of solid phase red cell adherence plate titration method on the Neo Iris automated system (Werfen, Immucor) and to compare it with the performance established for the reference tube method, used in our lab since decades. Another objective of the study was to try to determine plate titer thresholds for the main antibody specificities responsible for hemolytic disease of the fetus and newborn to trigger fetal monitoring by ultrasounds and measurements of the peak systolic velocity in the middle cerebral artery.

Methods: An home-made anti-D internal quality control (IQC) prepared and calibrated using the international anti-D Standard (16/332) as well as an anti-c commercial quality control (IH-QC

Bio-Rad) were used to determine the intraassay and interassay imprecisions, regarding the score and the titer results. Patients samples for testing were chosen during a 12-months assay period, regarding the specificity of the antibodies and the tube titer in order to cover a wide range of situations. Comparison of the results obtained from the same clinical samples with both methods was carried out.

Results: Better results were obtained with the solid phase red cell adherence plate titration method on the automated system for intra-assay imprecision. But interassay imprecisions show similar coefficient of variation (CVs) compared with the CVs established for the manual tube titration method (10%–15%). Titers obtained on tube and plate methods were compared for 131 samples from pregnant women: 37 anti-D, 41 anti-c, 31 anti-E, 22 anti-K. For anti-D (RH1), anti-E (RH3) and anti-c (RH4), titres were on average 2.3, 2 and 1.5 times higher respectively in the plate technique than in the tube technique. For anti-K (KEL1), differences in titres were less marked, averaging less than 1 dilution (0.6).

Summary / Conclusions: Automated anti-red blood cell antibodies titration by solid phase red cell adherence plate method on the Neo Iris shows similar interassay imprecision CVs compared to the tube method. But better imprecision CVs could be obtained using only fresh titration and revealing cells. Less inter-laboratory variability is expected with this method. Comparison of plate and tube titers show significant differences, with disparities depending on samples and antibody specificities. For anti-RH antibodies, titer results with the automated plate method are often higher than those with the tube method. For anti-K antibodies, results are more similar. In order to be able to establish threshold titers using the plate technique, linked to a risk of severe fetal or neonatal hemolytic disease, further studies on a larger number of samples need to be envisaged and results require to be validated with clinical data from pregnancy outcomes.

P664 | Non-invasive fetal KEL*01.01 genotyping using digital PCR

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Background: K-negative women may produce antibodies against fetal K antigen from Kell system leading to hemolytic disease of fetus and newborn (HDFN). In this setting, non-invasive prenatal testing (NIPT) of the fetal KEL*01.01 helps to identify high-risk pregnancies.

Aims: To validate a digital PCR (dPCR) protocol for fetal KEL*01.01 genotyping.

Methods: 104 DNA samples (extracted using easyMag, Biomerieux) from plasma of 41 pregnant K-negative women (13 to 33 week of gestation) were tested retrospectively to determine KEL*01.01, KEL*02 alleles and SRY gene in triplex assay in two replicates using

Naica 6-color dPCR (Stilla). The results were compared to fetal/neonatal *KEL*01.01*, *SRY* genotypes and sex.

Results: The dPCR results of *KEL*01.01/*02* and *SRY* genotyping were concordant with the genotype of pregnant women and their children. Mean value for *KEL*02* dPCR events in 104 tests was 814.23 (SD \pm 403, range 209-2702 events). Mean values for *KEL*01.01* dPCR events in 32 tests of plasma from women carrying K-positive fetuses and 70 tests of plasma from women carrying K-negative fetuses were 32.88 (SD \pm 17.1, range 7-72 events) and 0.12 (0.33 \pm SD, range 0-2 events), respectively. Mean values for *SRY* dPCR events in 78 tests of plasma from women carrying male fetuses and 26 tests of plasma from women carrying female fetuses were 14.99 (SD \pm 8.0, range 0-37 events) and 0.0 (SD \pm 0.0, range 0.0 events), respectively. For *KEL*01.01* and *SRY* dPCR reactions both limits of blank were 1 read and the limits of detection were 5 and 4 reads, respectively.

Summary / Conclusions: High accuracy of validated dPCR protocol determining fetal *KEL*01.01* genotype as well as fetal (*SRY*) and total (*KEL*02*) cfDNA fractions at the same experiment allows to implement the test into routine non-invasive diagnostics of immunized pregnant women.

P665 | Prevalence of maternal alloantibodies and their impact on outcomes of fetuses/neonates - a survey among tertiary level hospitals

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Background: Hemolytic disease of the fetus and newborn (HDFN) can have devastating consequences. Important efforts are made to prevent alloimmunization and the appearance of irregular antibodies (IAs) that can cause HDFN, but they still appear even in well-cared-for pregnancies.

Aims: To describe the causes of alloimmunization and the impact of IAs in a series of 1112 pregnancies.

Methods: The study involved a retrospective survey of pregnant women's demographics, IA detection, causative factors, and the effects on fetuses and newborns. The research, conducted across 25 Spanish hospitals from 2018 to 2020, analyzed IAs and their impact using a pre-existing classification system (de Haas, Vox Sanguinis, 2015).

Results: Data from 1,055 pregnant women from a total of 1112 pregnancies were analyzed. The most common IAs were anti-D (395 cases, 31.7%), anti-E (193, 15.5%), anti-c (157, 12.6%), anti-M (127, 10.2%) and anti-Kell (120, 9.6%). The parental phenotype was studied in 52% of cases, with 17% incompatible with the maternal IA, 9% compatible, and 26% not reported. The neonatal phenotype was analyzed in 51.7% of pregnancies, with 37% incompatible and 14% compatible neonates. The cause of anti-D sensitization was known in 47% of cases. Among these, 49.5% were due to incorrect prophylaxis during a sensitizing event or childbirth, and 44% to a "natural" event during a previous or the current gestation or abortion, without prophylaxis

failure. In 6.5% of cases, transfusion of RhD positive blood was identified as the cause. The cause of anti-Kell sensitization was known in 29.2% of cases, of which 45.7% were attributed to an incompatible blood transfusion and 54.3% to a sensitizing event in. For anti-c, the cause was known in 41.4% of cases, where 84.6% were due to a sensitizing event and 15.4% were due to an incompatible transfusion. In 24% of anti-E cases, the cause was known, with 67% resulting from a sensitizing event and 33% from an incompatible transfusion. Very severe HDFN was observed in 8.4% pregnancies, severe HDFN in 3.2%, and a mild form of HDFN in 9.2%. Of those with very severe HDFN, 62% presented a single IA specificity, while 38% had two or more. In severe cases, 78% had one specificity and 22% more than one. In mild cases 78.4% had one IA. During pregnancy, IAs considered high risk had no clinical impact on the fetus in 80.9% of cases, anemia appeared in 11.8%, hydrops in 4.4%, delayed fetal growth in 1.4%, and 1.4% died. At childbirth, high-risk IAs were present in 441 pregnancies, of which 57.6% were asymptomatic. Icterus was mild in 14.1 % of cases and severe in 12.5%. Severe anemia that required transfusion appeared in 9.1% of cases, while 4.1% were premature or low-weight neonates. One case received exchange transfusion (0.2%).

Summary / Conclusions: IAs continue to emerge during pregnancy, posing potential severe consequences for both fetuses and neonates. The main causes, such as incorrect prophylaxis and transfusions, are well known and deserve further training and process improvements to eradicate them. To improve gestational care, additional efforts should be directed toward characterizing immunization, as there is a lack of information in many cases.

P666 | Detection, monitoring, and implication of alloimmunization during pregnancy—five-year experience in Donostia University Hospital

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Background: Erythrocyte blood groups act as foreign antigens when encountered by the immune system of another organism, potentially leading to development of specific alloantibodies capable of causing erythrocyte hemolysis upon binding. Alloantibodies due to previous stimulus, known as irregular antibodies (IA) can have significant clinical implications in pregnancy due to risk of hemolytic disease of the fetus and newborn (HDFN) where maternal IgG-type alloantibodies destroy the fetal red blood cells due to antigens absent in the mother. IA can develop during pregnancy or as a result of previous immunization, and the clinical manifestations vary from mild to potentially fatal. Early identification and follow-up of IA in pregnant women is essential to prevent HDFN.

Aims: The objective was to determine the prevalence of alloimmunization during pregnancy in our population over the last 5 years, identify the most common specificities, and understand the prevalence of their clinical and analytical impact on the newborn.

Methods: This is a descriptive retrospective unicentric study where all requests for IA corresponding to first-trimester (1T) screening (IAS) during pregnancy in Gipuzkoa (Spain) were collected from April 2019 to January 2024. Subsequently, alloantibodies detected in the second and third trimesters, not included in the 1T IAS due to later development during pregnancy, were also collected. From positive IAS, the following variables were collected: age, blood group, previous pregnancies and transfusions, IA specificity and titration, risk group, delivery induction and data of HDFN.

Results: In the last 5 years in the province of Gipuzkoa, 19,266 IAS were performed for 1T screening. Of these, 145 were positive (0.75%): 59 were due to IgD gammaglobulin prophylaxis, 4 were auto-antibodies, 3 were secondary to gel reactivity interference, in 4 cases no specificity was found, and in 75 cases, one or more specific alloantibodies were detected, corresponding to a prevalence of positive IAS due to alloimmunization in 1T of 0.41%. Additionally, 43 positive IAS due to alloantibodies in the second and third trimesters not diagnosed in 1T were added. Therefore, the total number of alloimmunizations during gestation in this period was 118 (0.61%). 66.95% were present since 1T, and the remaining 33.05% developed during pregnancy. In 78.47% of cases, there was a history of previous pregnancies, and in 10.71%, a history of transfusion. The most frequent specificities were: anti-M (38.98%), anti-Lea (16.94%), and anti-E (11.86%). Of the total, 15.25% corresponded to the high-risk group (9 anti-D, 5 anti-c, 4 anti-K), 64.41% to intermediate risk, and 20.34% to the low-risk group. During follow-up only 6.78% reached titers higher than 1/32. In 21 cases, delivery was induced from week 38 due to the risk of HDFN. HDFN data were found in 4 cases (3.39%), two of them severe, related to alloimmunization by anti-D or anti-K antibodies.

Summary / Conclusions: The prevalence of alloantibodies during gestation in our setting was 0.61%. In 2/3 of cases were present from the beginning of pregnancy and in 1/3, they developed during pregnancy. In 78.47% of cases, there was a history of previous pregnancies, and in 10.71%, a history of transfusion. The most common alloantibody was anti-M, followed by anti-Lea and anti-E. 15.25% of detected alloantibodies corresponded to the high-risk group for HDFN. HDFN data were found in 4 cases, two of them severe, related to alloimmunization by anti-D or anti-K antibodies.

P667 | Clinical management of anti-Kell alloimmunization in pregnancy—a case report

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Background: Few reports have been published of the current clinical management of anti-Kell alloimmunization in pregnancy; its low frequency of occurrence means that the few series published have covered very ample time periods in which different kinds of clinical management have overlapped.

Aims: The aim of this paper is to present our experience in clinical management in a pregnant woman positive for the anti-Kell antibody.

Methods: SM, caucasian, age 22, single kidney, hypothyroidism in replacement therapy, PARA 1001, was observed in Mestre at a gestational age of 12 weeks, A CcDeekK, DAT negative, IAT positive, anti-Kell titer 1/1024. Her husband: PF, caucasian, age 28, A CcDeeKk. In both subjects extensive red blood cell phenotyping and genotyping were performed. Moreover, an analysis of the fetal DNA was performed, confirming that the fetus was K positive. The patient was treated with plasma exchange and IVIg at the Transfusion Medicine Department in Padua (see table I for details).

Results: The anti-Kell titer remained stable during pregnancy. A cesarean section was performed at a gestational age of 34 weeks. Newborn: PF, weight 2750g, length 47 cm, APGAR score 9, 10, 10 at 1st, 5th and 10th min. A CcDee, DAT positive for IgG but not for C3 and/or C4; after elution, it was not possible to identify the specificity of the antibodies bounded to the newborn's red blood cells as the panels tested consistently gave negative results. A Kk serological typing was performed in cordonal and neonatal blood: both samples were Kell negative, but genotyping confirmed Kk. The newborn presented a mild form of FNHD treated with phototherapy. In a further control performed four weeks after delivery, phenotype and genotype were concordant (Kk).

Summary / Conclusions: In our opinion this case report presents several points of interest. Firstly, the precocity and completeness of the

P667 - Table 1: Pre / Post plasma exchange anti-Kell titre

Date	Pre / Post Plasma Exchange anti-Kell Titer	Date	Pre / Post Plasma Exchange anti-Kell Titre
01/23/2023	2048 / 1024	03/16/2023	512 / 256
01/20/2023	1024 / 256	03/30/2023	1024 / 512
02/09/2023	512 / 256	04/06/2023	1024 / 512
02/16/2023	1024 / 256	04/12/2023	1024 / 1024
02/23/2023	512 / 256	04/23/2023	2048 / 1024
02/27/2023	512 / 256	04/27/2023	1024 / 512
03/02/2023	512 / 256	05/04/2023	2048 / 1024
03/09/2023	512 / 256	05/11/2023	2048 / 1024

diagnostic process which allowed us to confirm that the fetus was genotypically Kell positive. Secondly, the composite therapeutic approach based on a combination of plasma exchange and IVIg administration which allowed to keep under control the antibody titer. Thirdly, the discrepancy at birth between phenotype (K-) and genotype (K+). This discrepancy was resolved four weeks after birth, thus hypothesizing a masking of the antigen by the maternal IVIg treatment during pregnancy.

P668 | Use and clinical relevance of fetal hemoglobin measurement in cases suspected of fetal-maternal-hemorrhage

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Background: Measurement of fetal-maternal hemorrhage (FMH) can be of diagnostic benefit during pregnancy and newborn. In the Region of Southern Denmark (RSD) with approximately 12,000 births per year FMH is measured using dual-color flow cytometry (FC-FMH) with a combination of anti-hemoglobin F (HbF) and anti-carbon anhydrase (CA). Fetal red cells are identified as HbF positive and CA negative. Detection limit of the method has been verified at 0.07% of maternal red cells equivalent to approximately 1.5 mL of packed fetal red blood cells (RBC). At 22 weeks gestational age (GA) the median fetoplacental blood volume is 55 mL, equivalent to 33 mL packed RBC. Thus, before 22 weeks GA only a relatively large and recent FMH is detectable by FC-FMH and the policy in the RSD is not to perform this test if GA is <22 weeks.

Aims: To compare the clinical indications for performing FC-FMH measurement with the frequency of detecting a significant FMH and the GA at the time of sampling.

Methods: For the year 2023 all requests for FC-FMH measurement in the Region of Southern Denmark were reviewed with regard to

clinical indication, GA at the time of sampling, outcome of birth and FMH volume measured by FC-FMH.

Results: In 2023, there were 42 requests for FC-FMH measurement in the RSD. Two of these were clearly erroneous and were cancelled. Thus, the rate of ordering FC-FMH was 40/12,000 (0.3%) births. Most of the requests 35/40 (88%) were by the obstetric departments, the remaining 5/40 (13%) by the pediatric departments. The median GA at the time of sampling was 31 weeks (range: 15-42 weeks). At the time of ordering, 6/40 (15%) requests for FC-FMH were cancelled after consulting the ordering physician mainly because they concerned cases in early stage of pregnancy (missed abortion, late abortion or unknown). In only 1/34 (3%) pregnancies in which FC-FMH testing was performed was the fetal RBCs volume above the detection limit of 1.5 mL. In this case, the FMH was 0.96% of maternal red cells equivalent to 21 mL of packed fetal RBC. This newborn had a hemoglobin concentration at birth of 2.7 mmol/L (4.35 g/dL) after an emergency cesarean section was performed at 30 weeks GA. The child was pale at delivery but stable and the clinicians concluded that the anemia must have developed over a relatively long time. Upon review, 5/40 (13%) of the requests were considered outside policy because they were ordered on pregnancies that were <22 weeks GA. Consequently, a total of 11/40 (28%) of the initial requests for FC-FMH were considered clinically unnecessary.

Summary / Conclusions: FMH-measurement by flow cytometry is requested at a rate of 0.3% of births in RSD. With only 1/34 (3%) positive findings the utility of FC-FMH appears to be low in the current clinical setting, and 11/40 (28%) of the requests for FC-FMH were clinically unnecessary. Refining the ordering criteria to increase the pretest probability of a positive result is essential as is being vigilant not to perform the testing on pregnancies <22 weeks GA.

P669 | Evaluation of F cells after cesarean delivery of twins with the use of cytometry and microscopy

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Background: Although fetal hemoglobin disappears gradually after birth, small amounts of it are also found in adults. Adult F cells (HbF⁺ CA⁺) usually differ from fetal blood cells in a lower content of this hemoglobin and usually account for 1%-2% of all erythrocytes in most healthy adults. However level of F cells is differentiated, that may be genetically determined or acquired. There are different opinions about F cells in pregnant woman and the findings are inconclusive.

Aims: Evaluation of F cells in maternal blood in single and twin pregnancy with the use of cytometry and microscopy methods. Comparative analysis of cells before and after delivery by cesarean section in relation to chorionicity (monochorionic and dichorionic twins).

P668 - Table 1 Overview of the clinical indications for ordering FC-FMH testing.

Indication	N	% total
Foetus mortuus	9	23
Fetal anemia (by ultrasonic evaluation)	6	15
Neonatal anemia	6	15
Abortion < 22 weeks GA	5	13
Bleeding	3	8
Birth related	3	8
Maternal trauma	2	5
Not tested (missed abortion, late abortion or unknown)	6	15
Total	40	100

Methods: Blood samples from 34 woman were tested (24-before and after cesarean delivery of twins; 11 monochorionic and 13 dichorionic, 10-single pregnancy). Two flow cytometry tests with anti-HbF, anti-HbF + CA, anti-GPA antibodies, and modifications of microscopic Kleihauer-Betke (supplemented DAPI staining) test were used.

Results: Results obtained by cytometric (anti-HbF, anti-HbF + CA, anti-GPA) and microscopic (with DAPI staining) methods in samples from mothers didn't differ significantly. Percentage of F cells in single-ton pregnancy and after one chorion twin delivery was similar and was about 3% (before and after cesarean section). We observed increase in F cells after two chorion twin delivery (before and after CC: 4.7%). It was also statistically significant increase in the number of F cells in the two chorion twin samples, as compared to F cells in one chorion twin delivery.

Summary / Conclusions: Analysis of F cells is an important part of diagnosis of fetomaternal hemorrhage (FMH). Accurate F cells discrimination is significant to prevent false FMH results and unnecessary dosage of RhD immunoglobulin. Determination of F cells counts appears to be also of clinical importance in monitoring and the treatment of diseases as: thalassemia, hereditary persistence of fetal hemoglobin and sickle cell disease.

P670 | Use and clinical relevance of human platelet antigen typing in cases suspected of fetal and neonatal alloimmune thrombocytopenia

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Background: Fetal and Neonatal Alloimmune Thrombocytopenia (FNAIT) can occur when a pregnant woman develops antibodies against Human Platelet Antigens (HPA) carried by the fetus. These antibodies can cause severe fetal thrombocytopenia and lead to central nervous system (CNS) bleeding. In suspected prenatal FNAIT, the practice in the Region of Southern Denmark is to HPA-genotype both the mother and father and/or baby (paired genotyping) and perform serological testing for HPA antibodies to which the mother could become sensitized.

Aims: To compare the clinical indications for performing paired genotyping with the frequency of detecting an HPA antibody.

Methods: We systematically reviewed all maternal records relating to pregnancies where paired HPA genotyping was performed in the Region of Southern Denmark between January 1, 2020, and December 31, 2023. HPA typing in this region was carried out using a real time PCR (inno-train HPA FluoGene) for the HPA-1, -2, -3, -4, -5, -6, -9 and -15 genes. The pregnancies in which paired genotyping was performed were categorized based on the type of clinical findings that raised suspicion of FNAIT such as fetal CNS abnormalities

P670 - Table 1 Overview of paired HPA genotyping procedures and the clinical findings that raised suspicion for FNAIT.

Clinical finding	Number of cases	Parental HPA mismatches Number of cases (%)	HPA-antibody detected
Mild ventriculomegaly	10	7 (70)	No
Moderate ventriculomegaly	5	4 (80)	No
Mild ventriculomegaly and choroid plexus cysts	3	3 (100)	No
Other fetal CNS abnormalities*	5	4 (80)	No
Previous pregnancy with HPA antibodies	1	0	HPA-5b**
Unexplained thrombocytopenia in the newborn	6	3 (50)	HPA-1a
Unexplained death shortly after birth	1	1 (100)	No
Total	31	22	1

* Choroid plexus cysts, hydrocephalus, asymmetry of the cerebellar hemispheres, interhemispheric cyst, hyperechoic anterior horns.

** HPA-5b antibody from a previous pregnancy.

detected by ultrasound, unexplained fetal death, the detection of neonatal thrombocytopenia or maternal HPA antibodies in a prior pregnancy. Cases with ultrasound detected fetal ventriculomegaly were classified as mild (≥ 10 and < 12 mm), moderate (≥ 12 and < 15 mm), or severe (≥ 15 mm). Mismatches in HPA-types between the mother and the father and/or baby, and the results of serological HPA antibody investigations, were recorded and correlated to the clinical findings.

Results: During the four-year study period, 31 paired HPA genotyping tests were performed, equivalent to an estimated average incidence of 31/48.000 (0.06%) of births per year. During the study period the number of requisitions for paired HPA genotyping testing increased steadily; 2020 (4), 2021 (6), 2022 (8) and 2023 (13). Table 1 shows the clinical findings that prompted the paired HPA genotype testing. In most cases [18/31 (58%)] the genotyping was prompted by some degree of ultrasound- detected fetal ventriculomegaly. In 22/31 (71%) cases there was ≥ 1 HPA genotype mismatch between the mother and the father and/or baby. An HPA-antibody was detected in only 1/22 (4.5%) of cases with HPA-discrepancy. In this case, the newborn presented with petechiae and a large hematoma in the arm.

Summary / Conclusions: The use of paired HPA typing rose 3-fold during the observation period. However, there was only one case where a new HPA antibody was detected. Thus, the clinical benefit of paired genotyping appears to be low. We found no association between the most common indication ventriculomegaly (58%), and FNAIT but this result requires further validation.

P671 | Validation of a new commercial kit for fetal RHD detection from maternal plasma

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Background: Alloimmunization against the Rhesus factor (RhD) can result in Morbus Hemolyticus Neonatorum (MHN) in RhD-negative pregnant women carrying RhD-positive fetus. Maternal antibodies cross the placental barrier and enter the fetal bloodstream, causing hemolysis of fetal erythrocytes in RhD-positive fetuses. This leads to fetal anemia, which in severe cases can result in miscarriage. Alloimmunization can be prevented through the administration of anti-D immunoglobulin during pregnancy and postpartum. However, anti-D prophylaxis is unnecessary if the RhD-negative mother is carrying an RhD-negative child. Thus, knowledge of fetal RhD status enables targeted administration of anti-D prophylaxis in RhD-negative pregnant women carrying RhD-positive fetuses, thereby preventing unnecessary health risks and resulting in cost savings in cases of RhD-negative fetuses.

Aims: The aim was to compare the new commercial RBC-FluoGene fetal RHD Q kit (inno-train) with currently used commercial and in-house kits. The study utilized EDTA blood samples collected during early gestational weeks with longer transport times to assess the new kit's capability to detect low amounts or partially degraded cell-free fetal DNA.

Methods: Standard EDTA blood collection tubes were used. 21 maternal peripheral blood samples collected between gestational weeks 12 and 26 (mean 19.6) and transported for a maximum of 5 days (mean 3.3 days) at room temperature were typed in parallel using the mentioned test systems. The blood samples were centrifuged at 1700 g for 10 min. The resulting plasma supernatant was then transferred to a 13 mL tube and stored frozen at -80°C until cell-free DNA (cfDNA) extraction. The extraction of cfDNA was carried out from 2 mL of plasma using the QIASymphony DSP Circulating DNA kit, following the manufacturer's protocol. The elution volume was 60 µL. All test systems are real-time assays based on the TaqMan probe principle. With RBC-FluoGene fetal RHD Q, exon 5 and 7 of the RHD gene and an internal control are amplified in triplicates on QuantStudio 7 Pro Real-Time PCR System (ThermoFisher Scientific). The commercial kit and the in-house kit detect RHD exon 5, 7, 10, and an internal control in two mixes without replicates. With the RBC-FluoGene fetal RHD Q Kit, at least 3 out of the 6 replicates must be positive to classify the sample as RHD positive.

Results: In all 21 samples, the RBC-FluoGene fetal RHD Q kit confirmed the results obtained with the commercial kit and in-house test method. 7 out of 21 samples were RHD negative and 14 samples were RHD positive. The mean Ct values were very similar across different tests. In RHD-positive samples, the mean Ct value for exon 5 was 35.5 / 35.3 / 35.6, and 34.8 / 36.3 / 36.2 for exon 7 with FluoGene, the commercial kit, and the in-house method, respectively.

Samples with longer transport times showed slightly higher Ct values compared to samples with shorter transport times: 36.1 vs. 35.0 for exon 5 and 35.0 vs. 34.6 for exon 7 with the FluoGene Kit.

Summary / Conclusions: Results obtained with the new commercial kit are fully consistent with those of the current commercial test kit and in-house method for fetal RHD genotyping. The kit enables reliable detection of fetal cell-free DNA in standard EDTA blood collection tubes, even during early gestational weeks and with prolonged sample transport times. Therefore, the RBC-FluoGene fetal RHD Q Kit can be utilized for fetal RHD detection in maternal plasma.

P672 | Red blood cell alloimmunization among pregnant women in India—a systematic review and meta-analysis

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Background: Red blood cell (RBC) alloantibodies in pregnancy may develop against the fetal antigens, that are of paternal origin. This phenomenon poses a substantial risk of Hemolytic disease of the fetus and newborn (HDFN) during pregnancy. HDFN, may contribute significantly to perinatal morbidity and mortality. Several studies conducted in India have assessed the alloimmunization to RBC antigens in pregnant women, but a comprehensive systematic review in this context is lacking in the published literature.

Aims: The authors undertook a systematic review and meta-analysis, to evaluate the current evidence on RBC alloantibodies among pregnant women in India, and suggest recommendations, if any.

Methods: Authors searched the MEDLINE, SCOPUS, CINAHL and Google Scholar bibliographic databases with no restriction in search dates to identify relevant studies. PRISMA flow diagram was used to select the relevant studies. Case reports, comments, letters, conference abstracts, editorials and review articles were excluded. The primary data of the relevant studies were extracted as raw numbers. An aggregate effect size, weighted by sample size, was computed to provide an overall effect size across the studies and 95% confidence intervals (CI) were calculated. The relative weighted contribution of each study was also assessed.

Results: Out of 933 potentially relevant articles, 16 studies with cumulative sample size of 36174 pregnant women were selected. A total of 647 alloantibodies were identified in pregnant women. The prevalence of RBC alloimmunization exhibited a wide variation ranging from 1% (95% CI; 0%–4%) to 10.84% (95% CI; 7%–16%) in different studies. The overall meta-analytical prevalence of alloimmunization was observed to be 1.78 per 100 pregnant women (95% CI; 1.6%–1.9%) with zone-wise prevalence of 1.98%, 1.52%, 3.46%, 1.25% and 1.92% in the South, West, North, Central and East zone, respectively as shown in Table 1. 1.78% still seems to be highly significant, considering that India is the most populous country of the world. More than 85% of alloantibodies identified were associated with the Rh blood group system. Among clinically relevant alloantibodies, anti-D ranked as the most common, followed by anti-E,

P672 - Table 1: Zone-wise distribution of alloantibodies among pregnant women.

S.No.	Zone	Antibody positive cases	Total cases	Prevalence (%)
1	NORTH (Meena sidhu et al, Heeya gupta et al, Saikat mandal et al)	56	1614	3.46
2	EAST (L. Dayalaxmi et al, Archana Naik et al)	14	730	1.92
3	CENTRAL (Sangeeta pahuja et al, Sunil Golia et al)	55	4116	1.33
4	WEST (Kahar et al, Anshika yadav et al, Spruha K. Dholakiya et al, Meenakshi gothwal et al)	223	14664	1.52
5	SOUTH (Shanthala AM et al, Jophy varghese et al, B suresh et al, Soumya das et al, Rajat jagani et al)	301	15250	1.98
Total		647	36174	1.78

combination of anti-D+C, anti-c. After Rh alloantibodies, the next most common clinically significant alloantibodies belonged to the Kell blood group system. Asymmetry was noted in the funnel plot drawn to examine the publication bias in the results; observation being significant ($p < 0.001$).

Summary / Conclusions: The analysis of existing literature in India represented significant (1.78%) prevalence of RBC alloimmunization in pregnant women in India and highlighted that anti-D is the commonest allo-antibody found. Based on these results authors recommend adoption of RBC alloantibody screening as a standard of antenatal care for all pregnant women across India, and striving for 100% access of anti-D immunization.

fetuses were 18.83 (SD \pm 9.2, range 4-38 events) and 0.37 (SD \pm 0.9, range 0-4 events), respectively. Mean values for SRY dPCR events in 70 tests of plasma from women carrying male fetuses and 61 tests of plasma from women carrying female fetuses were 16.91 (SD \pm 8.7, range 3-46 events) and 0.16 (SD \pm 0.6, range 0-4 events), respectively. For HPA-1a and SRY dPCR reactions the limits of blank were 2 and 1 events, respectively, and the limits of detection were 6 events for both reactions.

Summary / Conclusions: The validated dPCR protocol determining fetal HPA-1a genotype as well as fetal (SRY) and total (HPA-1b) fractions of cfDNA in the same experiment has high accuracy which enables to implement the test into routine non-invasive diagnostics of immunized pregnant women.

P673 | Fetal HPA-1a genotyping using digital PCR

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Background: HPA-1a-negative pregnant women may produce antibodies against fetal HPA-1a antigen inducing alloimmune thrombocytopenia of a fetus and newborn (FNAIT). Non-invasive prenatal diagnostics predicts the fetal HPA-1a genotype assessing the risk of the disease.

Aims: To validate a digital PCR (dPCR) protocol for the detection of fetal HPA-1a together with fetal and total fraction of cell-free DNA.

Methods: 131 DNA samples (extracted using easyMag, Biomerieux) from plasma of 27 pregnant HPA-1a-negative women (15 to 30 week of gestation) were tested retrospectively to determine HPA-1a/1b (ITGB3), alleles and SRY gene in triplex assay in two replicates using Naica 6-color dPCR (Stilla). The results were compared to fetal/neonatal HPA-1a, SRY genotypes and sex.

Results: The dPCR results of HPA-1a/1b and SRY genotyping were concordant with the genotype of pregnant women and their children. Mean value for HPA-1b dPCR events in 131 tests was 853.4 (SD \pm 1939, range 206-16422 events). Mean values for HPA-1a dPCR events in 80 tests of plasma from women carrying HPA-1a-positive fetuses and 51 tests of plasma from women carrying HPA-1a-negative

P674 | Effect of different sensitization events on human leukocyte antigen alloimmunization among patients awaiting renal transplant

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Background: Anti HLA antibodies can be formed by exposure of foreign HLA molecules to the patient's immune system, this is known as alloimmunization. It can be the result from previous blood transfusion, pregnancy, and previous transplant. HLA class I, class II antibodies have also been detected even in the absence of sensitization events.

Aims: To determine the effects of different sensitization events on HLA Alloimmunization among patients awaiting Renal Transplant, tested at National Hospital Kandy.

Methods: Cross-sectional descriptive Study design was carried out at National Hospital Kandy for Two-year period starting from January 2019 to December 2020. HLA antibody testing data was collected from 300 patients with renal failure who tested at HLA laboratory of Blood bank. Effects, associations, and prevalence of different sensitization events on HLA alloimmunization were assessed. Data analysis was facilitated by SPSS version 25.0. Project was Ethically cleared by

ethics review committee in National Hospital Kandy & Post graduate institute of medicine.

Results: Mean age of the study participants was 40.18 years. (SD = 13.2 years) and a significant male predominance was observed within the study sample (N = 210:69.7%). Among class I antibodies HLA-A and HLA-B were identified among majority of the study participants (N = 273:91.0%). Among Class II antibodies, HLA -DR was noted among most of the study participants (N = 63:20.8%). Majority of the study participants had experienced a transfusion exposure (N = 88:52.4%). Not having a previous transplantation exposure significantly associate with the generation of HLA-B antibodies and presence of transplantation exposure significantly associate with HLA-C antibody generation. Majority of the participants with class I HLA antibodies (n = 174:91.5%) had produced compatible results during CDC crossmatch. Pregnancy and transfusion were identified as more vulnerable exposures for HLA- A and HLA- B antibody formation. Vaccination was associated with the formation of HLA-C antibodies. When class II antibody distribution with different sensitization events was considered, it was observed that DR antibodies were more common among participants with transfusion and vaccination status, DQ antibodies were common among participants with transplantation exposures. Majority of the participants with DSA were detected as individuals with incompatible crossmatch results (64.7%).

Summary / Conclusions: HLA class I antibodies are common among majority of the renal transplant awaiting patients and HLA-C antibody is less prevalent when compared to HLA-A and HLA-B antibody. It is challengeable to obtain an adequate and cost-effective compatibility assessment by performing CDC crossmatch only. CDC crossmatch having own limitations like, it gives positive results for the DSAs with complement binding capacity only, non-complement fixing antibodies cannot be detected, Low titer antibodies can be missed, DSAs can be missed if donor antigen expression is weak and test results vary depending on the viability of the donor cells in the sample. Positive DSA and positive CDC crossmatch transplants should be avoided due to strong association with ABMR and graft loss. But positive DSA with negative CDC crossmatch is a risk factor for transplantation as it can cause poor long-term graft survival but can proceed with pre transplant desensitization in case as lack of donor availability.

P675 | Abstract withdrawn

P676 | A single-center, retrospective analysis of anti-M alloimmunization during pregnancy and pregnancy outcome

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Background: Hemolytic disease of the fetus and newborn (HDFN) is caused by maternal alloantibodies that actively cross the placenta during gestation and destroy fetal erythroid cells. As a result, fetal anemia, hydrops fetalis and intrauterine fetal death (IUFD) may occur. Anti-M

antibodies is one of the most common non RHD antibodies in the pathogenesis of HDFN, previously report low risk leads to severe hemolysis. However, we present several life-threatening cases due to anti-M alloimmunization, and review the general guidelines and management of anti-M antibody during pregnancy through the context of a case series.

Aims: To investigate the detection rate of anti-M antibody in irregular antibody detection during pregnancy and notice pregnancy outcome, in order to increase the success rate of pregnancy.

Methods: We report a 37-year-old healthy pregnant G3P3 woman with anti-M antibodies who had experienced two times of miscarriages. This led us to statistically analyze the status of all pregnant women identified with irregular antibodies, including anti-M antibodies, at CMUH over the past decade.

Results: Irregular antibody screening tests had been applied to 280 pregnant women in ten years, from which 43 results were positive. There were 6 cases of intrauterine fetal death (13%). The identified antibodies were primarily anti-E (37%), followed by anti-Mia (23%). Among those 43 positive patients, 8 cases were positive for anti-M antibodies (18%). There was a total of 17 gravidity, with 7 miscarriages (41%), and 5 cases occurring in the third trimester (71%). Of the 11 live births, 6 cases of premature birth, including a set of twins. Complications during labor and delivery included pre-eclampsia, placenta abruption, placenta previa.

Summary / Conclusions: Anti-M antibodies indeed pose a high risk of miscarriage in pregnant women. It's essential to document this in the medical records and to provide early prevention and management.

P677 | Automated antibody identification and titration tests using solid phase red cell adherence allows to determine the prophylactic or immune nature of circulating anti-D (RH1) in pregnancy

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Background: Since 1970, generalization of antenatal immunoprophylaxis by anti-D immunoglobulins (RhIg) injections made the interpretation of anti-red blood cell antibodies screening during pregnancy more complex. In 1999, the laboratory of the CNRHP set up the anti-D microtitration test that allows to quantitate low levels of anti-D (between 0.008 and 0.12 UI/ml). It consists of a gel titration test, using a 0.03 UI/ml anti-D standard and papain-treated D positive cells. Comparison of found anti-D concentrations with concentrations expected according to the dose and date of RhIg injection based on pharmacokinetic curves, allows us to conclude on the prophylactic and/or immune nature of the circulating anti-D.

Aims: We performed a study on Neo Iris (Werfen Immucor) to determine if a quantitative approach based on agglutination values calculated by the analyzer in a solid phase red cell adherence identification

test could help to determine the nature of anti-D. We also wanted to evaluate the potential of the automated anti-D titration test as a substitution method for the microtitration test.

Methods: - Agglutination values (/99) found in the D positive wells of the identification test were collected for anti-D standards ranging from 0.008 to 0.12 UI/ml. 18 blood samples of women with low level anti-D were analyzed and comparison was made between the /99 agglutination mean values of positive wells and the anti-D concentration found by microtitration. Using a 0.12 UI/ml anti-D standard, 2 quality controls and 33 patient's samples with anti-D concentrations ranging from 0.011 to 0.58 UI/ml were analyzed in the titration test. Scores (/99) were calculated adding /99 values for each dilution wells showing positive agglutination. Sample's /99 scores were reported to the 0.12 UI/ml standard /99 score and anti-D concentration could be calculated for each sample and compared to those found by microtitration test or continuous flow analysis.

Results: A good correlation was found between the mean of the /99 agglutination intensity values in the D positive wells of the identification test and the microtitration values (Spearman, $p = 0.0014$, $r = 0.69$). Linearity was observed between 0.008 and 0.03 UI/ml (Deming $p = 0.0003$, equation $Y = 17.5X - 34$). Anti-D titration test using /99 scores and a 0.12 UI/ml standard to report values gave concentrations that were well correlated to the microtitration test and/or to continuous flow analysis concentrations (Spearman, $p < 0.0001$, $r = 0.91$). Values were comparable and linearity was obtained between 0.06 and 0.18 UI/ml (Deming $p = 0.0008$, equation $Y = 0.9114X + 0.018$).

Summary / Conclusions: Comparison of the sample's agglutination values (/99) in the positive wells of the solid phase red cell adherence identification test with the mean values of the 0.03UI/ml anti-D standard allows to have a quantitative approach that could be useful at delivery, as anti-D concentrations 9 to 13 weeks after routine antenatal prophylaxis are expected to be within the linearity range. In other cases, when targeted Rhlg prophylaxis has been done a few weeks before sampling, an anti-D titration test could be performed with calculation of the /99 scores and report to the 0.12 UI/ml anti-D standard /99 score. It allows to calculate accurate anti-D concentrations and ensure they are within the expected range based on Rhlg dose and injection date. If the sample's found values are too high, anti-D immunization despite prophylaxis should be considered.

P678 | Fetal RHD detection from maternal plasma comparison of two assays developed according to the workshop report on extraction of foetal DNA from maternal plasma

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Background: Fetal RHD genotyping from maternal plasma is a groundbreaking technique in prenatal diagnostics. This non-invasive

method offers crucial insights into the RhD blood type status of the fetus, thereby aiding in the management of RhD alloimmunization potentially leading to hemolytic disease of the newborn (HDN). The clinical implications of fetal RHD genotyping are significant. For RhD-negative mothers carrying RhD-positive fetuses, knowledge of the fetal RhD status allows for timely interventions to prevent RhD alloimmunization. These interventions may include administration of Rh immunoglobulin (Rhlg) to suppress the maternal immune response, thus reducing the risk of HDN in subsequent pregnancies. Conversely, RhD-negative fetuses can be identified early, sparing unnecessary Rhlg administration and associated costs.

Aims: The aim was to compare the new commercial RBC-FluoGene fetal RHD Q kit (inno-train) with currently used inhouse kits. RBC-FluoGene fetal RHD Q detects cell-free fetal DNA in maternal plasma and detects the presence of exons 5 and 7 of the RHD gene in triplicates, identical to the currently used inhouse test system.

Methods: 35 plasma samples from different weeks of gestation and after max 5 days of transport have been typed in parallel with the mentioned test systems. DNA isolation was done using Qiamp Blood kit mini. All test systems are real-time assays based on the TaqMan probe principle. For amplification, the real-time devices FluoQube® and StepOne Plus were used. With RBC-FluoGene fetal RHD Q, at least 3 of the 6 replicates have to be positive to call the sample RHD positive. The currently used technique requires 5 of 10 replicates to be positive to call the sample RHD positive.

Results: 32 out of 35 samples showed concordant results. 19 samples were typed with both systems as RHD positive; 9 samples were typed with both systems as RHD negative; 3 samples showed very early Ct values which was caused by a maternal partial RHD gene (RHD*01W.3, *08N.01; *06.02). 3 discrepancies were observed. These are currently under investigation.

Summary / Conclusions: Both test systems showed reliable results and are suitable for detection of fetal RHD from maternal plasma. The 3 discrepant test results will be further investigated. In conclusion, fetal RHD genotyping from maternal plasma represents a remarkable advancement in prenatal diagnostics, offering a safe, accurate, and non-invasive means of assessing the fetal RhD status. The new RBC-FluoGene fetal RHD Q kit is suitable for fetal RHD detection in maternal plasma.

P679 | Does the current guidance for the use of prophylactic anti-D meet the requirement for pregnant women?

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Background: Haemolytic disease of the fetus and newborn (HDFN) is caused by maternal antibodies either pre-existing or formed from a previous pregnancy/birth. The most significant cause of HDFN is incompatibility between mother and fetus due to the Rh system. Sensitisation can occur via transplacental passage of D positive fetal red

blood cells (RBCs) during pregnancy, with the formation of anti-D. Administering prophylactic anti-D (PAD) immunoglobulin at routine stages during pregnancy and at delivery to non-sensitised D negative women has reduced the risk of sensitisation. PAD dose relies on accurate calculation of feto-maternal haemorrhage (FMH) volume via the Mollison formula. This is based upon an expected maternal weight of 75 kg and associated packed red cell volume (PRCV) of 1800 mL. Maternal obesity has more than doubled over the previous decades, raising concerns that the Mollison formula does not reflect the current population of pregnant women.

Aims: This study aims to understand if the current guidance for the use of PAD meets the requirements of the present-day pregnant population.

Methods: Weight, height, age and haematocrit were collected at Liverpool Women's Hospital antenatal clinic. PRCV of all study participants were calculated using formulas from Pearson (1995) and Nadler (1962) formulas and applied in a modified Mollison formula. Wilcoxon two-tailed signed rank test was applied to compare the calculated: Pearson and Nadler PRCV mean with the theoretical PRCV mean; Pearson (1535 ml) and Nadler (1501 ml); Results by flow cytometry that relate to significant FMH volumes by the Mollison formula (0.1%, 0.2% and 0.6%) with values obtained considering the Pearson and Nadler PRCV.

Results: Descriptive population data identified a median height of 165 cm (Max = 180cm; Min = 155cm), weight of 71.1 Kg (Max = 139.9kg; Min = 45kg), and haematocrit of 0.34 l/l (Max = 0.43l/l; Min = 0.25l/l). A statistically significant difference between the Mollison PRCV and the Pearson PRCV ($p < 0.0001$), or the Nadler PRCV ($p < 0.0001$) was identified. There was also a statistically significant difference between the PRCV calculated using the Pearson formula when compared to the Nadler formula ($p < 0.05$). Comparison of FMH volumes calculated using the Standard Mollison PRCV formula known to be clinically significant Vs those calculated with the Pearson or Nadler PRCV adapted Mollison formula demonstrated a statistically significant difference for all comparisons ($p < 0.0001$ and $p = 0.0001$ respectively). A statistically significant difference was also identified when comparing calculated FMH volumes using the Pearson PRCV adapted Mollison formula Vs the Nadler adapted formula ($p < 0.05$).

Summary / Conclusions: This study suggests that considering the true maternal PRCV in an adapted version of the Mollison calculation provides a more accurate and lower FMH result. This allows for improved management of D negative mothers by reducing unnecessary attendance at postnatal clinic for PAD top-up and reduces the associated financial and management burden for the NHS.

P680 | Positive Coombs test in newborn in a tertiary Portuguese hospital—cases review in the years 2021-2023

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Background: Hemolytic disease of the fetus and newborn (HDFN) is caused by maternal antibodies produced against the fetus/newborn red blood cells (RBC), causing its destruction. HDFN may occur due to ABO, Rh or other minor blood group incompatibilities. HDFN can cause fetal hydrops or neonatal jaundice. Direct Antiglobulin Test (DAT) detects maternal antibodies bound to fetus/newborn RBC and aids in the diagnosis of HDFN.

Aims: The aim of this study was to investigate the frequency, causes and consequences of DAT+ in newborns in a tertiary Portuguese hospital.

Methods: We retrospectively reviewed all results of DAT+ in newborns born in our hospital between 2021-2023, along with the laboratory and infants' medical records. The following data was recorded: sex, gestational age, DAT results and strength (MARSH score), infant/maternal blood group and antibody screening and identification, bilirubin peak level and treatment with phototherapy or immune globulin therapy.

Results: Over these three years 5849 babies were born. Out of the 1733 DAT performed, 210 were positive (12.1%). During this period there was a slight increase in the number of births (6.8%), however, there was a higher rate of DAT realized (34.2%), which resulted in a notable increase in positive results (40.2%). In 43.3% of the cases, DAT+ was due to ABO incompatibility, in which 75.8% belonged to O/A blood group and the other 24.2% belonged to B/O blood group, where O is the mother's and A or B is the newborn's blood group. 33.3% of the DAT+ cases were caused by Rh incompatibility, where 94.3% were anti-D antibodies, 2.9% were anti-D+anti-C and anti-D+anti-E and the other 2.9% were equally divided among anti-Cw and anti-E antibodies. In 17.6% of the cases, both factors mentioned were present, while in 5.7% it was unclear. Clinically, in 58.6% of the DAT+ cases patient developed jaundice, in 1.9% anemia and in 3.3% anemia and jaundice. Fetal hydrops or encephalopathy wasn't detected in any newborns. In 90.2% of the cases in which jaundice was detected, the patient received treatment: 94.6% needed phototherapy and the other 5.4% needed intravenous immune globulin as well. Over the studied period there was a significant increase in the number of cases requiring phototherapy (50.1%), but no exchange transfusion was necessary. There was a positive correlation between phototherapy and the bilirubin peak level. Thus, when the bilirubin peak is above 95th percentile, phototherapy was necessary in almost all cases; even when bilirubin peak level is between 75-95th and 40-75th percentiles, phototherapy was needed in 89% and 64% of the cases, respectively. In these cases, additional risk factors were considered like prematurity, low weight and development of jaundice under 24 h. DAT+ was considered as a risk factor in some cases too. 13 babies were transferred to ICU, 10 for intensive phototherapy due to blood group incompatibilities and 3 due to other complications.

Summary / Conclusions: While ABO incompatibility continues to be the main reason for DAT+, as concluded by other studies, other causes should also be explored. The relevant impact of DAT+ on the diagnosis of HDFN should be considered and that is present in our results since DAT+ was used as an additional risk factor. This has implication on HDFN treatment as the majority of newborns needed treatment even when bilirubin peak levels were under 95th percentile.

In general phototherapy was sufficient, intravenous immune globulin was rarely needed and none required exchange transfusion.

P681 | Evaluating ABO-related neonatal hemolysis—incidence and treatment patterns at a tertiary facility

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Background: ABO incompatibility has a low frequency of about 15% of all live births. However, it does not result in clinically significant haemolytic disease. Between three and six months of age, individuals naturally begin producing A and/or B antibodies to antigens they do not have. As a result, ABO haemolytic disease of the newborn can occur since the first pregnancy. It is more frequent in mothers with blood type O and happens in less than 4% of pregnancies.

Aims: We retrospectively analyzed births from the past five years with positive direct Coombs tests due to ABO incompatibility to investigate the association between these cases and: length of hospitalization, admission to the neonatal intensive care unit (NICU), and the relationship between elevated bilirubin levels and the need for phototherapy or exchange transfusion.

Methods: Between January 1, 2018, and December 31, 2023, our hospital systematically performed ABO and Rh group testing, along with direct Coombs test, on cord blood samples of all newborns. We retrospectively analyzed this cohort ($n = 6,998$) and identified newborns with positive direct Coombs test and ABO incompatibility. The polyspecific Coombs test was performed using Ortho BioVue System® cards of QuideOrtho® with analysis in the Ortho-Vision automated immunohematology analyzer. The monospecific Coombs test was conducted with a manual technique using DG Gel 8 Direct Coombs cards of Grifols® and their DG Gel DC scan device.

Results: 2.1% of neonates ($n = 148$) exhibited a positive Coombs test indicative of ABO incompatibility. This is within the expected range of prevalence for this condition. Hospitalization duration followed a median of 3 days across all neonates. Haemoglobin and bilirubin had median values of 16 g/dL and 9.3 mg/dL, respectively. Among newborns requiring admission in intensive care unit for phototherapy ($n = 38$), a median hospitalization period of 4 days was observed. Their median haemoglobin was of 16.3 g/dL, while the median bilirubin level increased to 14.2 mg/dL, with a maximum recorded value of 23.75 mg/dL. Only one neonate required exanguinotransfusion, underscoring the mild clinical course associated with ABO incompatibility in most cases.

Summary / Conclusions: ABO incompatibility in newborns generally presents with a mild clinical course, aligning with existing literature. This suggests a good prognosis for most affected infants. Vigilance in monitoring bilirubin levels is crucial, as elevations can occur beyond 48 h. However, phototherapy initiation should be guided by established bilirubin thresholds based on the newborn's age for optimal management. ABO incompatibility typically manifests with limited and

non-chronic health consequences. Early and consistent communication with pediatricians is essential for ensuring appropriate follow-up and prompt intervention if necessary.

P682 | A study to identify potential risk factors for maternal alloimmunization

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Background: Red blood cell (RBC) alloimmunization during pregnancy is a significant cause for neonatal mortality and morbidity. The explanation for why only a select group of patients (responders) form alloantibodies to donor RBCs despite multiple exposures through transfusion or pregnancy is both complex and poorly understood. Separate studies have identified epidemiological factors, such as sex, race and certain medical conditions, to be independently predictive of alloantibody formation. Given the associated morbidity and mortality in transfusion and pregnancy, determining specific factors associated with increased risk for RBC alloimmunization has been a focus of active investigation as this will help the clinician in frequent monitoring and potentiate the safe clinical outcome.

Aims: To identify the potential risk factors associated with maternal RBC alloimmunization.

Methods: In this prospective observational study (from November 2018 to June 2019), antenatal females attending Obstetrics clinic after taking consent enlisted into the study. Age, demographic data, identified epidemiological factors and certain medical conditions were collected. Blood grouping and red cell antibody screening of antenatal mothers were performed at 1st visit and again antibody screening was repeated after 28 weeks of gestation by column agglutination technology using 3 cell panel. Positive cases were identified using 11 cell panel. Using statistical methods like odds ratio, relative risk, correlation and regression association between different demographic data and identified epidemiological factors and certain medical conditions were calculated.

Results: A total of 652 pregnant women who attended antenatal care were enrolled in the study following inclusion-exclusion criteria. Out of 652 antenatal mothers, 18 (2.76%) had positive antibody screening (ASP) and 634 (97.2%) of the participants had negative antibody screening (ASN). The odds ratio (95% CI) for presence of bad obstetric history in antibody screening positive group as compared to antibody screening negative group was 8.06 (3.1-20.97) and relative risk (95% CI) was 7.25 (3.02-17.12). The odds ratio (95% CI) for H/o Previous Blood Transfusion in antibody screening positive group as compared to antibody screening negative group was 2.78 (0.89-8.73) and relative risk (95% CI) was 2.67 (0.94-7.34). The odds ratio (95% CI) for Maternal Rh D Antigen: Negative in antibody screening positive group as

compared to antibody screening negative group was 38.42 (10.86-135.92) and relative risk (95% CI) was 32.05 (10.1-101.6). The odds ratio (95% CI) for Transfusion in Present Pregnancy: Present in antibody screening positive group as compared to antibody screening negative group was 0.19 (0.01-3.21) and relative risk (95% CI) was 0 (0-1.51). There was no statistically significant difference between the 2 groups in terms of age in years ($W = 6169.500$, $p = 0.556$). There was a statistically significant difference between the two subgroups ASP and ASN in terms of gravida ($W = 8256.500$, $p = 0.001$), with the median gravida being highest in the group: antibody screening positive group.

Summary / Conclusions: This study identifies that bad obstetrics history, increasing gravida, maternal Rh D negative blood group, history of previous blood transfusion are risk factors for maternal red cell alloimmunization. Whereas age of the mother and blood transfusion in present pregnancy does not increase the risk of RBC alloimmunization among antenatal women.

P683 | Evaluation of the causes of a positive Direct Antiglobulin Test (DAT) in newborns

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Background: Some new-borns have unexpected haemolysis after delivery with a positive DAT, high bilirubin and lower than expected haemoglobin level. These can be due to IgG antibody crossing the placenta and attaching to the baby's red cells and causing red cell destruction. Use of routine antenatal anti-D prophylaxis (RAADP) to prevent production of immune anti-D due to sensitisation with Rh D Positive red cells can cause a positive DAT as this is an IgG preparation and can cross the placenta. The other and more common reason for a positive DAT has been ABO incompatibility between the baby and the mother—usually where the mum is blood group O, and the baby is A or B.

Aims: To evaluate the cause of positive DAT's in new-borns and to see if the level of antibody present correlates with the degree of positivity.

Methods: All positive DAT results are monitored and those in new-borns were assayed further using elution to remove the antibody coating the red cells with subsequent testing to either identify the antibody or to determine whether A or B cells were present. A titre of the antibody present in the mothers' plasma was also tested to determine the level. All antibody testing and titres were done using the Immucor NEO Iris™. The elution's were performed using Immucor Gamma-ELU-KIT II and the DAT's were tested using BioRad cards.

Results: In total 9 new-borns were tested, and the results are shown in Table 1 below:

The majority of positive DAT's were due to ABO incompatibility and the titres of anti-A/B were high at ≥ 256 . One baby that had a DAT 1+ with anti-B coating the red cells required an exchange transfusion as the bilirubin went to 621 $\mu\text{mol/L}$. This was so unexpected at the time that other genetic testing was undertaken to ensure there was

P683 - Table 1: Table to show the blood groups of mother and baby and results of any antibody in the eluate and its strength in the plasma.

Baby Group	DAT	Mum Group	Antibody Screen	IgG Titre
O+	1+	A-	RAADP in eluate	Anti-D = 1
A+	3+	O-	RAADP in eluate	Anti-D = 1
A+	2+	O+	Anti-A1 4+ in eluate	Anti-A1 = 512
A+	1+	O+	Anti-A1 2+ in eluate	Anti-A1 = 256
B+	1+	O+	Anti-B 2+ in eluate	Anti-B = 256
A+	1+	O-	Anti-A 3+ in eluate	Anti-A1 = 256
A+	1+	O+	Anti-A 2+ in eluate	Anti-A1 = 512
A+	1+	A-	RAADP in eluate	Anti-D = 2
O+	2+	O+	Anti-Jk(a)+E in eluate	Anti-Jk(a) = 2 E = 8

no other reason for this bilirubin result, however, the test came back negative. Any positive DAT's due to prophylactic anti-D were low and did not cause a raised bilirubin and this was also the case in the baby with anti-Jk(a) and E on their red cells.

Summary / Conclusions: The most common cause of a positive DAT in new-borns is ABO incompatibility between mum and baby with the titre of anti-A/B often being quite high and this can also cause extremely high bilirubin levels. The strength of the DAT did not correlate with the antibody titre as the majority were only 1+ but these were the result of extremely high titre anti-A/B results.

P684 | Red cell alloimmunization in Intrauterine transfusion cases in the Royal Hospital (10 years experience)

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Background: Trans-placental passage of maternal antibodies that bind to fetal erythrocyte antigens lead to fetal or neonatal hemolysis. If untreated, fetal anemia can lead to fetal heart failure, hydrops or even death. Intrauterine Transfusion (IUT) is the basis of treatment for fetal anemia for a variety of causes. Maternal allo-antibodies will cause haemolytic anaemia, although the Rh or D antigen is the most common cause of red cell alloimmunization, however antibodies to c, Kell, Duffy, and Kidd antigens can be the other major causes of red cell allo-immunization in pregnancy.

Aims: The main aim of this study was to monitor the blood group antibodies that can cause severe intrauterine haemolysis for foetus in the Royal hospital which led to IUT.

Methods: A retrospective study of all IUT cases done in the Royal Hospital since 2013. Twenty seven cases of IUT were done over the period from 2013 to 2023.

Results: Among which twenty five were due to allo-immunisation i.e. 92%. 81% of these cases were Rh negative and 19% of the cases were Rh positive. All the Rh Negative cases were due to presence Anti-D, 52% were anti-D&C, 33% were Anti-D solely, anti-D&E were 10%, 5% were anti-D&Jk^a cases. All anti-D cases were with high anti-D titres as 2048. Although the majority of the Rh negative cases are Anti-D&C, however due to the un-availability of molecular testing it is not clear if there is involvement of anti-G or not. The Rh positive cases were due to anti-Kell, anti-Js^b, anti-Jk^a and a case of anti-E, Jk^b, S with auto-e who was a Sick cell disease patient.

Summary / Conclusions: Although all Rh negative cases needed IUT were anti-D, it is confirmed that antibodies against Kidd system can be severe and needs IUT. 99% of the cases survived and monitored after birth

P685 | Does breastfeeding increase transfusion needs in newborns with haemolytic disease due to anti-D?

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Background: The anti-D antibody is responsible for the most severe forms of hemolytic disease in the fetus and newborn (HDFN). Depending on the severity, the therapeutic approaches may range from intrauterine transfusion (IUT) to postnatal exchange transfusion (ET), but sometimes hemolysis persists over time and there is speculation about the possibility that breastfeeding contributes to it.

Aims: To assess the association between the presence of anti-D in breast milk and the persistence of HDFN.

Methods: Descriptive analysis of two cases of HDFN with persistent hemolysis after several ETs and presence of high-titer anti-D in breast milk.

Results: Case 1. Newborn O, D+, born to woman O, D-, correct prophylaxis with anti-D gamma globulin in previous pregnancy, immunized with anti-D titer of 512 and anti-C titer 1 (anti D+G) in week 32. Measurement of MCA-PSV ruled out the presence of fetal anemia at that time and in subsequent measurements. At week 37, pregnancy was terminated due to MCA-PSV of 88.8 cm/sec and fetal hemoglobin (Hb) of 8.5 g/dl. The newborn did not present hydrops, but Hb and bilirubin (Bb) in cord blood were 4.4 g/dl and 6 mg/dl. Exchange transfusion with red blood cells O, D- was performed. At discharge, Hb level was 13.5 g/dl. At 2 months, the Hb level decreased to 7 g/dl. Anti-D was demonstrated in breast milk at a titer of 16. Breastfeeding was discontinued and no further transfusions were required. Case 2. Newborn A, D+, born to woman A, D-, without correct prophylaxis with anti-D gamma globulin in previous pregnancy, presence of anti-D titer 128 and anti-C titer 8 detected in the first trimester. Subsequent controls showed an increase in titer but no ultrasound evidence of anemia or in PSV-ACM measurement. At week 29, anti-D titer was 2048 and anti-C titer 64, MCA-PSV 64.12 cm/sg (1.62

MoM), without hydrops, fetal Hb 7.58 g/dl. Maternal obesity prevented IUT. He was born in week 32, Hb and Bb in cord blood were 8.2 mg/dl and 6.9 g/dl. After four ETs with O D-, transfusion of a unit O D- and two doses of intravenous immunoglobulins (IVIg), anemia persisted with signs of hemolysis. Anti-D was demonstrated in breast milk at a titer of 64. Discontinuation of breastfeeding was advised.

Summary / Conclusions: Cases of persistent hemolytic anemia due to anti-D present in breast milk are very rare, as oral intake of Ig does not allow for good gastrointestinal absorption, although a synergy of action between the anti-D acquired intrauterinely and acquired through breastfeeding is expected. In our cases, it is possible that anti-D present in breast milk contributed to the persistence of anemia. Therefore, discontinuation of breastfeeding should be considered in cases that do not respond to standard treatment.

P686 | Anti-G with concomitant anti-D—a case report in a pregnant woman

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Background: Hemolytic disease of the fetus and newborn (HDFN) is due to maternal blood group antibodies that cause fetal red cell destruction. This process leads to fetal anemia and in severe cases can progress to edema, ascites, heart failure and death. The Rh blood group system remains the most common cause of HDFN. The incidence of HDFN caused by RhD sensitization has decreased markedly since the introduction of anti-D immunoglobulin (RhIg) prophylaxis. It is important at an early gestational age (GA) to determine the woman's blood group and to screen the presence of any alloantibody. If RhD negative woman is non-immunized by anti-D, RhIg is offered after events that may have led to fetomaternal hemorrhage, at GA 28 weeks, and again after delivery if the new born is RhD positive. The G antigen belongs to the Rh blood group system and is produced by RhD gene and RHCE gene. Anti-G antibody can lead to HDFN. Anti-G antibodies appear to be anti-D plus anti-C in serologic testing. Anti-D, anti-C anti-G can be distinguished by adsorption. For obstetric patients it is important to provide RhIG profilaxis to woman with anti-G to prevent immunization to D when indicated.

Aims: The aim of this study is to describe the clinical evolution of a woman sensitized to anti-D and anti-G.

Methods: Data collection through medical records.

Results: A 37-year-old pregnant woman was monitored during her 1st trimester visit. Her sample was sent for blood grouping, typing and antibody screening. The woman had a history of pregnancy and childbirth 2 years ago. At that time, the antibody screening was negative, and RhIG was administered at week

28 and after delivery. The newborn group was A+. The patient had no history of blood transfusion. Pregnant woman blood group was A Rh (D) negative, ccee (Rh phenotype) and the antibody screen was positive. Further workup revealed anti-C title 1/2 and anti-D title 1/128. In view of the presence of combined anti-C and anti-D, the presence of anti-G was suspected. Differential adsorption and elution were performed with the patient's plasma to evaluate the presence or absence of anti-G. The presence of combined anti-D and anti-G was confirmed. The ultrasound revealed a dichorionic diamniotic (DCDA) pregnancy. The fetuses were monitored with twice-weekly middle cerebral artery peak systolic velocity (MCA-PSV) values. Every 4 weeks blood sample and antibody screen were performed to the woman for antibody titles. At 35 weeks of gestation one of the fetuses developed signs of fetal anemia (MCA-PSV >1.5 multiples of median [MOM]), estimated fetal hemoglobin was 7.7 g/dl. Prepartum antibody titles were Anti-D 1/2048 and Anti-G 1/32. An attempt was made to perform an intrauterine transfusion; the procedure was rejected due to persistently unfavorable position of the fetus. The patient went into labor, twin vaginal birth. The 1st newborn weighed 2820 g, the umbilical cord group was A Rh (D) Negative, phenotype was Ccee, DAT was strongly positive and the hemoglobin and bilirubin results were 6.4 g/dl and 8.9 mg/dl. The 2nd newborn weighed 2500 g, umbilical cord group was A Rh (D) Negative, Ccee, DAT was strongly positive and the hemoglobin and bilirubin results were Hb 8.7g/dL and Brb 12 mg/dL respectively. In both new borns bilirubin gradually increased despite phototherapy and exanguinotransfusion was performed. Both babies had a good evolution.

Summary / Conclusions: With these results we can say that the HDFN was due to anti-G. Anti-G can explain the HDFN in a new born RhD negative, C positive.

P687 | Abstract withdrawn

P688 | Abstract withdrawn

P689 | Anti-K antibody, repeated intrauterine transfusions and further immunisation—how to survive complications

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Background: Prenatal care is at a high level in the Czech Republic and immunohaematology controls in pregnancies are well established. There are about 5000 (5%) positive screenings of irregular red cell antibodies per year that need further examination. Clinically significant antibodies are found in approximately 1500 (1.5%) pregnancies while about 500 (0.5%) fetuses have complementary antigens. Pregnant women with risk of haemolytic disease of fetus and new-born

are referred to centers with comprehensive diagnostic and therapeutic care. Severe anaemia with need of intrauterine transfusion develops in 25 to 50 fetuses per year.

Aims: These case studies aim to analyse two pregnancies with anti-K antibody in which severe anaemia of fetuses required repeated intrauterine transfusions. Further alloimmunisation appeared in both cases. Strategic choice of most appropriate erythrocytes for following transfusions is discussed.

Methods: Standardised serologic methods for immunohaematology examination of incompatible pregnancies were used. Identification of irregular antierythrocyte antibodies was done with certified panels of diagnostic erythrocytes (both papain and LISS/IAT). Complementary antigen/phenotype of mother and the foetus were revealed by certified diagnostic sera (foetal blood was obtained from cordocentesis before first intrauterine transfusion). IgG subtype and titration helped to assess clinical significance of antibodies. All results were reported to obstetrician and fetuses were monitored by duplex ultrasound (middle cerebral artery peak systolic velocity was measured to predict anaemisation). We found donors of appropriate phenotype in our donor-register. Compatibility testing and necessary adjustment of chosen erythrocytes were done. Transfusions were administered in the umbilical vein.

Results: In the first case, anti-K immunisation required five intrauterine transfusions. Additional antibody with specificity anti-c appeared before the fifth one and erythrocytes of different phenotype had to be chosen for this transfusion. In the second case, anaemisation occurred even earlier and situation became more complicated when after each intrauterine transfusion another antibody appeared. Concurrent detection of anti-K, anti-Fy(a), anti-S, anti-Jk(a) and anti-f antibodies deserved special preparation before every other transfusion including search for donors of appropriate phenotype.

Summary / Conclusions: In described cases of severe foetal anaemia due to anti-K antibody both pregnancies became transfusion-dependent. Intrauterine transfusion is an invasive procedure and among others carries a risk of further alloimmunisation. Perfect cooperation within a multidisciplinary team of obstetric and immunohaematology specialists is needed to manage this challenge.

P690 | Anti-G with concomitant Anti-D and severe Hemolytic Disease of Newborn (HDFN)—a case report

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Background: The G antigen belongs to the Rh blood group system and is produced by the RhD and RHCE genes. It is present in almost all D-positive or C-positive red blood cells, while absent from those with D-negative and C-negative red cells. This phenotype was first described by Allen and Tippett in 1958. Some reports showed

evidence of HDFN due to anti-G. The D antigen is highly immunogenic, and the presence of anti-D can cause severe HDFN.

Aims: Report a case of anti-G in combination with anti-D in a pregnant woman detected after delivery with a severe Hemolytic disease of the newborn.

Methods: A 42-year-old woman, first pregnancy by In Vitro Fertilization (IVF), monitored during her antenatal visit, with controls during first and second trimester with antibody screening negative. The patient had no history of blood transfusion and RHIG (RH Immunoglobulin) administration at 28 weeks of pregnancy. At 30 weeks of pregnancy, the patient goes to the emergency room due to bleeding. There was no RHIG administration at this moment.

Results: During the control of 38 weeks of pregnancy, echography showed the fetus with decreased variability and poor uterine dynamics; therefore, it was decided to induce labor by urgent cesarean section. At birth, the newborn presented Hb: 4 g/dL, coagulopathy, Bilirubin: 8 mg/dL, LDH: 5305 UI/L, and the maternal-fetal screening showed Antibody screening was positive (4+) by gel technique. Antibody identification 15 cell panel, gel technique, revealed the presence of anti-C and anti-D specificities. Given the presence of combined anti-C and anti-D, anti-G was suspected, and further testing by double adsorption and elution confirmed the presence of anti-D and anti-G antibodies.

Summary / Conclusions: Anti-D can cause severe HDFN. Anti-G could cause HDFN with high titers of anti-G consistent with moderate to severe HDFN. The differentiation of anti-D, anti-C, and anti-G specificities during pregnancy is fundamental for proper antenatal management of the patient. The HDFN with a combination of antibodies anti-G and anti-D is rare and can lead to confusion identified anti-G due to this antibody in panels of identification revealing the presence of anti-C and anti-D specificities. Due to having combined anti-C and anti-D, the presence of anti-G should be suspected.

P691 | Maternal-fetal conflict due to anti-K and anti-Jkb antibodies detected during the second trimester of pregnancy, with fetal demise at 37 weeks of gestation

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Background: We describe the case of a pregnant woman admitted due to third trimester thrombocytopenia, referred from another healthcare center. Immunohematological studies were repeated, leading to the diagnosis of alloimmunization with anti-Kell (K) and anti-Jk (b) antibodies. Ultrasound studies did not show fetal affection, but fetal demise occurred at 37 weeks. Fetal immunohematological studies confirmed the maternal-fetal conflict, with a positive direct Coombs test (DCT). Anti-Kell and anti-Jk(b) antibodies were identified in the eluate of fetal red blood cells, with a positive Kell and

Jk(b) phenotype. The autopsy findings indicated signs of pulmonary and other organ hemorrhage, as well as extramedullary hematopoiesis. Congested vessels associated with edema were observed in the brain. In conclusion, based on these findings, it cannot be determined that the cause of fetal death was due to this conflict.

Aims: Discuss the finding of anti-Kell and anti-Jkb antibodies and their implication in the course of pregnancy, including their possible association with fetal death.

Methods: This is a descriptive observational study, clinical case report type.

Results: We confirm the fetal-maternal conflict due to antibodies of anti-Kell (K) and anti-Jkb specificity using elution techniques, subsequently identifying them. The relationship with fetal demise could not be determined.

Summary / Conclusions: This clinical case illustrates a fetal-maternal conflict caused by the presence of anti-Kell (K) and anti-Jkb antibodies, which unfortunately resulted in fetal demise at 37 weeks of gestation despite timely monitoring. Although immunohematological studies confirmed the fetal-maternal conflict and identified anti-Kell and anti-Jkb antibodies in the eluate of fetal red blood cells, as well as a positive Kell and Jk(b) phenotype, neither the ultrasound studies during gestation nor the findings of fetal pathological anatomy definitively established that the cause of death was due to perinatal hemolytic disease resulting from this conflict.

P692 | A retrospective review of cell-free fetal DNA screening errors with information technology as a contributory factor reported to SHOT

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Background: Non-invasive high throughput cell-free deoxyribonucleic acid (cffDNA) screening for RHD was introduced in the United Kingdom in 2016 to improve management of D-negative pregnancies. In non-immunised D-negative pregnancies, cffDNA screening prevents unnecessary administration of anti-D immunoglobulin (Ig) where fetus is predicted as D-negative and decreases the number of clinic appointments required. Information technology (IT) can support safe and appropriate management of anti-D Ig by improving visibility of cffDNA screening results and enabling algorithms based on pregnancy status, maternal D-type and cffDNA results. Serious Hazards of Transfusion (SHOT) has been collecting data relating to errors in anti-D Ig management associated to cffDNA screening and IT.

Aims: Identify common errors in management of anti-D Ig relating to cffDNA screening results where IT was identified as a contributory factor.

Methods: Retrospective audit of SHOT reports analysed from 2019 to 2022 using a search of the terms 'cffDNA' and 'IT'.

Results: 46 cases were identified where IT contributed to error in anti-D Ig management relating to cffDNA screening results. Errors in the clinical area accounted for 28/46 (60.9%) of cases while 18/46 (39.1%) occurred in the laboratory. There were two cases where the primary error occurred in the laboratory, but additional errors were identified in the clinical area. In more than half of the cases, 28/46 (60.9%), cffDNA screening results were available in clinical and laboratory IT systems ($n = 7/28$ laboratory, $n = 21/28$ clinical) but were not checked prior to anti-D Ig issuing/administration. Other common laboratory errors included failure to heed warning flags in the laboratory information management system (LIMS) ($n = 3/18$) and cffDNA results checked from previous pregnancy ($n = 3/18$). In two cases, the lack of rules in LIMS for warning when anti-D Ig issued on a negative cffDNA result was identified as a contributory factor. In one case, the warning flag was not added to the clinical notepad by laboratory staff, however the result was available in the clinical IT system which was not checked prior to anti-D Ig administration. In the clinical area misinterpretation of results including misreading maternal blood group or 'not test' as a negative cffDNA screening result accounted for 4/28 clinical cases. Lack of interoperability between IT systems in the clinical and laboratory areas resulted in three transcription errors ($n = 2/28$ clinical, $n = 1/18$ laboratory) leading to two unnecessary anti-D Ig administrations and one omission of anti-D Ig. In three laboratory cases, the cffDNA screening results were available from the test provider but as were not entered/authorised on LIMS these were not available in the clinical IT system, leading to two unnecessary anti-D Ig administrations and one case of late administration.

Summary / Conclusions: IT can support best practice however, lack of interoperability between different IT systems requiring manual intervention can lead to transcription errors and delays. IT systems should support clear and standardised method of result display to avoid misinterpretation of results. Algorithms in LIMS should contain clear information that is not easily overridden. The SHOT data shows that IT has a potential to improve safety but only when it is configured and used correctly.

P692-A | Prevalence and specificity of clinically significant red cell alloantibodies in pregnant women at a tertiary-care facility

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Background: More than 50 red blood cell (RBC) alloantibodies are known to cause hemolytic disease of the fetus and newborn (HDFN). Although Rh immune globulin (RhIG) prophylaxis has significantly reduced the incidence of pregnancies complicated by anti-D, the need to detect and monitor maternal alloantibodies

capable of causing HDFN is still a concern. From our experience, there are few clinically significant RBC alloantibodies that require concern in pregnant women, but the migration of populations that we are currently seeing could change this reality in the near future.

Aims: The present study aims to assess the prevalence and specificity of red cell alloantibodies in pregnant women at a tertiary-care facility.

Methods: In this retrospective study, the prevalence and specificity of unexpected RBC alloantibodies known to cause HDFN in pregnant women at a tertiary-care facility during a 5-year period were compiled and analyzed. Patient selection was carried out by computerized search of patient data based on an obstetric location and the presence or history of RBC antibody between January 1, 2019, and December 31, 2023.

Results: A total of 691 obstetric patients with one or more RBC alloantibodies were identified during the 5-year period. The most common alloantibody found was anti-D in 645 (93.34%), followed by anti-M in 16 (2.32%) and anti-E in 9 (1.3%). The vast majority of anti-D identified had very low titers ($n = 615$, 89.5%), meaning previous anti-D prophylaxis with RhIG. The other antibodies identified were: anti-Lea in 6 pregnant women (0.87%); anti-Leb, anti-K, anti-Kpa and anti-G each one in 3 (0.43%); anti-C, anti-Fya and anti-Jka each one in 2 (0.3%); anti-N, anti-Cw, anti-c, anti-e and anti-Jkb each one in 1 (0.14%). Associations of different antibodies occurred in 16 (2.32%) of the studied population, the most common being anti-D + anti-M, in 4 (0.59%).

Summary / Conclusions: As expected, the most frequently identified alloantibody was anti-D, as a result of anti-D prophylaxis with RhIG. The other most common alloantibodies were anti-M and anti-E. Although we have identified 3 alloantibodies in immigrant women that have not been detected in natives in recent years - anti-c, anti-e and anti-N -, these results are not enough to say that there is a significant difference in the prevalence and specificity of RBC alloantibodies in pregnant women related to immigration. We're likely to see this change in the coming years, as immigration into this country is a growing trend. It will be necessary to analyze the years to come to see if there is a significant difference.

Clinical transfusion—neonatal and pediatric transfusion

P693 | A case of hemolytic disease of fetus and newborn caused by high-titer irregular antibody

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Background: Hemolytic Disease of Fetus and Newborn (HDFN) is caused by maternal antibodies and has been reported to cause fetal edema and anemia in severe cases, including death. The transfusion need and perinatal outcomes are related to the type of Ab, with RhD isoimmunization being the most frequent and serious during pregnancy.

Aims: HDFN caused by irregular antibodies is a rare, but possibly life-threatening condition. We report a rare case of HDFN due to high-titer irregular antibodies to Rh blood groups.

Methods: ABO/RhD grouping was performed using the ORTHO VISION platform. The irregular antibody identification was performed using a tube technique. Indirect and direct antiglobulin tests were performed by using the tube method. The titers were determined using a saline indirect antiglobulin test by preparing serial doubling dilutions of the plasma. The endpoint for the titer was 1+ agglutination.

Results: A woman in her 30s, at 33 weeks of gestation, was referred to Sapporo Medical University Hospital. She had experienced two parties and had given birth once. During the first visit to our hospital, maternal blood group typing was A, and RhD test results were negative. Irregular antibodies were detected in the blood, confirming the presence of anti-D and anti-C antibodies. Antibody titers were 65,536 for D, C, c, E, e phenotype and 32 for C, c, e phenotypes. Measurement of the middle cerebral artery peak systolic velocity and antibody titers were subsequently performed for periodic fetal monitoring, and antibody titers decreased from 65,536 to 16,384 and then to 8192 for D, C, c, E, e cells, while those for C, c, e cells increased from 32 to 64. Birth was induced at 37 weeks with vaginal delivery. The female newborn was typed as A- and D-positive and confirmed to have anti-D and anti-C antibodies. The titer of them were 1024 for D, C, c, E, e phenotype and 16 for C, c, e phenotype, respectively. The direct antiglobulin test result was positive for IgG specificity. After birth, the concentration of hemoglobin in infant blood was decreased to 8.6 g/dL and that of total bilirubin level was increased to 18.6 mg/dL, which led to transfusion of 1 unit of RBC on the third day after birth and blood exchange transfusion on the fourth day after birth. One month later, the anemia did not improve, and she received 1 unit of RBC.

Summary / Conclusions: The prevalence of anti-D-induced HDFN has decreased because of the common use of Rh immunoglobulin; however, serious fetal anemia usually occurs in pregnancies complicated by anti-D antibodies. The rate of hemolysis and disease severity were determined by the amount of antibody. When high titers of antibodies are identified in obstetrics, it is important to monitor the fetal course prior to birth and ensure appropriate management.

P694 | Cell salvage as a key part of intraoperative blood management in pediatric patients undergoing cardiac surgery—a single center experience

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Background: It is well known that cardiopulmonary bypass (CPB) is an important part of cardiac surgeries. In the pediatric population, the use of CPB requires large volumes of allogeneic blood transfusions, which are associated with postoperative morbidity and mortality. The development of Patient Blood Management programs has led to an increase in the use of autologous transfusions as they decrease transfusion reactions, transfusion-transmitted diseases, and immune-related complications. Pre-operative autologous donation and intraoperative cell salvage in children often had both technical and logistic limitations, making them not applicable to pediatric patients. Actually, advancements in new cell salvage devices allow efficient blood salvage in small children. In the case of cardiac surgery with CPB this technique allows not only to recover blood lost in the surgical field but also the residual bypass pump blood. This procedure concentrates the red cells, increasing the hematocrit before re-infusion

Aims: Describe the experience of cell saver blood transfusion in pediatric patients undergoing heart surgery and evaluate the impact of cell saver blood (CSB) transfusion on post-operative hemoglobin concentration, in our hospital from September 2020 to December 2023.

Methods: An observational, retrospective study was conducted on 182 patients with congenital heart disease who received CSB transfusion during or after cardiac surgery with CPB. Red blood cells (RBCs) concentrate was obtained by recovering blood lost in the surgical field, as well as from the CPB circuit after the procedure.

Results: 182 patients were enrolled in the study. The median age was 8 years old (range 1-19). The median weight was 25.5 Kg. Patients were classified according to the RACHS-1 score (Risk Adjustment for Congenital Heart Surgery). 65 (35.7%) children had a RACHS-1 score of 2, 87 (47.8%) patients had a score of 3, 10 (5.5%) patients of 4, 16 (8.7%) patients were heart transplant and 4 cannot be classified. The median time on pump was 111.50 min. The median volume priming was 900 ml and the median of the average hematocrit on CPB was 29%. We obtained 144 RBCs concentrates of which 136 were transfused and 8 were not used. The median volume of the products was 350 ml (100 – 1975), which represented 13.4 ml/Kg, with median of hematocrit 54 and 17.4 of hemoglobin. We made quality control by measuring free Hb as a direct hemolysis value. All values were within normal limits. Of the 136 transfused patients, 72 (52%) did not receive RBCs homologous during surgery. Their laboratory values post-surgery were: Median Hb: 12.1 mg%; median hematocrit: 35.5%; platelet count: 152500/mm³. Only 42 of the 136 received a RBCs homologous transfusion in the first 24 h post-surgery, not because of Hb value but for different reasons such as active bleeding. 56 patients did not receive any allogeneic RBCs component at all.

Summary / Conclusions: Despite efforts to reduce the use of blood components, allogeneic RBCs transfusions in children undergoing cardiac surgery with CPB cannot be avoided. However, we demonstrate that children who received CSB transfusion had hemoglobin values outside the transfusion range in the post-surgical control, which may induce a decrease in post-operative transfusions. CSB transfusion proved to be safe and effective as an adjuvant technique of

autologous transfusion in pediatric patients undergoing cardiac surgery with CPB.

P695 | Pediatric blood transfusions—unveiling adverse reactions in Colombia

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Background: Adverse transfusion reactions (ATR) represent undesired responses in patients occurring temporally with the administration of blood components. These reactions can significantly impact the quality and efficacy of transfusions, as well as the well-being of recipients. Identifying the causes behind these reactions is crucial for prevention. Reports from various countries indicate that rates of ATR are 1.3 to 2.6 times higher in pediatric populations compared to adults

Aims: To investigate whether similar trends are observed within the pediatric population in Colombia

Methods: This retrospective study conducted in Colombia from January 1, 2018, to December 31, 2022, examines transfusions and ATRs. Data were collected via the Hemovigilance Information System (SIHEVI-INS) managed by the National Institute of Health, incorporating voluntary reports from nationwide clinics and hospitals. ATRs were reported by medical personnel using standardized forms following guidelines set by the International Society of Blood Transfusion. A transfusion committee reviewed cases to ensure accuracy in classification of imputability and severity. Exclusions were made for missing data, procedural errors, or unlikely events. The study included 2,097,179 patients receiving 6,637,363 transfusions, with 6830 ATRs recorded after excluding 1293 cases. Demographic data were sourced from national statistics

P695 - Table 1. Summary information of blood transfusions and adverse reactions in Colombia between 2018 and 2022

Age (years)	ATR	Recipients	Transfused blood component	
			Red cells	Platelets
0 to 17	1561	196,687	224,514	137,670
≥18	5268	1,900,492	3,660,661	1254359

P695 - Table 2. Gender differences in ATRs and recipients by group age

Age (years)	Male ATR	Female ATR	Male Recipients	Female Recipients
0 to 1	130	71	65,348	51,936
2 to 14	608	448	45,516	42,119
15 to 65	1859	2184	423,461	528,012
>65	777	753	318,000	309,432

Results: The analysis reveals significant differences in ATRs between pediatric and adult patients. Pediatric recipients, though comprising only 9.4% of total recipients, accounted for 22.9% of total RATs (table 1). Most pediatric ATRs were non-severe, but severe cases were recorded, including fatalities. Adults, constituting the majority of recipients, also experienced ATRs, with a lower proportion of severe cases. Pediatric transfusions exhibited a male bias and a higher rate of ATRs per units transfused compared to adult transfusions, with a greater proportion of allergic reactions and fewer febrile non-hemolytic reactions. Pediatric recipients were more likely to experience ATRs across various blood components compared to adults (Table 2)

Summary / Conclusions: ATRs in pediatric population were three times higher than adult patients. These findings underscore the importance of understanding and addressing the factors contributing to ATRs in pediatric populations, to improve the safety and efficacy of transfusion practices

P696 | Five-year review of laboratory insights in hemolytic disease of fetus and newborn

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Background: Although ABO incompatibility is the most common cause of hemolytic disease of the fetus and newborn (HDFN), the symptoms are generally mild, without severe anemia or requirement for phototherapy.

Aims: In this study, we investigated all cases of HDFN of our institute and aim to discuss clinical characteristics and considerations during transfusions and laboratory testings.

Methods: We reviewed the medical records of newborns regarding HDFN due to ABO incompatibility for the past 5 years of our institution. We defined newborns with ABO typing discrepancy and incompatibility in crossmatching with mother's blood group as HDFN. The laboratory results such as ABO blood type of mothers and newborns, direct antiglobulin test (DAT), total bilirubin were collected. History of transfusion and phototherapy were also taken.

Results: During the five-year period from 2019 to 2023, a total of 275 newborns were diagnosed with HDFN by ABO blood type testing and crossmatching. As for the mother's blood type, group O was the predominant with 259 cases, followed by B and A type, 11 and 5 cases respectively. For the newborns', group A was the most common with 151 patients, followed by group B with 108 and AB with 16 cases. The most common type of incompatibility of mother/newborn was O/A, accounting for 54.9%, followed by O/B, B/AB, and A/AB at 54.9%, 4.0%, and 1.8%, respectively. The DAT test was conducted on only half of the group O mothers,

and among them, 38.5% had trace or positive results. For the 16 cases of non-O group mothers, only 6 underwent the DAT test, and all of them was negative. 21.6% of newborns from group O mothers received transfusions, and 31.3% of newborns from non-O blood type mothers received transfusions. Phototherapy was administered to newborns of O blood type mothers at 49.4%, and to newborns of non-O blood type mothers at 43.8%.

Summary / Conclusions: The blood bank laboratory should consider the possibility of HDFN in newborns when conducting pre-transfusion tests such as blood typing and select blood types for transfusions, especially in cases involving Group O mothers. Unless the institution provides O-RBC regardless of the newborn's blood type, ABO back typing for newborns can contribute to safe blood transfusion practice.

P697 | Abstract withdrawn

P698 | Analysis of the effectiveness of targeted platelet transfusion in neonatal alloimmune thrombopenia—a report of six cases

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Background: Neonatal alloimmune thrombocytopenia (NAIT) is defined as an uncommon platelet disorder caused by maternal alloimmunization to human-specific antigens (HPAs) that are paternally inherited, resulting in low fetal/neonatal platelet levels and debilitating effects on the newborn. The most common antibodies involved are Anti-HPA. The role of Anti-HLA antibodies is a subject of controversy although their involvement in platelet transfusion refractoriness is well established. NAIT is the cause of the majority of cases of intracranial hemorrhage caused by thrombocytopenia, with an incidence between 10% and 30%. For this reason it is important to determine the presence of Anti-HPA and/or Anti-HLA antibodies to perform targeted transfusion therapy in order to reduce hemorrhagic complications.

Aims: -Describe the case series: minimum platelet level, hemorrhagic events, treatment received and response to treatment. Study the effectiveness of targeted platelet transfusion in newborns with neonatal alloimmune thrombocytopenia.

Methods: We have analyzed six confirmed cases of NAIT at Cruces University Hospital, Spain. Antibodies were detected using a multiplex bead-based assay for detecting human platelet antibodies (Pak-Lx) and genotyping of parents and newborns was performed to confirm HLA/HPA antigenic discrepancies. Platelet yields at 24 hours were analyzed using the Calculation of Corrected count Increment (CCI) formula for both non-directed and directed platelet transfusions. Data were analyzed using the Kolmogorov-Smirnov test.

Results: The antibodies identified in maternal serum were Anti HPA-1b ($n = 1$) Anti HPA-5b ($n = 2$) Anti-HLA ($n = 3$). In two cases, there were discrepancies in the HPA-15a/b genotypes without antibodies being identified. **Table 1** describes the characteristics of patients included in the study; platelets at birth, minimum platelet value, hemorrhagic events, and maternal and neonatal medical history. Four targeted platelet transfusions were performed. In one case, platelet yield could not be analyzed because blood analysis was not performed at 24 h. The mean platelet yield for non-directed transfusions ($n = 8$) was 9.92 (-4 min, 37.47max). For targeted platelet transfusion the mean yield was 39.86 (23.6 min, 54.53). Although there was an increase in the transfusion yield for targeted platelets, no statistically significant differences were obtained due to the number of samples.

Summary / Conclusions: NAIT is the most frequent cause of thrombopenia in the term newborn and its identification as well as perform targeted platelet transfusion is a safe and important strategy to prevent hemorrhagic complications.

P699 | Abstract withdrawn

P700 | Abstract withdrawn

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P702 | Abstract withdrawn

P698 - Table 1.

Lower platelet count ($\times 10^3/\mu\text{L}$)	Maternal antibodies	Maternal genotype	Newborn genotype	Targeted platelet transfusions
13	Anti-HPA-5b	HPA 5a/5a	HPA5a/5b	1
44	Anti-HPA-5b Anti-HLA	HPA 5a/5a	HPA 5a/5b	0
26	Anti-HLA No Anti-HPAs found	HPA 15b/15b	HPA 15a/15b	0
7	Anti-HLA No Anti-HPAs found	HPA 15b/15b HPA 3a/3a HPA 1a/1a	HPA 15a/15b HPA 3a/3b HPA 1a/1b	1
11	Anti-HLA	No HPA discrepancies	No HPA discrepancies	1* Not appreciable
5	Anti-HPA-1b	HPA 1a/1a	HPA 1a/1b	2

Clinical transfusion—therapeutic apheresis

P703 | Implementation of an automated red blood cell exchange program in a tertiary referral hospital—benefits and challenges over seven years

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Background: Automated red blood cell exchange (RBCX) is a valuable treatment option for acute and chronic complications of sickle cell disease (SCD). It involves replacing the patient's red blood cells (RBCs) with health donor blood to significantly reduce hemoglobin S concentration without exposing the patient to an increased risk of iron overload.

Aims: We aimed to evaluate the characteristics and outcomes of patients with SCD enrolled in a RBCX program at a tertiary referral hospital.

Methods: Automated RBCX performed with Spectra Optia (TerumoBCT®) was introduced in January 2017 for acute events, and in February 2018 for chronic indications. Single needle procedures have been available since April 2019 (software 12.0 TerumoBCT®). The device calculation post-apheresis targets were hematocrit around 30% and Hb S below 25%–30%. Extended phenotype RBC matching was selected to minimize alloimmunisation. A retrospective analysis was performed of a consecutive series of patients who underwent RBCX, from January 2017 to December 2023, regardless of age. We evaluated patient and procedural characteristics, efficacy and safety issues of RBCX and clinical outcomes.

Results: A total of 36 patients, including 26 pediatric cases, and 300 procedures were included in the analysis. The mean age was 9 years (range:1-16) in the pediatric group and 32 years (range:18-62) in the adult population. RBCX was performed in an acute setting in 27 subjects (20 of whom were pediatric) and 9 patients (6 of whom were children) were enrolled in a chronic RBCX program. When RBCX was prescribed in an acute setting, the main indications were acute chest syndrome and pre-surgery in 66% of cases. Regarding venous access, the majority of procedures (85%) were carried out through a central venous catheter. In the chronic RBCX program, the main indication for admission was recurrent vaso-occlusive crisis (66%). While a port (Vortex, AngioDynamics®) was used in all pediatric patients, ultrasound-guided peripheral venous access was available in all adult patients. Genotype or extended phenotype was available in all patients. An erythrocyte alloantibody had been previously detected in 4 cases and was newly developed during the RBCX treatments in 3 patients (anti-Lea, anti-Kpa and anti-C). Regarding the performance of the procedures, 186 (62%) were single-needle

modality and in 47 (15%) required blood priming. In the acute RBCX subgroup, the Hb S target was achieved in 91% of the treatments. Minor venous access flow problems were identified in 4 cases (11%). All patients showed clinical improvement post-RBCX treatment. When analyzing chronic RBCX procedures, in 96% of cases the post-apheresis Hb S target was reached. The mean time duration in the single-needle option was longer (120 min) compared to the dual-needle modality (89 min). Overcoming flow difficulties were observed in 31 procedures (12%). In 3 cases apheresis failed due to unresolved flow problems. At follow-up, all patients showed a decrease or even disappearance (33%) of vaso-occlusive crises. Safety concerns included one mild allergic reaction, one possible hypocalcemia and one port hematoma.

Summary / Conclusions: In our series, automated RBCX is a feasible, safe and effective modality for the management of SCD patients in both acute and chronic settings. Chronic RBCX program has shown clinical benefit. Therefore, it would be interesting to conduct a prospective study to evaluate the factors associated with clinical response and to assess the impact on patient quality of life.

P704 | Efficacy of high volume versus standard volume plasma exchange in patients with alcohol related Acute on Chronic Liver Failure (ACLF)

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Background: Acute on chronic liver failure (ACLF) is a clinical syndrome with a rapid dynamic course and a high in-hospital mortality. While high volume therapeutic plasma exchange (HV-TPE) is a well-established treatment modality in patients with acute liver failure, its efficacy in treating ACLF patients as well as its comparison to standard volume TPE (SV-TPE) is yet to be established. ACLF carries a high in-hospital short-term mortality (more than 40%) which varies based on the initial severity and the number of organ failures.

Aims: To compare the efficacy of HV TPE with SV TPE in alcohol related ACLF patients.

Methods: Twenty consecutive patients with alcohol related APASL-ACLF Grade 1 & 2 were enrolled into the study (10 in HV-TPE arm and 10 in SV TPE arm). Patients in both the arms underwent TPE using a continuous flow centrifugation plasma exchange device (Optia Spectra Terumo® BCT, Lakewood, Colorado, USA) in addition to standard medical treatment (SMT). In HV TPE arm, 15% of ideal body weight or 8 to 12 litres was exchanged in a single session. In SV TPE arm, one to 1.5 times plasma volume of the patient was removed in a single session at a PE rate of 40ml/minute. In both the arms, plasma exchanged was replaced with an equal volume of replacement fluid (group specific Fresh Frozen Plasma according to the calculated plasma volume to be replaced). A total of 5 procedures were

performed on alternate days. The procedure was performed via a peripheral vascular access using a 16-gauge needle for blood withdrawal (inlet) and return. In patients with inadequate peripheral venous access, central venous catheters (Jugular / Femoral, preferably jugular) were assessed to ensure uninterrupted inlet flow. The change (baseline to post TPE) in laboratory parameters, clinical severity scores along with 30 and 90 day mortality rates were noted and compared between the two groups. The adverse events were also noted in both the groups.

Results: A total of 44 and 52 TPE procedures were performed in HV and SV arm respectively (average of 4.4 and 5.2 procedures/patient respectively). Both HV and SV TPE were effective in significant reduction of serum bilirubin ($p = 0.0023$ vs 0.0000) ALP ($p = 0.0069$ vs 0.0117), INR ($p = 0.0050$ vs 0.0051), serum ammonia ($p = 0.0483$ vs 0.0197) and clinical severity scores-AARC ($p = 0.0001$ vs 0.0002), MDF ($p = 0.0008$ vs 0.0017) and MELD ($p = 0.0050$ vs 0.0095). However, HV TPE was found to be more effective in correction of all severity scores however the differential impact was significant only for MELD and CTP ($p < 0.05$). Mortality at 30 day (30% vs 30%, $p = 1.00$) and 90 days (40% vs 60%, $p = 0.40$) were not significantly different in either group. Procedure related adverse events was observed in 6.8% of HV TPE and 5.8% of SV TPE procedures.

Summary / Conclusions: HV TPE is more effective in improving clinical severity scores like MELD and CTP. However, difference in impact on mortality could not be established in our study.

P705 | Abstract withdrawn

P706 | Shedding new light on GVHD—a 17-year journey through extracorporeal photopheresis efficacy and survival insights

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Background: Extracorporeal photopheresis (ECP) is a leukapheresis-based procedure used to treat both acute and chronic graft-versus-host disease (aGVHD and cGVHD), among other conditions. Owing to its significant effectiveness and outstanding safety profile, ECP has established itself as a primary treatment option for steroid-refractory GVHD. This study examines our 17-year research experience with ECP, including some under-reported complications.

Aims: The primary aim is to determine the response rate, including the degree of improvement defined as the proportion of patients achieving an overall response (OR) at day +28. Secondary aims include assessing demographic variables, type of GVHD, underlying diagnosis, treatment-related adverse events, and overall survival.

Methods: This retrospective, observational study included adult and pediatric patients older than 4 years who received extracorporeal

photopheresis from January 2006 to February 2023. The most common indication for this therapy was graft-versus-host disease, more specifically, glucocorticoid (GC)-resistant GVHD, defined as a disease that progresses by day 5 or shows no response to treatment by day 7. The only exclusion criterion was hypersensitivity or allergy to psoralen.

Results: We analyzed 24 patients and 572 ECP procedures, comparing 14 responders to 10 non-responders. Responders were younger, with a mean age of 30 (range 13-51), compared to non-responders, who had a mean age of 42 years (range 22-63, p -value 0.046), with no significant difference in gender distribution (female 35.71% vs. 40%, $p = 0.839$). The response rate, defined as a complete or partial response after 4 weeks, was 58%. Responders received more GVHD therapies (mean 6) compared to non-responders (mean 4, $p = 0.011$) and had a longer ECP course with a mean of 31 sessions (range 8-97) versus 14 sessions (range 1-37, $p = 0.041$). ECP complications were similar, but non-responders experienced more anemia (17.27% vs. 0.23%, $p = 0.041$). The mean overall survival for responders was significantly higher at 76.7 months compared to 29.81 months for non-responders ($p = 0.009$, HR: 0.43).

Summary / Conclusions: In summary, our 17-year retrospective study reinforces ECP as an effective treatment for steroid-resistant graft-versus-host disease, demonstrating notable improvement in patient outcomes, especially among younger individuals with a longer course of treatment. The findings indicate a significant survival advantage for responders to ECP treatment, despite some underreported complications like anemia in non-responders.

P707 | Amotosalen-UVA Pathogen Reduced Plasma, Cryoprecipitate Reduced (PRPCR)—an optimized cost-effective component for therapeutic plasma exchange

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Background: Cryoprecipitate poor plasma (CPP) is underutilized for therapeutic plasma exchange (TPE). Mafrá (American Society of Hematology Abstract # 2629, 2024) reported a meta-analysis of TPE for thrombotic thrombocytopenic purpura (TTP) showing decreased mortality with CPP compared to plasma. Solvent detergent plasma (SDP) is used for TTP and albumin for other TPE to reduce risk of transfusion-transmitted infection (TTI). FDA approved amotosalen-UVA Pathogen Reduced Plasma, Cryoprecipitate Reduced (PRPCR: Cerus, Concord, CA) for TPE of TTP and other indications to reduce risk of TTI.

Aims: To characterize the hemostatic functions of PRPCR.

Methods: Thrombin Generation (expressed as endogenous thrombin potential- ETP), Fibrinogen (FIB), Factors II, V, VII, VIII, IX, X, XI were

P707 - Table 1.

	PRPCR	PR Plasma
Thrombin Generation-ETP (nM · min)	1156 ± 208	1581 ± 154
Fibrinogen (mg/dL)	1.47 ± 0.15	2.28 ± 0.49
Factor II (IU/mL)	0.77 ± 0.07	0.89 ± 0.12
Factor V (IU/mL)	0.66 ± 0.12	0.86 ± 0.17
Factor VII (IU/mL)	0.83 ± 0.21	0.77 ± 0.23
Factor VIII (IU/mL)	0.15 ± 0.05	0.92 ± 0.35
Factor IX (IU/mL)	1.00 ± 0.19	1.00 ± 0.25
Factor X (IU/mL)	0.86 ± 0.13	0.94 ± 0.19
Factor XI (IU/mL)	1.02 ± 0.29	0.92 ± 0.21

P707 - Table 2.

	PRPCR	PR Plasma
vWF RIPA (IU/mL)	0.10 ± 0.00	0.95 ± 0.38
ADAMTS 13 (IU/mL)	1.32 ± 1.8	0.90 ± 1.6
Protein C (IU/mL)	0.99 ± 0.14	0.79 ± 0.18
Protein S (IU/mL)	0.83 ± 0.10	0.97 ± 0.23
α -2 plasmin inhibitor (IU/mL)	0.86 ± 0.13	0.76 ± 0.07
IgG (mg/g total protein)	989.9	993.5
IgA (mg/g total protein)	13.7	14.2

measured by one-stage coagulation assays. VWF was measured by Ristocetin platelet aggregation (RIPA). ADAMTS13 was measured by FRET assay. Protein C, Protein S, and α -2 plasmin inhibitor (α -2 PI) were measured by standardized assays. IgG and IgA were assessed by nephelometry and SDS PAGE. The ability of FIB and vWF from PRPCR to support platelet adhesion and aggregation were determined in microfluidic chambers at various wall shear rates. For these experiments, we used hirudinized whole blood or reconstituted plasma-free blood (RBC + platelets + PRPCR or PR cryoprecipitated fibrinogen complex, IFC).

Results: (Tables 1-2). In PRPCR: FIB, Factor VIII, and vWF were reduced compared to plasma. Factors II, V, VII, IX, X, XI, Protein C, Protein S, and α -2 PI were conserved. IgG levels were retained without qualitative changes. Thrombin generation was reduced with retained activity (Table 1). Microfluidic assays at low shear (300 s^{-1}) confirmed PRPCR (FIB = 50 mg/dL) surface coating promoted platelet adhesion, which was reduced compared to IFC (FIB = 300 mg/dL). Integrin α IIb β 3 played a key role in adhesion with complete inhibition by abciximab for both conditions, indicating the GPIIb-IX-V complex is only partly involved. Perfusion of reconstituted blood over immobilized vWF binding peptide (100 μ g/mL), showed absence of platelet adhesion with PRPCR compared to IFC full activity. Perfusion of reconstituted blood over immobilized collagen (200 μ g/mL) at high wall shear rate (3000 s^{-1}) resulted in no platelet aggregation with PRPCR compared to IFC.

Summary / Conclusions: PRPCR retained thrombin generation, anti-thrombotic proteins, and ADAMTS13. Residual fibrinogen

supported platelet adhesion at low shear. Collagen induced platelet aggregation was negligible at high shear due to depletion of high molecular weight vWF. PRPCR retains functional hemostatic capacity for TPE without increased platelet thrombotic activity while providing retention of IgG and IgA, and the benefit of pathogen reduction.

P708 | Therapeutic apheresis in neurological disease—a retrospective study

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Background: Therapeutic plasma exchange (TPE) is a treatment for acute attacks of neurological disease aimed at removing pathogenic components from the plasma. TPE can be used alone or in combination with other therapies. Recent advances suggest that TPE is as equally effective as immunoglobulins, to be used as a first-line treatment. However, many hospitals have not adopted it yet.

Aims: To analyze the response rate and safety profile of TPE in immunoneurological diseases.

Methods: We have a retrospective observational study from patients with acute attacks of immunoneurological diseases treated with TPE in a tertiary-care hospital between May 2016 and January 2024. The main target was to analyze the clinical response. Non-response was defined as the absence of improvement or progression of symptoms after 7 sessions of TPE. Patients' records were reviewed with respect to demographics, diagnosis, treatment history, TPE sessions, clinical responses, IVIG support, and adverse events. TPE sessions typically ranged from 2 to 6, with additional sessions based on medical criteria. We used a continuous-flow centrifugation system. Each session was carried out 1-1.5 times at the predicted plasma volume with a frequency of either daily or every 48 hours. The substitution solution used was albumin. Fresh frozen plasma was only administered to one patient due to a hemostatic disease not secondary of TPE.

Results: Retrospective study from 57 patient treatment with TPE were analyzed. These patients had a median age of 59(16-87) years and were predominantly women ($n = 30$ [52.6%]). The incidence of neurological disease is represented below.

P708 - Table 1.

Frequencies of DIAGNOSIS	Counts	% of Total
SD.Guillain Barré	20	35.1%
Myasthenia Gravis	12	21.1%
SD.Miller Fisher	8	14.0%
Acute disseminated Encephalomyelitis	8	14.0%
Chronic inflammatory polyneuropathies	8	14.0%
Multiple sclerosis	1	1.8%

A total of 387 TPE sessions were performed. TPE was conducted as first-line in 52.6% ($n = 30$) of cases, and others after IVIG or immunosuppressive therapy. The median processed plasma volume was 2500 mL for each cycle. 84.2% ($n = 48$) of patients responded favorably to TPE, resolving acute episodes. Patients who received IVIG ($n = 25$) showed response in 76% ($n = 19$) and non-response in 24% of cases ($n = 6$), with no statistically significant differences ($p = 0.16$). The median number of TPE sessions per patient was 6, with no statistically significant differences ($p = 0.37$) between use as first-line and others. Similarly, no difference was observed in the IVIG group ($p = 0.30$). The incidence of acute relapse was low 9.3% ($n = 5$), no statistically significant differences ($p = 1$) observed in the IVIG group. There was no differences in relapse between patients treated with TPE as first-line and others. The diagnoses in relapse were Myasthenia gravis ($n = 4$) and Miller Fisher syndrome ($n = 1$). The median time of relapse was 360 days. No adverse events were associated during TPE.

Summary / Conclusions: TPE has a relevant role in the management of acute immunoneurological diseases and it is recommended as first-line therapy with a grade 1A or 1B recommendation in "ASFA 2023". This study demonstrates that TPE is effective with responses to 84% of patients. Associating IVIG to TPE has not demonstrated an improvement in the rate response. Our data suggests that the TPE treatment is safe and equally effective as the IVIG but for deeper results more research is needed.

P709 | Therapeutic plasma exchange in patients with neurological diseases—a four-year Albanian experience

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Background: In Albania we have been using the Therapeutic Plasma Exchange procedure since 2012 mainly for neurological diseases. In our country plasma exchange, since its introduction remains the first line of treatment for autoimmune neurologic disorders.

Aims: Our aim was to evaluate our results in the treatment of autoimmune neurologic disorders with TPE as well as the risk benefit ratio in our treated patients.

Methods: We retrospectively reviewed the medical records of patients with neuroimmunologic disorders that were treated with TPE in our centre during 2020-2023. In total there were 107 patients (52 male, 55 female). 66 of them were diagnosed with Guillain-Barré syndrome (GBS), 20 with myasthenia gravis (MG), 5 with chronic inflammatory demyelinating polyneuropathy (CIDP), 3 children with autoimmune encephalitis, 6 with neuromyelitis optica, 6 with multiple sclerosis (SM), 1 with anti-IgLON5.

Results: Total number of procedures performed for 107 patients was 500, with a median number of TPE sessions per patient 4 [1-14]. TPE was done through central venous catheters in all cases. The median age was 50 (range: 5-85) years. We found that 95.5% of patients had a noticeable improvement of symptoms and only 2 patients relapsed

and returned to our center with neurological symptoms undergoing additional sessions. Patients diagnosed with autoimmune encephalitis that received TPE procedure were totally recovered. There were only three cases of mortality due to respiratory failure, all of them GBS post covid. 51 patients experienced only mild side effects. These side effects were, complications related to catheter insertion (7 patients), hypotension (39 patients), hypocalcaemia (2 patients) and bradycardia (16 patients). Only 11 of them experienced both hypotension and bradycardia, and 2 patients experienced hypotension, bradycardia and hypocalcemia.

Summary / Conclusions: The improvement rates were very encouraging and side effects were mainly mild without impairing general conditions of the patients. Benefits outweigh the risks suggesting that TPE is a safe and an effective treatment option in autoimmune neurological disorders.

P710 | Abstract withdrawn

P711 | The outcome of patients undergoing therapeutic plasma exchange for pulmonary haemorrhage associated with leptospirosis in National Hospital and Colombo South Teaching Hospital of Sri Lanka

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Background: Leptospirosis is a widespread and emerging zoonotic disease that mostly affects communities living in resource-poor settings. It has a global incidence of 1.03 million cases and 58,900 deaths per annum, with the highest disease morbidity and mortality in South and Southeast Asia. Sri Lanka has the highest incidence of leptospirosis worldwide, with an estimated pooled case fatality ratio of 7%. Lung involvement in leptospirosis occurs in 20–70% of patients and accounts for a mortality rate of more than 50% in those diagnosed with the severe disease. Despite high morbidity and mortality, no definitive treatment is available for pulmonary hemorrhage due to leptospirosis, at present. Several studies and case reports have demonstrated that therapeutic plasma exchange (TPE) can be used as a treatment modality for patients with pulmonary haemorrhage in leptospirosis. This study aims to describe the outcome of patients who underwent TPE for pulmonary hemorrhage associated with leptospirosis.

Aims: The aim of the study was to determine the outcome of patients undergoing therapeutic plasma exchange for leptospirosis associated with pulmonary haemorrhage with regard to mortality, hospital stay and long-term consequences and disability from the disease.

Methods: This descriptive study was conducted in two major hospitals in the Western Province of Sri Lanka from January 2021 to June 2022, including all patients referred for therapeutic plasma exchange. Hospital records of relevant patients were traced, and data were collected.

Results: The study included 86 leptospirosis-confirmed positive cases, with 78 (90.7%) males and 8 (9.3%) females, with a mean age of 48.8 (SD 15.2). The mean duration of fever on admission was 4.08 days, with pulmonary hemorrhage diagnosed between three and nine days. The mortality rate among the patients who underwent TPE was 25.58% ($n = 22$). The duration of hospital stay varied from 4 to 28 days, with a mean duration of 10.77 days. Among the 64 patients who survived following treatment with plasma exchange, 24 had short-term and long-term complications due to leptospirosis. From the 86 patients who underwent TPE, all had acute kidney injury, 52 (60.5%) had acute liver injury, 36 (41.9%) had myocarditis, and coagulopathy was observed in 27 (31.4%) patients while three patients had rhabdomyolysis, and two had pancreatitis. The study also revealed a significant association between smoking and severe pulmonary hemorrhage syndrome ($p < 0.001$), whose severity was assessed using pre-TPE oxygen demand.

Summary / Conclusions: The study revealed a significant improvement in survival from the disease, with treatment of plasma exchange warranting further studies for evidence-based inclusion of it as a treatment modality for leptospiral pulmonary hemorrhage.

P712 | Sensitization of therapeutic plasmapheresis patient to ethylene oxide gas

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Background: Ethylene oxide (EtO) is a low temperature gaseous process widely used in hospitals and the medical equipment industry, including apheresis devices. Since 1975, EtO-associated allergic reactions during hemodialysis have been reported, but not for the therapeutic plasmapheresis (TPE) patients.

Aims: We now report a case of EtO-associated allergic reaction in TPE patient.

Methods: A 62-year-old male patient with end-stage kidney disease was scheduled for a kidney transplant. In preparation for the surgery, he was set to undergo TPE on an every-other-day basis. His treatment utilized a Spectra Optia Apheresis System (Terumo BCT) along with a specific kit. The total plasma volume to be processed was estimated at approximately 3604 mL, based on the patient's weight of 90.1 kg and an alternative plasma volume to 40 ml/kg. The procedure was initiated with an inlet velocity of 37 mL/min. Anticoagulation was managed with ACD-1, and 14 units of human serum albumin (HSA) were used as the replacement fluid. The patient's vital signs were stable.

Results: Approximately 40 min after initiating a routine apheresis treatment, the patient exhibited signs of an allergic reaction, developing urticaria and swollen eyes. Initially, he was administered 4 mg of chlorpheniramine; however, his condition worsened, with severe

itching and a drop in SpO₂ levels to 90%–91%. A second dose of 4 mg chlorpheniramine was given, and the apheresis machine's inlet velocity was reduced to 10 ml/min in an attempt to mitigate the reaction. Once the symptoms began to subside, the inlet velocity was cautiously increased to 20 ml/min. Despite these measures, the patient's itching intensified once more, and his SpO₂ decreased to 90%. At this point, 100 mg of hydrocortisone was administered, and oxygen was supplied at a rate of 2 L/min via a nasal cannula to address the respiratory distress. Following these interventions, the patient's condition stabilized to a tolerable level, allowing for the completion of the TPE session. For the second TPE session, a desensitization protocol for HAS was implemented due to previous adverse reactions. This protocol involved gradually increasing the concentration of HAS, starting at 20 mg/ml, from an initial rate of 0.2 ml/hr up to 30 ml/hr, with increments every 15 minutes. After successfully administering HAS at 30 ml/hr without any adverse effects, and priming the TPE kit with normal saline, the TPE procedure commenced with an initial inlet velocity of 30 ml/min. This session was completed as planned, without any complications. In the third TPE session, the process was initiated only with the priming of the kit using normal saline, omitting the desensitization step for HAS, and maintaining the initial inlet velocity at 30 ml/min. This session also proceeded without any adverse effects and was completed successfully. The successful completion of the fourth session without desensitization, coupled with the knowledge that the desensitization of HAS remains effective for approximately 24 hours, indicates that the cause of the adverse reactions in previous sessions was not HAS sensitivity.

Summary / Conclusions: This a case of adverse effect of EtO, but there were few reports about TPE. This case underscores a critical consideration in the context of TPE treatments: replacement fluids used during the procedure, such as FFP or HAS and materials and substances involved in the sterilization of medical devices, such as EtO, as potential sources of patient adverse reactions.

P713 | Efficacy of therapeutic apheresis procedures in a tertiary hospital—alignment with scientific guidelines

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Background: The ninth edition of the ASFA guidelines, published in the Journal of Clinical Apheresis (JCA) in 2023, reassesses the indications for therapeutic apheresis in some cases compared to the previous from 2019. Clinical responses to these procedures should correlate with the level of indication specified in the newly guidelines.

Aims: To evaluate the appropriateness of therapeutic apheresis indications according to the ninth edition of the ASFA guidelines and assess the concordance of procedure efficacy in a tertiary-level hospital over a three-year period.

P713 - Table 1

Pathologies (n = 22)	Patients (n = 82)	Type of Procedure	Procedures (n = 262)
Sickle cell disease	15 (18.29%)	RBC exchange	39 (14.88%)
Myasthenia gravis	9 (10.97%)	TPE	35 (13.36%)
ABO incompatibility	8 (9.75%)	TPE	10 (3.82%)
GBS	8 (9.75%)	TPE	19 (7.25%)
TTP	6 (7.32%)	TPE	31 (11.83%)
PV	5 (6.10%)	Erythroapheresis	8 (3.05%)
Optic neuritis	5 (6.10%)	TPE	30 (11.45%)
ME	4 (4.88%)	TPE	14 (5.34%)
NMDA encephalitis	4 (4.88%)	TPE	20 (7.63%)
Hyperleukocytosis	3 (3.66%)	Leukocytapheresis	5 (1.90%)
MG hyperviscosity	3 (3.66%)	TPE	14 (5.34%)
Secondary polyglobulia	2 (2.44%)	Erythroapheresis	2 (0.76%)
Hypertriglyceridemia	2 (2.44%)	TPE	3 (1.15%)
AIHA	2 (2.44%)	TPE	8 (3.05%)
Hemochromatosis	1 (1.22%)	Erythroapheresis	3 (1.15%)
Rare indications	5 (6.10%)	TPE	24 (8.02%)

P713 - Table 2.

Indication (n = 82 patients)	Improvement	No improvement
I (n = 36)	28 (77.78%)	8 (22.23%)
II (n = 30)	26 (86.67%)	4 (13.34%)
III (n = 15)	10 (66.67%)	5 (33.34%)

Methods: A retrospective study spanning three years (2021-2023) assessed the indication and recommendation grade based on the 2023 ASFA guidelines comparing it with the previous 2019 edition. Additionally, the efficacy of these procedures was evaluated using validated clinical and analytical parameters. Therapeutic apheresis procedures were performed using the Spectra Optia® system.

Results: During the study period, 262 therapeutic apheresis procedures, were conducted on 82 patients, as shown in Table 1; 76.82% using central access and 23.17% using peripheral access. For patients undergoing TPE (69.51% of total), the mean number of procedures was 3.54 per patient. According to the 2023 ASFA guidelines, the described procedures fell into categories I (43.90%), II (36.59%), and III (18.29%). Remarkably, 97.56% had a level of indication that overlapped between the 2019 and 2023 guidelines. The remaining 2.44% that did not agree with the previous guidelines included a plasma exchange in 2021 for autoimmune necrotizing myositis, performed under grade IV indication that is currently a grade III; and three leukaphereses for hyperleukocytosis in 2022, performed under grade II indication that is currently a grade III according to the 2023 guidelines.

Table 2 describes the relationship between the indication categories according to the 2023 ASFA guidelines and the improvement or stability of the patients.

Summary / Conclusions: Most of the therapeutic apheresis procedures realized in our unit are indicated for hematological and neurological pathologies. The appropriateness of these procedures, based on the more recent guidelines, was 97.56%, belonging the majority to categories I and II, showing a higher percentage of favorable clinical outcomes compared to indication grade III. We observe a very good correlation between the recommendations in these guidelines and our clinical practice, allowing us to identify specific cases where the indications have been changed compared to the previous ASFA edition. Efforts to adequate the indications to scientific evidence guarantee effective and safe results for patients.

P714 | Use of therapeutic plasma exchange in paediatric disease conditions in a national paediatric tertiary care center in Sri Lanka

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Background: Therapeutic Plasma Exchange (TPE) is increasingly used as a treatment modality for various Paediatric clinical conditions. TPE removes pathogenic substances such as autoantibodies, circulating immune complexes, lipoproteins from plasma and is being used as a standard therapeutic modality in a variety of diseases. TPE procedures have being done for emerging clinical conditions such as cytokine storm in COVID 19, acute fatty liver with hepatic encephalopathy and following various infections.

Aims: Our aim was to describe paediatric disease conditions for which TPE was used in a National Paediatric Tertiary Care Center in Sri Lanka.

Methods: All patients who underwent TPE at National Paediatric Tertiary Care Center from June 2021 to November 2023 were included in this retrospective study. Data was collected from the registries available at the Transfusion Medicine Unit and bed head tickets of patients. Data was entered into an Excel database and was analyzed using SPSS statistical software.

Results: 212 patients underwent TPE during the study period. There was equal gender distribution and nearly two thirds were below 10 years of age. 119 patients with body weight <25 kg underwent blood priming prior to TPE. Highest number of patients who underwent TPE had neurological disorders (65%). Among other diseases 8% had haematological conditions, another 8% infections and 5.6% were renal diseases. Of the neurological disorders autoimmune encephalitis accounted for the highest number (45.6%) and 38.4% presented with Guillen Barre Syndrome. Optic neuritis was 6% and Transverse Myelitis was 5%. Among haematological diseases Thrombotic Thrombocytopenic Purpura was the commonest (58.82%). Haemolytic Uraemic Syndrome and Haemophagocytic Lymphohistiocytosis accounted for 35.29% and 5.88% respectively.

Summary / Conclusions: TPE appeared to be beneficial in most of the paediatric neurological disorders. More research is required to conclude its usefulness in paediatric clinical practice.

P715 | Therapeutic plasma exchange for heparin induced thrombocytopenia during treatment of anti-glomerular basement membrane disease

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Background: Heparin induced thrombocytopenia (HIT) usually appears following exposure to unfractionated heparin (UNH). It is a disorder characterized by platelet-activating anti-heparin/platelet 4 immune complexes (HITAbs), which cause thrombocytopenia and increase thrombosis risk. Although the use of therapeutic plasma exchange (TPE) to treat HIT is not widespread, some studies support its role in the treatment of severe or refractory HIT, as well as in patients with persistent HITAbs who need urgent cardiothoracic surgery with re-exposure to UNH. Given the paucity of evidence (ASFA 2023 category III), there is no consensus on the number of procedures needed nor the replacement fluid of choice. TPE partially removes pathogenic ultra-large immunoglobulin G (IgG) HITAbs, thus the number of procedures required could be fewer than the 5-6 procedures needed for smaller antibodies.

Aims: To highlight the role of TPE in the management of HIT.

Methods: Retrospective case report of a patient who developed HIT during TPE.

Results: An 85-year-old woman presented with diffuse alveolar hemorrhage and rapidly progressive renal failure, and was diagnosed of anti-glomerular basement membrane disease. Treatment with steroids, cyclophosphamide and TPE was started. TPE was performed on alternate days processing an average of 1.2 plasma volumes, using saline/albumin solution and fresh frozen plasma (FFP) as replacement fluid and anticoagulation with UNH. Five days from the start of TPE,

platelet count decreased from 327,000/ μ L to 55,000/ μ L. The 4T score was intermediate (5 points). The HITAbs immunoassay was positive (12.3 U/mL, cutoff: 1/ml), supporting the diagnosis of HIT. Thrombotic complications were ruled out. Heparin was immediately discontinued and 7 additional TPE processes were performed with the same technical characteristics but using ACD-A (adenine, citrate, dextrose – A) as anticoagulant instead of UNH. After the cessation of heparin and the initiation of TPE with ACD-A, platelet count improved rapidly, recovering normal counts (104,000/ μ L after 1 TPE on day 2; and 169,000 / μ L after 3 TPE on day 5). HITAbs negativized after 5 procedures (on 10th day). The response pattern of HITAbs is shown in Table 1. Her condition evolved satisfactorily and was discharged some weeks later.

Summary / Conclusions: There is little scientific evidence on the treatment of HIT with TPE, which is usually reserved for selected patients. In this case, the primary indication for TPE was a rheumatological disease already under treatment with TPE and immunosuppressive treatment, factors that may contribute to explain the low clinical expression and the rapid and satisfactory evolution. Our patient achieved platelet normalization on day 5 of TPE, in line with published data on TPE as HIT initial therapy (Onuhua, Transfusion, 2020). Moreover, she showed a declining pattern of antibody titers until complete negativization. She did not show the rebound effect that has been described in some articles, which might be explained due to an inhibitory effect on HIT antibody-mediated platelet activation by plasma IgG, as in vitro data suggest (Jones, Blood, 2018). In conclusion, TPE can be a therapeutic alternative in selected cases of HIT, with plasma probably being a preferable replacement fluid, although more studies are necessary on this topic.

P716 | Abstract withdrawn

P717 | Analysis of a cohort of patients with autoimmune diseases submitted to therapeutic plasma exchange in a tertiary hospital during the years 2018–2024

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P715 - Table 1. Monitoring of HITAbs pre- and post-TPE

TPE number (ACD-A)	Pre (U/mL)	Post (U/mL)
1°	6.5	5
2°	4.8	0.9
3°	4.1	0.7
4°	1.6	0.3
5°	1.1	0.3
6°	0.7	0.5
7°	0.8	0.3

Background: Therapeutic plasma exchange (TPE) is a safe therapeutic tool with evidence-based indication in some autoimmune diseases. The guidelines of the American Society for Apheresis (ASFA) classify the indications for the use of therapeutic apheresis into four groups. Its application in those conditions included in the first category has been widely demonstrated, however, most centres continue using this therapy as a rescue treatment, despite a poorly established indication.

Aims: To analyze the experience of 59 cases of TPE performed in our hospital in the last five years in patients diagnosed with autoimmune diseases.

P717 - Table 1.

Category	Condition	Number of patients
I	Guillain Barre Syndrome (1st line)	13
	MG (acute)	8
	Demyelinating polyneuropathies	2
	NMDA receptor antibody encephalitis	2
	Chronic focal encephalitis	1
	Catastrophic Antiphospholipid Syndrome (CAPS)	2
II	Lupus	1
	Multiple sclerosis	5
	MG (long-term)	1
	Neuromyelitis optical spectrum disorder	3
III	Guillain Barre Syndrome (2nd line)	4
	Anti-glomerular basement membrane disease	1
	Chronic inflammatory demyelinating polyradiculoneuropathy	1
	Paraneoplastic neurological syndrome	1

Methods: Retrospective, single-center cohort study that included all TPE procedures performed from January 2018 to January 2024 in autoimmune pathology.

Results: Fifty nine cases of TPE were analyzed in a total of 58 patients, one of them with two indications implying different categorization. The mean age of the patients was 57 years with a predominance of males (57.6%). An average of 5.68 sessions were performed per patient every 48 h: 64.4% during the first episode and the rest during an outbreak. We used albumin in 81.4% of cases ($N = 48$), Fresh Frozen Plasma (FFP) in 13.6% ($N = 8$), both in 5.1% ($N = 3$). The autoimmune diseases and indication group for which TPE was performed are shown in Table 1. In 52.54% of cases they were classified in II-III category. Complications were only found in 17 patients and solved with symptomatic treatment. Two cases were related to catheter, characterized by flow resistance or bleeding. The rest were associated to the procedure including hypotension (self-limited or severe) and mild anaphylactic reactions (rush or pruritus) as the most relevant. A high clinical response rate has been observed with a success rate of 78%. There is a statistically significant association between belonging to an indication group and the improvement of the pathology after TPE ($p = 0.016$), coinciding our experience with the recommended of the ASFA guidelines. The pathology that benefited most from TPE was myasthenia gravis (MG) where 66.7% improved completely and 33.3% improved partially. On the other hand, the most disadvantaged pathology was vasculitis, where 42.90% did not obtain a response, 21.4% responded partially, and 35.7% responded fully. We found no statistically significant association between clinical improvement and plasma exchange at the first episode rather than at relapse ($p = 0.91$). No significant differences were found in terms of improvement

(including partial improvement) between patients who underwent TPE in the first five days after diagnosis and those who had it later ($p = 0.27$).

Summary / Conclusions: Used as a first-line, TPE has been shown an effective therapy. In our center we have used TPE in many cases with indication category II-III, despite the low scientific evidence, without actually showing significant benefit. In our experience it proved to be a safe therapy, with few mild adverse effects, unrelated to the type of replacement solution used.

P718 | Abstract withdrawn

P719 | Abstract withdrawn

P720 | Management of Juvenile Dermatomyositis (JDM) with Therapeutic plasma exchange (TPE)

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Background: JDM is a rare immune mediated disease involving swelling and inflammation in blood vessels of skin and muscles which make proximal muscle weakness and skin manifestation.

Aims: There are some studies done TPE as a treatment modality for management of JDM. This case is based on JDM management with TPE as a rescue therapy.

Methods: A 9 year-old previously healthy boy developed periorbital swelling with facial puffiness, photosensitive rash over the nasal bridge 2 months ago. He developed Arthralgia, swelling and itching whole body and treated as allergy and responded. 7 days later periorbital swelling and facial puffiness reappeared. There was restriction of movement, limping, and calf pain with difficulty to stand. All movements restricted due to pain. No food or drug allergy and family history of rheumatoid disease. Mild bilateral periorbital oedema, facial puffiness, oral ulcer and Gottorn's papules noted over metacarpophalangeal joints with positive Raynaud's phenomenon. Bilateral upper limb and lower limb proximal muscle weakness noted with 4/5 power and preserved tone and reflexes. Myositis with skin manifestation suggestive JDM and started oral Prednisolone, Methotrexate weekly, Folic acid daily. As worsening of proximal muscle weakness while on oral prednisolone, IV methylprednisolone started. As poor response, oral prednisolone was increased. Methotrexate was withheld due to elevated liver enzymes. But patient developed worsening proximal muscle weakness with new onset of bulbar muscle involvement—nasal regurgitation and dysphonia and referred to transfusion team for TPE as a rescue therapy. Child was assessed and planned to do TPE every other day. Pre-procedure investigations normal. A total of 1000 mL of plasma was removed and replaced with a 5% albumin and Normal saline. He was hemodynamically stable throughout the procedure,

except mild itching developed without urticaria which was related to his disease condition, and it was managed with antihistamine. The patient showed improvement in dysphonia after the 1st cycle. Proximal muscle weakness improved markedly following 4 cycles, and the child was able to walk without difficulty. After completing 6 cycles of TPE, child was able to walk without support, and swallowing had improved, and NG tube was removed. The CPK level was reduced from the initial 2759 to 40 u/l. Physiotherapy continued to strengthen muscle power. He was started with second-line treatment with IV Rituximab 400 mg two weekly, and methotrexate increased to 12.5 mg weekly and was discharged without residual weakness.

Results: His investigations (FBC, S.Ca, Coagulation profile, Renal Profile, S.Albumin) were normal throughout procedure. LDH level was high (954 u/l). Blood picture, Chest xray, Lung function, 2D-Echo normal. Anti-Nuclear Antibody and Montoux negative. Electromyography suggestive of inflammatory myopathy/myositis.

Summary / Conclusions: This case proves TPE, as rescue therapy in Multi-Disciplinary treatment of JDM. Need further evidence to confirm and include JDM as an indication for TPE international apheresis guideline.

P721 | Leukocytapheresis—new challenges in therapy of the patients

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Background: Leukocytapheresis (LA) is a specific apheresis technique with predominant collection of leukocytes. It is used routinely for the reduction of the extent of leukocytes in patients with hyperleukocytosis as well as in hematopoietic stem cell collections (PBPC) in mobilized donors and patients. Recently, LA plays an increasing role in more specific antitumor therapy by the use of chimeric antigen receptor (CAR) T cells in the treatment of some B lymphoproliferative diseases.

Aims: The aim of the study was to evaluate the results of productive leukocytapheresis procedures, i.e., MNC, and PBPC collections in different groups of patients and donors. Understanding the process would be helpful in optimizing apheresis procedures.

Methods: MNC and PBPC collections were performed in groups of: **Non-mobilized patients and donors** MNC were collected for extracorporeal photochemotherapy “off line”(ECP), for CAR T cells, and for DLI in: patients: with acute and chronic GVHD (a/c GVHD, 119 procedures, 8 patients), and patients with B cell lymphoproliferative diseases - ALL, DLBCL (155 procedures, 147 patients); healthy donors: who were collected for DLI (7 procedures, 7 donors). **Mobilized PBPC patients and donors** PBPC were collected for autologous and allogeneic transplantation in: patients: with non-Hodgkin lymphoma and multiple myeloma for autologous HSCT transplantation (132 procedures, 79 patients); healthy donors: who were collected for allogeneic HSCT transplantation (46 procedures, 38 donors, Zarzio). Collections were performed using Spectra

Optia, v. 11, Terumo, CMNC, MNC. The precollection numbers of leukocytes, CD 34+ cells, CD 3+ cells in blood, as well as the numbers of leukocytes, percentage (%) of MNC, CD 3+ cells, and CD 34+ cells in products were evaluated (Sysmex XN 10, BD FACS Canto II).

Results: Total blood volumes processed (\times TBV) were in patients and donors: GVHD 1.4 (1-1.7), ALL/DLBCL 2.6 (2.1 - 3.3), DLI 1.4 (0.8-1.6), PBPC autologous 3.6 (2-5.4), PBPC allogeneic 3.2 (1.3 - 4. 7). The results are expressed as medians and their ranges. We found in MNC products: Number of leukocytes: GVHD 7 (4-12), ALL/DLBCL 14 (2-113), DLI 7 (4-12), PBPC autologous 246 (36-829), PBPC allogeneic 328 (119-536) $\times 10^9$; Percentage of MNC: GVHD 86 (45-97), ALL/DLBCL 93 (28-99), DLI 86 (54-98), PBPC autologous 69 (20-98), PBPC allogeneic 72 (30 - 91)%; percentage of CD 3+ cells: GVHD 52 (19-92), ALL/DLBCL 86 (64-97), DLI 42 (14-50), PBPC allogeneic 24 (4-47)%; the median yield of CD 34+ cells from one collection in PBPC autologous 5.7 (1-50), PBPC allogeneic: 5 (0.2-18) $\times 10^6$ /kg b.w. of the patient, the median yield of CD 3+ cells from one collection in MNC for CAR-T was 5.4 (0.4-28) $\times 10^9$.

Summary / Conclusions: Spectra Optia was proved to be an efficient system in the process of MNC and PBPC collections. We obtained the sufficient numbers of MNC, CD 34+, and CD 3+ cells for therapy of the patients in majority of procedures. Percentage of MNC in the products was in non-mobilized patients and donors higher than in mobilized PBPC donors. Percentage of CD 3+ cells in non-mobilized patients was higher than in donors of DLI non-mobilized, and donors of PBPC mobilized. The cause of such differences is not clear yet, and will be studied in near future. No serious adverse reactions in the course of collections have been observed.

P722 | Abstract withdrawn

P723 | A comprehensive case report of refractory acute disseminated encephalomyelitis in a pediatric patient emphasizing successful intervention with therapeutic plasma exchange

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Background: ADEM is an inflammatory demyelinating typically monophasic disease predominately affecting the central nervous system. Children and young adults are commonly affected. Diagnosis is challenging due to the heterogeneity of clinical presentation. Therapeutic aim in ADEM is to minimize the immune mediated neuronal damage. Early treatment improves both acute and long-term clinical outcome.

Aims: TPE is accepted as second-line therapy in ADEM. We wanted to see the clinical efficacy of TPE in steroid refractory ADEM.

Methods: Ten-year-old girl presented to local hospital with fever, vomiting and diarrhea for 3 days which was associated with increased drowsiness, emotional liability, and difficulty in walking. During the hospital stay she developed one episode of generalized tonic clonic seizure associated with increased tonic activity, urinary incontinence, and loss of consciousness. Following the seizure, patient continued to

exhibit persistent drowsiness. She was given IV Midazolam and started on broad spectrum IV antibiotics and antivirals considering a potential diagnosis of meningoencephalitis. Due to persistent drowsiness, she was transferred to Lady Ridgeway Hospital for Children (LRH). She was a diagnosed patient with seizure disorder at the age of 2 years and was on Sodium Valproate. Treatment had been discontinued for four years, as she was seizure-free for more than two years. She is the only child of a non-consanguineous parents. No family history of seizure disorders. No birth trauma. Developmental milestones were age-appropriate except delayed gross motor development during childhood. Immunization was up to date. On admission to LRH, child was hemodynamically stable. GCS 11/15 but was drowsy and irritable. Hypertonicity in limbs with reduced power noted. Reflexes were difficult to elicit. Pupils equally reacted to light.

Results: Full blood count, serum electrolytes, coagulation profile and CRP were normal. Cerebrospinal fluid (CSF) full report showed mild pleocytosis with normal CSF protein and sugar. CSF for NMDR antibodies, virology screening for hepatitis B, C, HIV, HSV, CMV and EBV were sent. EEG showed generalized slowing of electrical activity. USS brain and CECT showed mild cerebral oedema. MRI brain revealed multiple subtle T2W/ FLAIR high signal intensities with restricted diffusion suggestive of ADEM. She was started on high-dose IV steroids daily for five days. IVIG 2g/kg was given over 2 days. Despite these interventions, patient deteriorated leading to respiratory acidosis. She was transferred to medical intensive care unit, intubated, and ventilated. TPE was considered as a rescue therapy. One total plasma volume (TPV) removed and replaced with 5% albumin. Five cycles were done every other day. Child was improved and extubated after 3rd cycle of TPE. Her clinical condition markedly improved at the completion of 5 cycles and discharged after one month of hospital stay.

Summary / Conclusions: Management of ADEM needs multidisciplinary team approach. The positive response to TPE shows its potential in management of cases refractory to conventional treatment.

Clinical transfusion—evidence based transfusion medicine practice

P724 | Efficacy of VaxCCP transfusion in immunocompromised patients with SARS-CoV-2 infection

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P724 - Table 1. Baseline characteristics of patients treated with VaxCCP.

Clinical features	Patients treated with VaxCCP (n = 167)
Age, years	67 (IQR 54 - 75)
Men - female	105 (63%) / 62 (37%)
Cause for being immunocompromised	
Oncohematological diseases	156 (93.4%)
Autoimmune diseases	7 (4.2%)
Solid organ transplant	4 (2.4%)
Vaccinated	146 (87.4%)
Mild SARS-CoV-2	71 (42.6%)
Pneumonia caused by SARS-CoV-2	62 (37.1%)
Persistent SARS-CoV-2 infection	34 (20.3%)
Use of concomitant remdesivir or other antivirals	140 (84.3%)

P724 - Table 2. Response to VaxCCP.

Measure of efficacy	Patients treated with VaxCCP (n = 167)
Decrease in viral load at non-infective levels (CT>30)	87 of 129 (67%)
WHO scale, points	
Pre VaxCCP values	4 (IQR 3-5)
Post VaxCCP values at 15 days	1 (IQR 1-2)
Restart of onco-hematological treatment	62 of 75 in whom onco-hematological treatment was delayed (83%)
Survival at 6 months	81% (CI 95% 75-87)
Survival at 12 months	72% (CI 95% 64-79)
Causes of death (n = 39)	
Primary disease progression	21 (53.6%)
SARS-CoV-2 infection	9 (23.2%)
Other	9 (23.2%)

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Background: Immunocompromised patients are at high risk of both severe acute and persistent SARS-CoV-2 infection because of a deficient immune response. These patients could, therefore, benefit from treatment with high-titer anti-SARS-CoV2 post-vaccine plasma (VaxCCP) but literature is still scarce.

Aims: To investigate the efficacy of high-titer anti-SARS-CoV2 VaxCCP in immunocompromised patients with mild/severe acute and persistent SARS-CoV-2 infection.

Methods: We retrospectively reviewed all immunocompromised patients treated with VaxCCP between June 2021 and December 2022 at the Hospital Clinic of Barcelona. VaxCCP was obtained from male donors with infection of SARS-CoV-2 in the previous 6 months and at least two doses of SARS-CoV-2 vaccine, and whose anti-SARS-CoV-2 titer was $\geq 20,000$ BAU/mL (Abbott ARCHITECT SARS-CoV-2 IgG assay). Efficacy was evaluated by: (1) the decrease in viral load to non-infective levels (cycle threshold >30), (2) clinical improvement according to the WHO scale, (3) restarting of onco-hematological treatment if previously stopped, and (4) survival at 6 and 12 months. A multivariate analysis for mortality at 12 months was performed.

Results: During the analyzed period, 167 patients received VaxCCP. Their main baseline characteristics are shown in Table 1. Response to VaxCCP assessed according to above mentioned efficacy criteria is summarized in Table 2. In multivariate analysis, older age (HR 1.03, 95% CI 1.01-1.06, $p = 0.02$) and having metabolic comorbidities (HR 2.30 95% CI 1.02-5.25, $p = 0.04$) were associated with a higher mortality risk at 12 months. Sex, previous vaccination and ABO group did not have a significant effect. Regarding safety, 9 minor allergic reactions were reported.

Summary / Conclusions: VaxCCP seems to be useful in immunocompromised patients with SARS-CoV-2 infection.

P725 | Convalescent plasma to treat COVID-19 in immunocompromised patients

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Background: At the start of the COVID-19 pandemic, treatment options were scarce. Historically, convalescent plasma has been regarded as a safe and promising treatment and many clinical trials with COVID-19 convalescent plasma (CCP) were performed, mostly resulting in insignificant outcomes on mortality or clinical recovery compared to placebo or standard care. However, recent studies have shown potential benefits of CCP with high (neutralizing) antibody titers for immunocompromised patients, especially in patients with low or absent (neutralizing) antibody titers. In the Netherlands, high-titer anti-SARS-CoV-2 IgG CCP may be requested for immunocompromised patients in compliance with compassionate use and is provided by Sanquin Blood Supply for this patient population only.

Aims: To investigate the impact of CCP on the clinical status of immunocompromised patients 7 days after transfusion, measured on the

P725 - Table 1: Mean difference of WHO-CPS before and after CCP transfusion, categorized by clinical status at intake

	Mean difference of WHO- CPS (SD)	df	p	Cohen's D
Overall	0.68 (2.35)	404	<0.001	0.290
Clinical status at intake (score)				
Ambulatory mild disease (1-3)	0.68 (1.60)	76	<0.001	0.422
Hospitalized: moderate disease (4-5)	1.16 (2.31)	226	<0.001	0.501
Hospitalized: severe disease (6-9)	-0.39 (2.56)	100	0.133	-0.151

WHO clinical progression scale (WHO-CPS), a scoring system ranging from 0 to 10, a higher score indicates a worse clinical status.

Methods: A quasi-experimental quantitative one-group pretest-posttest study was performed. Information was collected by Sanquin blood supply between May 2022 and January 2024 from over 30 hospitals in the Netherlands, with a final cohort of $N = 405$ patients. The collected data consisted of demographics (age, sex), indication for receiving CCP, WHO-CPS status at intake and 7 days after treatment and whether prevalent SARS-CoV-2 variants at time of plasma donation matched the variants at time of administration. Information on circulating SARS-CoV-2 variants was obtained from the Dutch National Institute for Public Health and the Environment. Statistical analysis was performed by a series of paired t-tests, measuring outcome as mean difference, where a positive score indicates a decrease in WHO-CPS, thus clinical improvement. Cohen's D was added to estimate effect size. Five subgroup analyses were performed.

Results: The overall analysis showed a significant mean difference between the clinical status at intake and 7 days after treatment (table 1). Stratification for WHO-CPS status at intake showed the largest effect in patients with mild (WHO-CPS 1-3) or moderate (WHO-CPS 4-5) disease. Patients with severe disease (WHO-CPS 6-9) showed no significant differences. Stratification for age showed significant effects in the patient group aged 45-64 years (mean dif. = 0.67 (SD = 2.25), $df = 122$, $p = 0.001$, Cohen's D = 0.297) and the patient group aged 65-79 years (mean dif. = 0.77 (SD = 2.36), $df = 216$, $p < 0.001$, Cohen's D = 0.326). No significant effects were found for the age groups 18-44 years and 80 years and over. No significant differences were found comparing sex, indication and SARS-CoV-2 variants.

Summary / Conclusions: COVID-19 convalescent plasma transfusion appears to be a significant factor in improving clinical status for immunocompromised patients. The largest effect is seen in patients with mild or moderate disease. The study will be finalized in the upcoming months and supplemented with extra subgroup analyses.

P726 | Do red blood cells from chronic mountain sickness patients qualify for transfusions?

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Background: Chronic Mountain Sickness (CMS) manifests in high-altitude residents above 2500 m, characterized by symptoms associated with elevated hematocrit levels typically ranging between 60% and 80% caused by excessive erythrocytosis (EE). This results in elevated hemoglobin, exceeding 21 g/dL for men and 19 g/dL for women. A commonly employed approach for CMS management involves phlebotomy, typically extracting 4.5-9 dL of blood, which is subsequently discarded. In contrast, there is a scarcity of blood supply, particularly in low and medium-income countries in the Andes region such as Ecuador, Peru, or Bolivia, which also have a high incidence of CMS.

Aims: Our study aimed to delve deeper into the characterisation of CMS patients, focusing particularly on the properties of their red blood cells (RBCs) and evaluating the option to use the RBCs of CMS patients for transfusion purposes.

Methods: We investigated 62 male volunteers residing permanently in La Rinconada, Peru, 5100 m above sea level. Following the prevailing international consensus, a diagnosis of CMS was established when the Qinghai CMS score was ≥ 6 , with the presence of EE. We identified 36 individuals as CMS patients and 26 as healthy controls. RBC deformability, a critical parameter for assessing the quality of RBCs for transfusion purposes, was evaluated using ektacytometry. In addition, aggregation indices, M (stasis) and M1 (very low shear) were acquired with an aggregometer.

Results: Mean RBC volume and mean RBC haemoglobin concentration are not statistically significantly different ($p = 0.14$ and $p = 0.75$, respectively) and mostly within reference values. Small variations in these parameters may be influenced by the distinct count of reticulocytes (3.4 ± 1.5 vs. 2.6 ± 1.2 for CMS patients vs. controls), given their 24% higher volume and 16.7% lower haemoglobin concentration compared to mature RBCs. Microscopic examination of RBCs did not unveil any discernible differences in shape between CMS patients and controls. Viscosity measurements based on a normalized hematocrit of 40% revealed no significant

differences between CMS patients and controls. We investigated three different shear stresses (0.3 Pa, 3 Pa and 30 Pa) with the shear of 3 Pa being the physiologically most relevant condition - no significant differences were observed between CMS patients and controls ($p = 0.43$, $p = 0.87$ and $p = 0.33$, respectively). Similarly, RBC aggregation indices (M and M1) exhibited no discernible differences between CMS patients and controls. Finally, as a measure of RBC stability, we monitored the elongation index (at a constant shear of 30 Pa) over a 10-min duration. No significant differences were detected between CMS patients and controls. In essence, the mechanical and rheological properties of RBCs from CMS patients closely resemble those of controls.

Summary / Conclusions: Considering that blood bank products are processed as blood constituents—not whole blood—the above-mentioned physical properties of RBCs are most relevant. In light of these findings, we propose that blood obtained through phlebotomy for treating CMS patients holds the potential as a viable source for blood transfusions. However, this would require a systematically scheduled phlebotomy for CMS patients with a controlled substitution of plasma, iron and amino acids. Certainly, the suggestion to employ RBCs from CMS patients for transfusions, as proposed through the *in vitro* investigations of blood samples in this study, warrants validation through clinical trials.

P727 | Plasma proteome profiling reveals signatures of inflammation and dysregulation of hemostasis in acute myeloid leukemia patients undergoing intensive-transfusion treatment

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Background: Bone marrow aplasia is a common feature in acute myeloid leukemia (AML) patients during their remission induction treatment. This causes long periods of thrombocytopenia and potential complications such as bleeding and anemia. In order to prevent and treat these complications, frequent platelet and red blood cell transfusions are administered. However, studies in this patient population have shown that platelet counts have a limited association with bleeding and hence, additional factors are needed to improve monitoring of these patients. Unbiased proteomics have been employed in several diseases to map its progression and association with potentially affected pathways in patients. Therefore, plasma protein levels can add valuable insights to our understanding of the patient's health state throughout treatment. Importantly, changes in the plasma proteome in AML patients receiving transfusion products have not yet been studied. Thus, defining protein profiles during this phase is key

to use as reference point prior to tracing patients' treatments and their responses.

Aims: We aimed to map out the plasma proteome of AML patients during treatment receiving intensive supportive transfusions compared to healthy controls.

Methods: We employed unbiased mass spectrometry (MS)-based proteomics on longitudinal plasma samples ($n = 37$) from 11 AML patients during intensive-transfusion treatment phase. Plasma from healthy individuals ($n = 11$) was used as baseline control. In brief, plasma samples were processed to obtain proteins which were digested overnight. Peptides were analyzed using a LC-MS based proteomics approach and results were interpreted using statistical analysis and evaluation of protein dynamics between the two groups based on Pearson coefficient correlations.

Results: A total of 456 proteins were quantified in plasma samples from AML patients and healthy controls. We detected a constant increase of HAMP in AML patients throughout their transfusion treatment phase likely driven by a dysregulation in iron metabolism and inflammation. We also found lower levels of CAMP in this phase probably caused by the bone marrow aplasia experienced by these patients. In addition to an expected acute phase response, we also observed significant alterations in proteins levels involved in complement (e.g. C9 and MASP2) when comparing AML versus healthy individuals. Global correlation analysis revealed additional affected protein dynamics. These alterations included proteins associated with coagulation (i.e. F10 and SERPIND1) and complement cascade (i.e. C3 and C9) suggesting a disbalance in the interplay of coagulation and complement systems. Furthermore, we found a decreased abundance in proteins enriching for lipoprotein remodeling (i.e. APOA1 and PON1) throughout the treatment phase in these patients pointing towards a potential unbalanced lipid metabolism.

Summary / Conclusions: The plasma proteome from AML patients during intensive transfusion treatments shows a disbalance in inflammation, endopeptidase inhibitors activity, lipoprotein remodeling, coagulation and complement. These alterations could be associated with differences in transfusion responsiveness and bleeding risk. Analysis of bigger cohorts are currently being used to delineate these observed effects and their potential association with bleeding events and bone marrow recovery.

P728 | Low voxelotor concentrations exacerbate sickle cell formation at low oxygen pressure

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Background: Sickle cell disease (SCD) is the most prevalent inherited monogenic blood disorder, affecting millions of people worldwide. The formation of the abnormal hemoglobin S (HbS) leads to

hemoglobin (Hb) polymerization and red blood cell (RBC) deformation and rigidity, primarily at low oxygen conditions and pathology. The novel drug Voxelotor binds to and stabilizes Hb-tetramers in the high-O₂ affinity R-state, thereby preventing HbS polymer formation.

Aims: We hypothesized that under hypoxic conditions Voxelotor bound Hb-R-state-tetramers scavenge the limited amounts of O₂, thereby depriving non-Voxelotor bound T-state Hb from O₂. Consequently, at low doses of Voxelotor this will lead to increased HbS polymerization, potentially resulting in clinical manifestations. If accepted, this may have implications for monitoring therapy efficacy, drug discontinuation strategies and individual drug exposure. Validation requires a robust assay to quantitatively determine sickle cell formation at single cell resolution and at sufficiently large numbers, which is currently not available

Methods: To this end, we have developed a novel high throughput method combining imaging flowcytometry and artificial intelligence to discriminate the morphological changes in sickle RBCs using limited number of cells (1-2 μ L). With this method we are able to discriminate and quantify discocytes, sickle cells and intermediary morphologies (holy leaf cells and granular cells) with high accuracy, low inter-sample variability (CoV<0.043), and at single cell resolution. The method was independently validated on healthy controls ($n = 10$) and SCD patients ($n = 124$).

Results: This method was applied to understand how different Voxelotor concentrations at hypoxic conditions affect sickle cell formation. At 1% O₂, we found a clear dose-dependent (0, 10, 30, 60, 90 and 120 μ M) increase in discocytes and a decrease in holy leaf cells. At high Voxelotor concentration, we observed the expected dose-dependent response, showing a decrease in the more severe sickle cell morphologies, granular cells and sickle cells. In contrast, these severe SCD morphologies increased at low drug concentrations, peaking at 30 μ M Voxelotor with a 2-fold increase compared to 0 μ M voxelotor. Gradually making O₂ molecules available to non-Voxelotor bound T-state Hb-tetramers by increasing O₂ concentrations (1%, 2.7%, 5.3%, 10.7%, 15% and 21%) moved the peak of increased sickle cells and granular cells from 30 μ M Voxelotor at 1% O₂ to 10 μ M at 2.7% O₂ followed by a dose-dependent decrease in granular and sickle cells at 5.3% O₂. This indicates that low concentrations of Voxelotor under hypoxic conditions increase sickle cell formation by disturbing the R-state versus T-state Hb balance in RBCs of SCD patients

Summary / Conclusions: These findings have important clinical consequences and indicate that therapy management and therapy compliance are essential in disease management. The clinical importance of this mechanism is further supported by a recent case-report, which describes a SCD patient with two hospital admissions due to VOCs and a severe drop in Hb, 3 or 4 days after abrupt discontinuations of Voxelotor treatment (Nagalapuram et al, AJH, 2022). In addition, our newly developed high-throughput AI-based imaging flow cytometry assay enables us to elucidate and predict the efficacy of different concentrations of Voxelotor or other disease-modifying drugs, potentially resulting in individualized drug doses in SCD patients.

P729 | Abstract withdrawn

P730 | Situational analysis of clinical blood transfusion practices among physicians at the Cameroon Baptist Convention Health Services—a step towards quality services

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Background: Prescription of blood components is the starting point for blood transfusion activities. To meet these needs, blood banking ensues to provide what the physicians need. Thus having a perspective of the physician's understanding of blood transfusion is paramount in achieving available, prompt, safe, and quality blood to patients. Moreover, considering the backbone that blood transfusion provides to most services in the hospital, we embarked on having an in-depth perspective of this practice in the Cameroon Baptist Convention Health Services (CBCHS).

Aims: We thus sought to understand physicians' knowledge and attitudes on some concepts in clinical transfusion practice. In addition, we were interested in describing their attitude towards voluntary non-remunerated blood donation.

Methods: This was a cross-sectional survey from October 2022 to April 2023 involving 10 hospitals within the CBCHS among prescribing healthcare physicians. A questionnaire was developed and pre-tested before recruitment of participants. This comprised of 23 questions wherein the responses were required in various formats among which; 10 of them were single-tick box answers, 8 short phrases answers, 4 open ended questions and 1 multiple-tick boxes answer. Consenting participants were included and questionnaires with incomplete answers were excluded. Microsoft excel 2013 was used to analyze the data.

Results: Out of the 100 participants, 92 were included in the study. The majority were females (57.6%), with physicians having less than 5 years of clinical experience (64.1%). Whole blood/pRBCs were needed more frequently (21.7%) compared with platelets (6.5%) and plasma (3.3%) in daily practice. Also, 48.9% admitted that unavailability of timely transfusions occurred in 4 to 7 requests. 57.6% based their transfusion decision primarily on the clinical state of the patient as opposed to 35.9% who prioritized hemoglobin trigger. Based on the WHO recommendation of hemoglobin trigger, the internal medicine department (86.9%) had the best transfusion practice compared with neonatology (22.8%) and pediatrics (4.4%). Moreover, only 8.6% used a transfusion guideline, with 21% never formally trained in blood transfusion and 71.1% using the single-unit transfusion protocol. With regards to patient blood management, 40.9% used complete blood count as the primary mode of diagnosing anemia against 5.5% who sought the etiology. Furthermore, oral hematinic (53.7%) was the most prescribed alternative form for the management of anemia as opposed to IV iron (8.8%) and erythropoietin synthetic analogs (2.5%). Finally, 61.9% had never engaged in at least one voluntary blood donation over the last 12 months.

Summary / Conclusions: Building interventions that could be contextualized, implemented, monitored, and evaluated would be necessary to improve the standards of the blood transfusion services in CBCHS. This involves training on blood component use, developing local transfusion guidelines, strengthening patient blood management programs, and improving voluntary blood donation rates among physicians

P731 | Abstract withdrawn

P732 | Hemostatic evaluation of thrombocytopenic patients receiving platelet transfusions, using the Total Thrombus Formation Analysis System (T-TAS)

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Background: Thrombocytopenic patients show an increased risk of bleeding, they frequently receive prophylactic platelet transfusions. Criteria for administering transfusions are a topic of debate and controversy. Assessment of their hemostatic status can be useful in this scenario.

Aims: Evaluating the potential value of the Total Thrombus Formation Analysis System (T-TAS) to assess the hemostatic status of thrombocytopenic patients receiving platelet transfusion.

Methods: Fifty-five thrombocytopenic patients (64.4 ± 13.8 y; platelets $25.5 \pm 20.3 \times 10^9/L$) were evaluated. Hematologic malignancy was the principal baseline diagnosis (39 patients, 70.9%). Platelet transfusion ($3.1 \pm 0.98 \times 10^{11}$) was indicated as prophylaxis, before an

P732 - Table 1. Clinical Parameters (n = 55).

Age (years), mean \pm SD	62 \pm 14
Gender, n (%)	Men 33 (60)
Underlying Pathology, n (%)	Hematological Malignancies 39 (69)
	Sepsis 3 (6)
	Liver Failure 8 (15)
	Chronic Renal Disease 2 (4)
	Post-surgery bleeding 3(6)
Fever, n (%)	Yes 9 (16)
Platelets shelf life (days), mean \pm SD	4 \pm 1
Platelet ABO compatibility, n (%)	Identical 16 (48)

P732 - Table 2. Analytical Parameters (n = 55).

	BT	AT
Hb (g/dL)	9.5 ± 1.7	9.2 ± 1.5*
Hto (%)	27.5 ± 5.2	27.0 ± 4.5*
Platelets (× 10 ⁹ /L)	25.5 ± 20.3	45.5 ± 24.1*
Fibrinogen binding (PAR1- 25 uM)	1490 ± 2333	1703 ± 2280*
CD62 (PAR1- 25 uM)	1247 ± 1306	1364 ± 1556*
T-TAS Area Under Curve (AUC)	44 (14–1427)	489* (16–1509)
T-TAS OT (min)	30 (4-30)	27* (5-57)
Thrombin Generation ETP (min)	1015 ± 293	1015 ± 284*

Parameters are expressed as mean ± SD, except for CCI 1 h, T-TAS, AUC y T-TAS OT, which are referred as median (interquartile range). **p* < 0.05 AT vs. BT.

invasive procedure or as therapy for bleeding (28, 18 and 9 patients, respectively). Bleeding was graded following the WHO bleeding scale, dividing patients into mild (≤1) or severe (≥2) bleeding (mb, MB), 36(65.5%) and 19(35.5%), respectively. Blood samples were drawn immediately before (BT) and after (AT) transfusion for complete blood count, flow cytometry assessment of PAR-1 (25 uM) induced platelet binding of fibrinogen (Fg) and P-Selectin (CD62) release, Thrombin Generation Essay (TG) and HD chip T-TAS test (Fujimori Kogyo Co). The study was approved by the local Ethics Committee. Patients gave written informed consent.

Results: Patients clinical features and test results are specified in Tables 1 and 2. The Corrected Count Increment (CCI) 1 h after transfusion was 11 (0-43). This meant an adequate response to platelet transfusion in 37 cases (67%) (CCI > 7.5). These results were similar between hematologic (HP) and non-hematologic patients (NHP) and between those with MB and mb. This was not affected by other conditions like fever, nor by platelet's shelf life. About TG, the overall results improved AT (*p* < 0.05). NHP and patients with MB showed poorer results in this test. In HD-T-TAS, the area under curve (AUC) improved AT (44(14-1427) vs. 489(16-1509)), (*p* < 0.01). BT&AT, AUC values correlated (*p* > 0.05) with hemoglobin, platelet count and platelet CD62 secretion. AUC values correlated negatively (*p* < 0.05) with platelet's shelf life and with TG results BT&AT. T-TAS results AT

were significantly poorer in HP (197 (16-1300) vs. 1202 (265-1509)). Those patients with worse results in T-TAS analysis needed more platelet transfusions in the following 7 days (*p* < 0.05).

Summary / Conclusions: This study suggests that T-TAS-HD chip is potentially useful for hemostatic evaluation in thrombocytopenic patients. These results are sensitive to platelet counts, hematocrit, and platelet reactivity. It also differentiates hematologic and non-hematologic patients.

P733 | Audit of transfusion rates in Australian lung transplant patients

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Background: Australia performs approximately 200 lung transplants (LTx) annually with Alfred Health performing almost 50% of these (ANZOD Registry, 2023). The transfusion rate in LTx reported from a single USA centre was 56% (Huddleston et al., 2020). Despite the frequent transfusion rate of LTx recipients, published data exploring use of blood components during LTx in Australia is lacking.

Aims: Identify fresh blood component administration rates during the index admission of LTx recipients and evaluate the impact of patient demographics, single versus double LTx, and use of CPB on transfusion rates.

Methods: A retrospective audit of all LTx performed at Alfred Health between the 1 January 2018 and 30 June 2023 was performed. Patient demographics, LTx specific variables and transfusion data (red blood cells [RBC], platelets, fresh frozen plasma [FFP] and cryoprecipitate) were extracted from laboratory, LTx clinical database and medical record systems. Fresh blood components transfused were only included if they occurred during the hospital admission of LTx. This data was then collated into Microsoft Excel spreadsheets.

Results: A total of 434 LTx patients were included in the audit. Of these 298 (68.7%) were transfused. Patient characteristics are shown in Table 1. Table 2 contains the indications for LTx and transfusion rates. The septic lung disease category included cystic fibrosis. A transfusion rate of 90.9% (*n* = 110) occurred for LTx on CPB and 61.1% (*n* = 324) in non-CPB. The use of CPB was highest in

P733 - Table 1.

	All transplants (n = 434)	Transfused (n = 298)/Not transfused (n = 136)	Single lung (n = 67)/Double lung (n = 367)	CPB (n = 110)/No-CPB (n = 324)
Men	256	165/91	48/208	63/193
Women	178	133/45	19/159	47/131
Years of age*	59 (5-73)	59 (5-73)/61 (14-73)	64 (26-73)/59 (5-73)	58 (5-73)/ 60 (9-72)
BMI*	24 (13-32)	24 (13-32)/25 (16-32)	25 (19-30)/24 (13-32)	24 (14-32)/25 (13-32)

ICU = intensive care unit, LOS = length of stay, BMI = body mass index.

* Data expressed as median (range).

P733 - Table 2.

Indication for transplant	Patients transfused RBC	Patients transfused platelets	Patients transfused FFP/cryoprecipitate	Total patients transfused
Obstructive lung disease (n = 142)	80 (56.3%)	31 (21.8%)	50 (35.2%)/24 (16.9%)	90 (63.4%)
Restrictive lung disease (n = 174)	76 (43.7%)	56 (32.2%)	74 (42.5%)/37 (21.3%)	106 (60.1%)
Septic lung disease (n = 43)	33 (76.7%)	11 (25.6%)	18 (41.9%)/10 (23.3%)	34 (79.1%)
Pulmonary hypertension (n = 40)	34 (85%)	31 (77.5%)	36 (90%)/24 (60%)	39 (97.5%)
CLAD (n = 32)	26 (81.3%)	13 (40.6%)	18 (56.3%)/12 (37.5%)	26 (81.3%)
Other (n = 3)	1 (33.3%)	1 (33.3%)	3 (100%)/1 (33.3%)	3 (100%)
Total (n = 434)	250 (57.6%)	143 (32.9%)	199 (45.8%)/108 (24.8%)	298 (68.6%)

pulmonary hypertension (n = 34, 85%). Transfusion rates in double LTx recipients was 73.0% and 44.8% in single LTx. Most patients who received RBC were transfused post-operatively (n = 235, 94%) compared to intra-operative (n = 83, 33.6%). 18 patients (7.2%) received RBC pre-operatively, of these 6 were receiving extracorporeal membrane oxygenation support. FFP was administered to 20 patients pre-operatively, 14 of these patients had been taking warfarin.

Summary / Conclusions: Incidence of transfusion remains high in patients undergoing double LTx, compared to single LTx. Recipients with pulmonary hypertension had a particularly high transfusion rate. LTx requiring CPB had a higher transfusion incidence compared to non-CPB. Further exploration of factors influencing transfusion rates is required to determine if transfusion practice in LTx should be revised.

P734 | Abstract withdrawn

P735 | Abstract withdrawn

P736 | Complexities of Red Blood Cell (RBC) transfusion in patients with Myelodysplastic Syndromes (MDS)—a prospective time-driven, activity-based study

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Background: Many patients with MDS need RBC transfusion, and safe transfusion depends on a complex interplay of processes and

resources. However, few data are available on the individual steps, decision pathways, and associated resources/costs involved.

Aims: To identify and quantify all healthcare resources required for each step of the pathways for MDS patients receiving inpatient (IP) and outpatient (OP) RBC transfusions. These process maps and timings form the basis for health economics analyses currently underway.

Methods: Prospective study of transfusion-related activities for MDS patients at Monash Medical Centre, a 640-bed university teaching hospital in Melbourne, Australia. Multiple patients and processes were observed, including phlebotomy for pre-transfusion testing, laboratory processes and transfusion in the day transfusion ward for OP, or the IP haematology ward. All steps are process-mapped, individually timed (minimum 3 timings per step) and all consumables, equipment and staffing involved in each step recorded.

Results: 31 process maps (22 laboratory, 8 clinical including phlebotomy, 1 pharmacy) describing >600 individual activities were developed. Table 1 shows the processes, steps involved, decision points, time taken and resources required for transfusing 1 RBC unit. For a routine uncomplicated patient (i.e. no alloantibodies or special product requirements, automated group & screen, computer crossmatch), the entire process takes an average total of mins for 440 mins for OP (478 mins in OP ward A, 401 mins in OP ward B) and 303 mins for IP (Table 2). For complex patients with more testing or product requirements, the process takes up to 918 mins for OP and 743 mins for IP (Table 2). Laboratory processes are the most complex, with up to 53 decision points and taking 87% of total time (647/743 mins) for complex IP and up to 77% (647/841 mins) for complex OP. For routine patients, laboratory processes take 68% (207/303 mins) total time for IP and up to 52% (207/401 mins) total time for OP.

Summary / Conclusions: This is the first detailed real-world study of the complex multistep, multidisciplinary processes required for supporting RBC transfusion in patients with MDS. Cost analysis of this entire process is currently underway and will help in future resource planning. Detailing the many steps and decision points can also help identify potential 'weak links in the chain' in these processes, and be used to improve transfusion procedural safety.

P736 - Table 1 Steps for transfusing 1 RBC unit

Process	Steps, N	Decision points, N	Time(mins), Mean (range)	N of staff (S) N of consumables/ equipment (C)
OP Phlebotomy	36	1	18 (10-26)	S:2 C:47
IP Phlebotomy	32	2	33 (31-37)	S:1 C:35
Laboratory (same for OP & IP)	Routine* patient:165 Complex# patient:up to 417	Routine*: 20 Complex#:53	Routine*:207(189-217) Complex#:647(602-793)	S:3 C:103
RBC administration, OP ward A^	105	38	253 (194-353)	S:9 C:156
RBC administration, OP ward B^	86	36	176 (146-215)	S:6 C:77
RBC administration, IP	38	23	63 (52-77)	S:4 C:47

IP, Inpatient; OP, Outpatient.

* Routine: no alloantibodies, no special blood products required #Complex: alloantibodies and/or special blood products required.

^Outpatient transfusions may occur in either ward A or ward B.

P736 - Table 2: Mean total time (phlebotomy+laboratory+RBC administration), in minutes

	OP ward A^	OP ward B^	IP
Routine patient*	478	401	303
Complex patient#	918	841	743

* Routine: no alloantibodies, no special blood products required

#Complex: alloantibodies and/or special blood products required.

^Outpatient transfusions may occur in either ward A or ward B.

P737 | Development of a scoring system using laboratory parameters and transfusion outcome to predict the severity of Autoimmune Haemolytic Anemia (AIHA)

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Background: Understanding severity of hemolysis in AIHA aid in treating and monitoring disease activity. Previous reports have classified severity into moderate and severe based on laboratory parameters irrespective of the outcome measures. Not all the laboratory parameters are available in resource poor settings. Inclusion of clinical aspect of disease with easily available laboratory parameters will improve the usefulness of severity scoring in resource constraint settings and draw actual picture of disease in total.

Aims: The study was aimed to assess if a scoring system including both laboratory parameters and transfusion outcome would help predict the severity of DAT positive AIHA.

Methods: The study was conducted in Department of Transfusion Medicine of a tertiary care center for period of two years after Institute ethical committee approval. DAT positive AIHA patients were

included. Poly-specific DAT (anti-IgG, C3d) performed by conventional tube test (CTT) and gel method. Values of hemoglobin, bilirubin, lactate dehydrogenase (LDH) and transfusion details were collected from hospital information management and blood bank management system software respectively. Monospecific DAT and IgG DAT dilution (anti-IgG 1:10, 1:30, 1:100, 1:300, 1:1000) were performed using gel method (Bio-Rad, Switzerland). DAT positive AIHA patients for whom IgG DAT dilution was not performed were excluded from the study. Data analyzed using Microsoft Excel. The parameters used for scoring include hemoglobin (scored from 0 to 4), either bilirubin or LDH (scored from 0 to 2), blood transfusion (scored from 0 to 3) and DAT strength by CTT (scored from 0 to 2). The cumulative score was calculated, and the maximum score given was 11. The severity of hemolysis in AIHA was classified as no active hemolysis, mild, moderate and severe based on the cumulative score.

Results: 45 IgG DAT positive AIHA patients were included. The median age was 30 years [Inter quartile range (IQR)1 = 19, IQR3 = 53] and 60% were females. Mean hemoglobin was 6.8 ± 9.3 (standard deviation) g/dL and 42% had it <6g/dL. The median total bilirubin and LDH levels were 2.3mg/dL (IQR1 = 1.4, IQR3 = 4.2) and 538 IU/L (IQR1 = 352, IQR3 = 1127) respectively. 26 patients received at least one blood transfusion, 18 required it multiple times. The median intertransfusion interval was 37 hours (IQR1 = 24, IQR3 = 52) and 13 patients received multiple transfusions within 48 hours of hospital admission. IgG DAT titer of 10, 30, 100, 300 and 1000 were present in 11 (24%), 10 (22%), 5 (11%), 3 (7%) and 16 (36%) patients respectively. The scoring system classified 8 patients as mild, 16 as moderate and 21 as severe AIHA. A positive correlation (Spearman rank coefficient of 0.7) of the score was observed with the IgG DAT titer.

Summary / Conclusions: The study tried to address the limitations of previous scoring systems by incorporating simple clinical, hematological, biochemical and serological aspects of the disease. The scoring

system could be easily adopted by most centers even in resource limited settings to assess severity of AIHA and the need for aggressive treatment and monitoring. This study is limited by relatively small sample size and usefulness only in IgG DAT positive AIHA patients.

P738 | CAR-T cell therapy vs ASCT—impact for a transfusion service

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Background: The standard treatment for Non-Hodgkin lymphoma who responds to salvage treatment after relapse is chemotherapy plus consolidation with autologous stem cell transplant (ASCT). Recently, Chimeric antigen receptor (CAR)-T-cell therapy has been proven to be beneficial for this disease. Both therapeutics strategies increase the blood support after the lymphodepletion (LD) or conditioning regimen (CR).

Aims: The purpose of the current study was to evaluate the cumulative transfusion burden in patients treated with CART or ASCT until haematologic recovery.

Methods: We performed a retrospective study of the clinical characteristics and transfusions requirements of the patients who received the commercial CAR T Cell axicabtagene ciloleucel (Yescarta) or ASCT between March 2019 and October 2023 in one center. Patients included were previously diagnosed with diffuse large B cell lymphoma, follicular lymphoma and primary mediastinal large B-cell lymphoma.

Results: A total of 88 patients (51% male) who received ASCT ($n = 36$) or CART ($n = 52$) were included. Median age was 60 years (range 19-75) for CART cohort and 53 years (range 28-71) for ASCT cohort. Median of previous therapies before CART was 3.5 (range 2-7) and 2 (range 1-6) for ASCT. The LD consisted in the combination of fludarabine and cyclophosphamide, while the CR combined carmustine, etoposide, cytarabine, and melphalan (BEAM). In the CART cohort, before the infusion, six patients (11.5%) had anaemia grade ≥ 3 and 4 (7.7%) thrombocytopenia grade ≥ 3 . No patients had severe anaemia or thrombocytopenia before ASCT. The median days until haemoglobin ≥ 8 g/dL and platelets $>20 \times 10^9$ /L for CART and ASCT cohort was 28 (IQR 6-45) vs 10 (IQR 8.5-23.2) and 44 (IQR 37-60) vs 13 (IQR 9-15), respectively. Overall, the more common ABO and Rh group was A Rh(D)+ (44.3%) and O Rh(D)+ (42.1%). A total of 715 blood components were transfused: 300 packed red blood cells (pRBC) units, 388 pooled platelets (PP), 17 fresh frozen plasmas (FFP)

and 10 COVID-19 convalescent plasmas. The cumulative transfusion burden in CART cohort was higher in comparison to ASCT cohort for pRBC [4 (range 1-52) vs 2 (range 1-10)] and PP [4 (range 1-28) vs 2 (range 1-16)]. Ten patients (19.2%) in CART cohort needed ≥ 10 pRBC and 8 \geq PP, five of them died of progression disease. In the first 30 days after infusion, transfusion of blood components was 42% and 91% in the CART and ASCT cohorts, respectively. One CART patient received 12 FFP (70.6%) due to disseminated intravascular coagulation secondary to shock septic. ASCT and CART represented 1% and 2.9% of the total irradiated blood components transfused in our center in the same period.

Summary / Conclusions: Transfusion is an important support for CAR-T and ASCT. Patients who received CART therapy required higher blood support and for a longer period, probable because they were more heavily treated.

P739 | Transfusion requests and PRBC deposition analysis

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Background: The Blood Bank of La Rioja, based at San Pedro Hospital (Logroño, La Rioja), caters to blood component demands from regional hospitals. Over 80% of Packed Red Blood Cells (PRBC) are transfused there. Despite declining donation rates, PRBC supply, especially O negative type, has risen. This calls for scrutinizing red blood cell transfusion requests and storage. La Rioja has seen decreasing blood donations, yet certain components, notably O negative PRBC, have increased.

Aims: Analyzing red blood cell transfusion requests and storage is crucial in this context.

Methods: We have analysed the distribution of PRBC in San Pedro Hospital over the last five years, taking into account the ABO and Rh group. An exhaustive examination of PRBC distribution within San Pedro Hospital over the preceding five years has been conducted, with due consideration given to the ABO and Rh group classifications. Similarly, the number and percentage of PRBC discarded at the San Pedro Hospital in relation to the number of PRBC obtained by whole blood donation and apheresis has also been reviewed during these years. In the same way, the most frequent reasons for which PRBC are withdrawn have been collected. Simultaneously, a review has been undertaken to scrutinize the number and percentage of discarded PRBC at San Pedro Hospital concerning the total PRBC obtained through both whole blood donation and apheresis methodologies during this temporal span. Furthermore, a compilation of the predominant reasons necessitating the withdrawal of PRBC has been meticulously documented. The data has been obtained from the e-Progesa and e-BDI Plus programs.

Results: The evolution of the distribution of PRBC according to ABO blood group and Rh from 2019 to 2023 and the observed variation is reflected in the following table:

P739 - Table 1.

2019		2020	2021	2022	2023	% Variation
PRBC Requests	7495	7792	7511	7751	7683	+2.44
O+	2596	2593	2586	2681	2669	+2.73
O-	921	951	979	1022	1109	+16.95
A+	2489	2598	2502	2630	2463	-1.05
A-	655	782	745	736	707	+7.35
B+	499	552	406	364	423	-17.96
B-	104	118	109	117	103	-0.97
AB+	188	167	165	167	163	-15.33
AB-	43	31	19	34	46	+6.52

P739 - Table 2.

2019	2020	2021	2022	2023	% Variation
PRBC donated 10156	10436	9810	9886	9254	
PRBC discarded 883	639	694	498	317	
(8.69%)	(6.12%)	(7.07%)	(5.05%)	(3.42%)	5.27%
The main reasons for discarding PRBC					
Expiration date 604	395	444	258	90	
(68%)	(62%)	(64%)	(52%)	(28%)	
Loss of storage 80	71	70	63	53	
temperature (9%)	(11%)	(10%)	(13%)	(17%)	

The number and percentage of PRBC discarded with respect to the number of red blood cell concentrates obtained and the main reasons for discarding PRBC throughout the years studied will be reflected below:

Summary / Conclusions: (1). In the analyzed five-year period, PRBC demand rose by 2.44%, notably for O- type, with a 16.95% distribution increase. (2). Discarded PRBC concentrates decreased by 5.27% from 2019 to 2023, indicating improved stock management. (3). Mainly, PRBCs are discarded due to expiry, which notably decreased in the last year. (4). Loss of storage temperature is the second cause, often due to returned red blood cells. Enhancing employee training handling donation bags is crucial for improvement.

P740 | Study of circulating erythrocyte microparticles in major thalassemia patients

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Background: Circulating microparticles derived from pathologic red blood cells notably in thalassemic patients, have been associated with

multiple physiopathological conditions such as oxidative stress and hypercoagulability state. Major thalassemic patients on regular transfusion program may exhibit a lower incidence of thromboembolic complications. This may be attributed to a decrease in microparticle levels due to repeated transfusions.

Aims: The aim of the study was to investigate circulating erythrocyte microparticles (CEMPs) level before and after transfusion in regularly transfused major thalassemic patients.

Methods: 38 β -thalassemia major adults patients and 30 age and gender matched normal controls were enrolled in the study. Blood samples were collected from patients before and after transfusion. Platelet-free Plasma was obtained following a specific centrifugation protocol at room temperature. Samples were aliquoted and stored at -80°C until use. Circulating erythrocyte microparticles enumeration was performed by high sensitivity flow cytometry. As for cell blood count (CBC), it was conducted on the patient group, prior to transfusion.

Results: The mean age of major β -thalassemia patients was 27 years [19 - 36]. The sex ratio was 1.62. All patients were splenectomized and regularly transfused. The mean values of CBC parameters appeared as follows: Hemoglobin (Hb) = 7.8 g/dL [5.3-10.3], Mean corpuscular volume (MCV) = 82.7 fL [77.6-88.8], Mean corpuscular hemoglobin concentration (MCHC) 33.7 g/dL [31.7-35.8], white blood cell (WBC) = 16.07 /mm³ [3.7-28.11], platelets (PLQ) = 655*10³/

mm³ [24-1088]. The comparison between circulating erythrocyte microparticles levels obtained in patients and controls groups was assessed by means of the Mann-Whitney U test and revealed higher levels in the patient group (CEMPs median = 9.1) compared to the control group (CEMPs median = 3.5) ($p = 0.03$). In addition, the analysis of circulating erythrocyte microparticle levels in β -thalassemia patients after transfusion indicate a decrease compared to pre-transfusion levels (CEMPs median = 4.3) with a statistically significant difference ($p = 0.04$).

Summary / Conclusions: circulating erythrocyte microparticles level in regularly transfused major β thalassemia patients was found to be higher than the normal range. Their level exhibit a significant decline post-transfusion.

P741 | Profile and performance of a transfusion service in a liver transplant program—preliminary data from a 4-year experience of Uruguay's national center

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Background: Liver Transplant Programs demand great resources from transfusion services assisting the institutions that host this type of solid organ programs. They present high transfusion requirements, a critical hemovigilance program and a fluid communication between hospital teams. It is key to know the transfusion profile of these patients and to understand how the transfusion service can be better prepared to support them in the most efficient way.

Aims: To describe the use of blood components regarding a national Liver Transplant Program. A preliminary performance of our transfusion service blood reserve protocol is included.

Methods: We performed a retrospective study of the period 2020-2023, that includes 80 liver transplant (LT) procedures at our center. For each case, the study includes parameters from the day of the transplant to an immediate follow-up of 7 days. We also described our transfusion service protocol of blood reserves for LT procedures, and analyze at what rate our threshold reserves are surpassed. Descriptive statistical analysis and Mann-Whitney tests were performed with SPSS 29.0.

Results: Out of 80 patients, 51 (63.75%) were male and 29 (36.25%) female. All received an orthotopic LT. The primary liver pathologies presented by the patients were: 19 (23.75%) non-alcoholic fatty liver disease (NAFLD), 16 (20%) alcoholic cirrhosis, 10 (12.5%) viral cirrhosis, 9 (11.25%) autoimmune cirrhosis and 17 (21.25%) as other causes, which include primary sclerosing cholangitis, hemochromatosis, autosomal recessive polycystic kidney disease and neoplasia. In those 80 LT procedures, 9 (11.25%) liver retransplantation procedures are included. All alcoholic and viral cirrhosis LT patients were male, and 8 of 9 autoimmune cirrhosis were female. 2 patients (0.025%) died during the immediate 7 day follow-up. 17 (21.25%) LT patients did not receive any blood component support during the procedure

(or the 7 days immediate follow-up). Overall, patients received a mean of 5.15 red blood cell (RBC) concentrates, 2.01 fresh frozen plasma (FFP) units, 1.11 platelet concentrates (PC) and 1.98 cryoprecipitates. There were no transfusion rate differences between male and female patients, overall and in each blood component subgroup (with a significance level of 0.05). Among primary liver pathologies, autoimmune cirrhosis patients required less RBC concentrates against NAFLD patients ($p = 0.022$), but overall these groups did not present statistical differences. For each LT patient, by protocol our center has a reserve stock of 10 units of RBC concentrates, 10 units of FFP, 2 therapeutic doses of PC and 10 cryoprecipitates. Overall, 30% (24) of LT blood usage exceeded at least one of the blood components from our protocol stock, with cryoprecipitates (10% of LT) being the most frequent transfused component that surpassed our initial stock reserved.

Summary / Conclusions: While analyzing our preliminary data from a 4-year experience at our center, we found that most LT patients are male (63.75%), and the most common cause of LT is NAFLD (23.75%) followed by alcoholic cirrhosis (20%), viral cirrhosis (12.5%) and autoimmune cirrhosis (11.24%). All the alcoholic and viral cirrhosis patients were male, and all but one of the autoimmune group were female. Autoimmune patients required less RBC concentrates than NAFLD patients. We matched our LT patients profile with our blood component protocol and found that in 30% of LT at least one component reaches or surpasses the reserved stock.

P742 | Granulocyte transfusions—an experience from a Brazilian cancer center

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Background: Infection associated with prolonged neutropenia caused by myelotoxic chemotherapy is still a major cause of extended hospitalization and mortality in cancer patients. Granulocyte transfusions (GTx) have been used to treat refractory infection in the severe neutropenic for over 50 years, yet its clinical efficacy remains controversial.

Aims: This study aims to analyze the characteristics of granulocyte recipients, collection yield and survival rates at AC Camargo Cancer Center, a Brazilian Oncological institution.

Methods: This is a retrospective study based on chart review of patients who received GTx from January 2013 to January 2024. We collected data on demographic characteristics, underlying disease and clinical outcomes (death and hospital discharge), as well as data on apheresis collection. Granulocyte donors had to meet same qualification criteria as whole blood donors and received mobilization with dexamethasone (8-10mg PO) and single-dose filgrastim 300mcg on the evening before collection (approximately 15 h before). Apheresis was performed using a continuous-flow cell separator (Spectra Optia Apheresis System, Terumo BCT), processing 6-7L of blood, preferably utilizing sedimenting agent.

Results: Fifty five GTx were administered on 10 adults and 5 pediatric patients; median age was 23 years old (range 3–52) and 53% were female. Acute leukemia (73%) and lymphoma (20%) were the predominant underlying diseases. Three patients had undergone allogeneic stem cell transplant and one patient presented with prolonged cytopenias after CAR T cell infusion. All patients had evidence of fungal infection, with 53% displaying both bacterial and fungal etiology. Skin (40%) and pulmonary (40%) sites were the most common primary locations; nine patients (60%) had more than one site of infection. Median granulocyte dose administered was 5.03×10^{10} (1.16 – 10.88), with 58% of GTx delivering an optimal dose of $\geq 0.6 \times 10^9$ granulocytes/kg. Six apheresis were performed without sedimenting agent due to a nationwide shortage and median yield was lower (1.33×10^{10} ; 1.16 – 1.81). Despite that, all collections yielded above the minimum requirement of 1×10^{10} granulocytes. Patients received a median of 3 (1 – 10) GTx. Pre-transfusion leukocyte counting (WBC) median was $360/\text{mm}^3$ (10 – 5720), absolute neutrophil counting (ANC) was not available in 62% of those due to severe leukopenia. Median post-WBC was $1490/\text{mm}^3$ (70 – 9901) and median post-ANC $1267/\text{mm}^3$ (0 – 9800). Sixty percent of these GTx resulted in a post-ANC $\geq 500/\text{mm}^3$. Pre-medication with antihistaminic and antipyretic was systematically administered and infusions were closely monitored by our Transfusion Practitioners and medical staff. We had only one potential adverse event related to GTx (oxygen saturation fluctuation) but it was mild and infusion was resumed after supportive measures. Mortality after 30 days of the first GTx was 47% and overall mortality was 67%, all within the same hospitalization period of the GTx.

Summary / Conclusions: While clinical efficacy evidence remains inconclusive, considering the high morbimortality of patients with refractory infections and severe neutropenia, GTx may be an adjuvant therapeutic option. Collection from donors after stimulating agents provided adequate dose and infusions were overall well tolerated. Due to limitations of sample size and the retrospective nature of our study we were not able to identify subgroups of patients that could have benefited from GTx but it may be a promising approach in the future.

P743 | 3-Factor versus 4-Factor prothrombin complex concentrates usage—a tertiary cardiac centre experience

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Background: Prothrombin complex concentrates available as 3-factor prothrombin complex concentrate (PCC) or 4-factor PCC. The 3-factor PCC has been available in Malaysia for the past 20 years since the fractionation program started in Malaysia. It is derived from our own plasma donors through fractionation process. Earlier it was widely used as treatment and prophylaxis in bleeding patients with single or multiple congenital deficiency factors IX, II or X when purified specific coagulation factor product is not available while 4-factor

PCC purchase from commercial companies and this cause huge financial implication between those two. However, nowadays patients benefit greater with the availability of purified specific coagulation factor. Hence, both PCC can be used for treatment and perioperative prophylaxis of bleeding in acquired deficiency of prothrombin complex factors, such as deficiency caused by treatment with vitamin K antagonists, when rapid correction of the deficiency is required.

Aims: This study was conducted to determine the use 3-factor PCC and 4-factor PCC in Hospital Sultan Idris Shah, Serdang and analyse its efficacy.

Methods: Retrospective study of all cases used 3-factor PCC and 4-factor PCC for treatment and the data collection was obtained from patient medical record, pharmacy information system and Lab Information System (LIS).

Results: A total of 94 patients were given 3-factor PCC and 75 patients 4-factor PCC from January 2022 to May 2023. Majority of bleeding secondary to overwarfarinization were given 3-factor PCC 57 cases (82.6%) while only 12 cases (17.4%) were given 4-factor PCC as reversal agent. Inversely in massive bleeding intraoperatively cases only 25.5% (21 cases) used 3-factor PCC meanwhile 74.4% (61 cases) used 4-factor PCC as adjunct therapy to control bleeding. In case of elevated INR and planned for urgent procedures only 1 case used 4-factor PCC and 15 cases treated with 3-factor PCC before going to any invasive procedure. Only 1 case in each PCC groups were used to treat bleeding due to overdose of Non-Vitamin K Antagonists Oral Anticoagulant (NOAC). The mean INR of 4.0 (range 0.9–16.0 in 3-factor PCC and range of 1–17.5 in 4-factor PCC group) before treatment and only one dose of 3-factor PCC needed in 77.7% of cases to achieve homeostasis meanwhile 98.7% in 4-factor PCC. Eighteen cases needed second doses of 3-factor PCC, meanwhile only 1 case in 4-factor PCC. Three cases of massive intraoperative haemorrhage need up to the 3rd doses of 3-factor PCC to achieve homeostasis. 3-factor PCC were given concurrently with blood products in 13% (12 cases), 65% (49 cases) in 4-factor PCC group and all of it were massive bleeding intraoperatively. Overall, mean doses of 3-factor PCC were 30iu/kg (range 10 – 50 iu/kg) to achieve homeostasis and 20iu/kg in 4-factor PCC (10 – 25 iu/kg).

Summary / Conclusions: The study shows, in cases due to overwarfarinization warfarin, lower doses of 3-factor PCC (10 IU/kg – 30 IU/kg) and 4-factor PCC (10 – 25 iu/kg), giving the factors concentrate without FFP were effective. Majority of 4-factor PCC were also used in massive intraoperative bleeding and it is effective to reduce usage of blood products.

P744 | Night-time transfusions—a single-center transfusion service perspective

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Background: In a hospital, limited human resources during nighttime may impact transfusion safety for patients receiving blood derivatives. Guidelines recommend avoiding non-urgent transfusions outside regular hours for patient safety and to respect their rest.

Aims: Evaluate the percentage of transfusions performed during nighttime hours (10 pm – 8 am) at our hospital. Evaluate within a representative sample of nocturnal transfusions, those that could have been avoided.

Methods: This retrospective observational study took place at the University Hospital Joan XXIII in Tarragona. Over six months in 2023 (June to November), we randomly collected data on transfusions initiated during nighttime hours, in total 78 samples. We recorded transfusion urgency, patient diagnosis, time of lab result validation, time of request, and time of transfusion.

Results: In 2023, 18.6% of all transfusions occurred at night (2085 out of totally 11218). We analyzed 78 transfusions, including 7 platelet pools and 71 red blood cell concentrates (RBCs). Urgency levels for RBCs were 2 immediate, 34 urgent, 33 routine, 1 scheduled, and 1 pre-surgery reserve. Among immediate cases, clinical urgency precluded delaying transfusion. All screening tests in the lab were always negative. For urgent requests, 50% occurred within 3 hours of lab result validation, 23.5% within 3-6 h, and 26.5% after 6 h. The average time from request to transfusion was 2 h, except for one case delayed due to patient fever. Routine requests, most avoidable, were divided as follows; 60% (20 requests) were from inpatient units, and 40% (13 requests) were from Emergency/Operating Room. For routine inpatient requests, 25% were within 3 h, 20% within 3-6 h, and 55% exceeded 6 h from lab result to request. From request to transfusion, 25% (5 requests) were delayed by 4-5 h, an acceptable timeframe for urgent requests. 45% were unavoidable night transfusions due to over 6 h between lab result and request. Regarding routine Emergency Room requests, 85% were within 3 h, 7.5% (1 request) within 3-6 h, and 7.5% (1 request) after 6 h. 77% percent (10 of 13 requests) were unavoidable due to late lab results when patients entered the ER.

Summary / Conclusions: It's difficult to assess if night transfusions can be avoided. Most delays are from delays between lab results and inpatient transfusion requests, both routine and urgent. For urgent transfusions, around 50% could have been avoided due to delays between haemoglobin results and transfusion requests. Further discussion is needed on whether these requests should have been categorized as routine instead of urgent. In routine inpatient transfusions, a debate arises as 50% exceeded 6 h between lab result and request. Investigating the reasons for such delays is crucial to determine potential reductions. Transfusion personnel face circumstances causing delays, like; fever or undergoing tests, leading to nighttime transfusions. Another debatable point is whether, in routine inpatient transfusions delayed to the night shift, it would be ethical to prioritize rest and transfuse the next morning, considering potential adverse effects. In conclusion, it's difficult to assess whether a regular nocturnal transfusion could be avoided due to influencing factors, some of which are beyond modification.

P745 | A clinical audit on night-time red cell transfusions at National Hospital of Sri Lanka

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Background: Nighttime red cell transfusions carry a risk to the recipients due to inadequate monitoring and difficulty in detecting and managing transfusion related adverse events as there is a lower number of staff available in the night when compared to daytime. Additionally, overnight transfusions can disturb the patients' sleep as well. Therefore, nighttime transfusions are not recommended by the transfusion services unless those are clinically essential.

Aims: The main objective of this audit is to get the details of nighttime red cell transfusions of the hospital and to determine whether the indications for those transfusions are justifiable.

Methods: The details of the patients who received red cell transfusions from 1900 hours to 0700 hours, each day are collected over a period of one month using the red cell issue registry, red cell request form and patients' clinical records. The clinical indications for transfusions which were used in the National Health Service (NHS) audit on overnight transfusions in 2008, were modified into three categories according to the local practice.

Results: A total number of 1992 red cell units were transfused during the study period. Out of those, 373 (18.7%) red cell units were transfused during the nighttime. 54.9% of the nighttime issues were received by males and the average age of the patients who received nighttime transfusions was 58.1 years while 63.3% of those were given to patients >50 years. Patients from medical and surgical specialties received 48.8% and 25.2% of nighttime transfusions respectively while 7.7% units were transfused at operation theaters. 18% of the total nighttime transfusions were received by ICU patients. Only 64.4% of units were transfused to the patients with acute clinical need with active bleeding or haemolysis and for patients with symptomatic anaemia (Category 1), while 8.1% of units were given to the patients with less acute clinical need (Category 2) who were transfused to optimize haemoglobin prior to a procedure or during haemodialysis. 27.3% of total nighttime transfusions were given to patients without any acute or subacute clinical need, in view of correcting the haemoglobin (Category 3). No acute transfusion related adverse events were reported for above 373 red cell transfusion episodes.

Summary / Conclusions: According to this audit, more than 1/3 of transfusions were given to patients without an acute clinical need and these can be minimized by educating the clinical teams regarding the risks associated with these transfusions. We recommend introducing policies to minimize unnecessary transfusions at night through the hospital transfusion committee and introduction of special patient consent for nighttime transfusions. Re-audit should be conducted after introducing these policies to see the progress.

P746 | Is the administration of anti-D prophylaxis in transfused patients with Rh-incompatible platelets justified?A Moya¹, A Sánchez¹, S Sánchez¹, B Pérez De Camino¹, R Capellan¹, G Castellanos¹, P Llamas¹¹Blood Bank, Hospital Universitario Fundación Jiménez Díaz, Madrid, Spain

Background: Standard practice in our setting for platelet transfusion is to respect the ABO group and as far as possible the Rh group, although the lack of availability of Rh-negative components limits our approach. The risk of alloimmunization in case of transfusion of Rh-incompatible platelets is lower than with transfusion of red cells but it is not exempt, which could be a future complication for the immuno-haematological studies and increase patient morbidity. In the specific case of D antigen, we rely on the profilactic administration of anti-D immunoglobulins to Rh-negative patients who are to receive Rh-positive platelets to avoid the previously described risk. The risk vs benefit profile and cost of this measure have been questioned on numerous occasions. Therefore, we believe it is interesting to evaluate the efficacy of this practice with data from our centre.

Aims: The aim of this study is to compare the efficacy of anti-D immunoglobulin administration in Rh-negative patients who received Rh-positive platelets versus those who did not receive such prophylaxis.

Methods: A single-centre, descriptive, observational, retrospective, descriptive study was carried out to collect data on Rh-negative patients with diverse pathologies who have been transfused with Rh-positive platelets. The sample comes from the Hospital Universitario Fundación Jiménez Díaz in Madrid during the period between 2020 and 2023. Patients with D-antibody prior to the study, who had changed their group to Rh-positive, who had undergone a massive transfusion of Rh-positive red blood cells and those for whom no sample was available after the date of the last administration have been excluded.

Results: The sample consisted of 124 patients, of whom 118 (95.16%) did not receive immunoglobulin administration and 6 (4.84%) did it. Of the 118 patients who did not receive anti-D prophylaxis, 47 subjects (39.83%) were lost to follow-up as no results were available after administration of anti-D prophylaxis. In addition, all cases in which prophylaxis was administered (6) were also lost to follow-up. Alloimmunization occurred in 7.04% (5 patients) of the subjects who did not receive anti-D prophylaxis (71).

Summary / Conclusions: Analyzing the results obtained, it can be noted that the risk of alloimmunization after transfusion of Rh-incompatible platelets and without administration of prophylaxis is lower than expected. The loss of patients follow-up who received anti-D gammaglobulin unfortunately limits the analysis of our data, and it did not allow the achievement of significant results. Given these findings, and taking into account the increasing transfusion requirement for platelets in tertiary hospitals, the difficulty in recruiting Rh-negative donors and the increasingly high costs of administering anti-D serum, it is necessary to extend the study to expand our sample and make it more representative in order to reach more detailed and enlightening conclusions.

P747 | An audit on outpatient and inpatient platelet transfusion practice before and after implementation of platelet transfusion guidelinesB Lamas¹, A Gálvez¹, C Sopeña¹, D Martínez¹, P Asensi¹, Gómez¹, M Santiago¹, P Solves¹¹Haematology, Hospital Universitari i Politècnic La Fe, Valencia, Spain

Background: Use of platelet transfusion has increased during last years. Taken into account that it is a scarce resource, blood banks must assure the appropriate utilization of platelets. Establishment and diffusion of PLT transfusion guidelines is the first step for clinicians to be trained in their use. Audits are a skill to monitor the adherence to the current guidelines.

Aims: Our objective was to compare platelet transfusion practise before and after of implementation a PLT transfusion guidelines in the Haematology Department of a Tertiary care Hospital.

Methods: After performing an audit on platelet transfusion, we established a platelet transfusion guideline in the haematology Department. Once the new document was spread among haematologists, we performed a second audit to monitor the adherence to the updated guidelines. Criteria for considering appropriateness of PLT transfusions were the same for both audits. In the prophylaxis setting a PLT threshold of <10000/μl was used and 20000/μl in case of risk factor for bleeding was present. For treatment purpose (bleeding grade 2 or greater on the modified WHO scale), PLT threshold was considered <50000/μl for severe bleeding and <30000/μl for mild non-life-threatening bleeding. For hospitalized stable patients receiving an autologous stem cell transplant no prophylactic transfusions were considered, just therapeutic. The audits consisted of reviewing the transfusion and clinical chart of patients to collect platelet counts before transfusion and clinical conditions related to the transfusion episode.

Results: We collected data from 255 transfusion episodes (AUD 1) and compared to 137 episodes collected after the implementation of the PLT transfusion guidelines (AUD 2). Results are shown in the table below. Prophylaxis was the main indication of platelet transfusion in both audits. Media ± SD of platelet counts in the prophylaxis indication were 12693 ± 8044 in AUD 1 and 13150 ± 10040 in AUD2 ($p = ns$). In the treatment indication, platelet counts were 32000 ± 11039 in AUD 1 and 21965 ± 14651 in AUD2 ($p = ns$). Overall percentage of adjustment to the guidelines was higher in AUD2 (64.2%) than in AUD1 (49%). Overall grade of appropriateness was higher for inpatients (58%) than for outpatients (48%), $p = 0.009$. When compared

P747 - Table 1.

	Audit 1	Audit 2	p value
number, n	255	137	
Prophylaxis/treatment	238/17	107/30	< 0.001
Inpatient/Outpatient	150/105	76/61	0.522
Adjusted to guidelines/not adjusted/not assessable	125/98/32	88/42/7	0.006

results from AUD2 to AUD1, adherence to guidelines improved in both the inpatient setting (67% vs 54.6%), and in the outpatient setting (60.6% vs 40.9%).

Summary / Conclusions: Available transfusion guidelines support clinicians to perform an evidence-based platelet transfusion. While it is true that current PLT transfusion guidelines are inconsistent and in some aspects variable among them, they have to be the base to collect more evidence. Since audits show a high degree of non-compliance with current recommendations, Blood banks must work on implementation guidelines to assure an adequate use of platelets. Adherence to guidelines will provide more homogeneous data, allowing a more precise analysis of the consequences of applying them, so we can continue improving the indications.

P748 | Evaluation of the provision of crossmatch compatible platelets for haematology patients who have acquired non-immune refractoriness

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Background: Platelet transfusions are vital to the ongoing management of haematology patients, however, many patients become refractory leading to increased use just to prevent bleeding, but their platelet count never seems to increase. Non-immune platelet refractoriness is the most frequent cause and treatment is often ineffective and a complicated challenge. HLA (human leucocyte antigen) matching and platelet crossmatching has led to increased costs and difficulties in management with significant delays in treatment.

Aims: To evaluate the provision of crossmatch compatible platelets for haematology patients who have acquired non-immune refractoriness using expression of P-selectin and complement (C1q) on platelets.

Methods: Platelet rich plasma (PRP) was prepared pre and post platelet transfusion from 8 haematology patients who had become refractory post stem cell transplant. A total of 34 apheresis and 47 pooled platelet donations were transfused and sampled so that crossmatching could be performed retrospectively on the patients. Both apheresis and pooled platelet donations were compared in this study to see if there were any significant differences between them which may affect the crossmatch result and the increment when transfused to haematology patients. Flow cytometry was used to demonstrate the expression of CD62P and C1q on platelets as markers of platelet activation and complement. The platelet increments after each donation were then studied to see if whether transfusing a compatible platelet donation gave a better increment than transfusing an incompatible one. A transfusion was deemed to be successful where the CCI (corrected count increment) was >5000 or the platelet count went over $10 \times 10^9/L$.

Results: The CD62P and C1q values in apheresis platelets were significantly higher than in pooled platelets, normal controls, and patients and this could be due to the apheresis machines where

centrifugation and filtration involves platelets coming into close contact with membranes that can potentially cause platelet activation. The elevated levels of CD62P and/or C1q in platelet donations did not give incompatible crossmatch results – there was a mixture of compatible and incompatible results with normal and elevated levels of these markers meaning that these alone were not the reason for the incompatibility. Platelet transfusions in patients did not affect the levels of CD62P or C1q on their platelets, which were already higher than normal controls, possibly due to some other underlying mechanism. Sometimes a platelet transfusion led to an increase or a decrease in both or one of these markers – there was no specific pattern. A successful transfusion could not be attributed to the provision of compatible platelets, since the percentage of successful transfusions were similar between the compatible and incompatible transfusion groups.

Summary / Conclusions: There was no evidence to suggest that crossmatched platelets would provide a better platelet increment than random donor platelets. Comparison of the two platelet manufacture methods showed apheresis platelets to have higher levels of CD62P and C1q on them, however, this did not account for the poor increments or incompatible crossmatches. There was no correlation between increased values of CD62P or C1q and a successful/unsuccessful crossmatch. Clearly more research is needed in the field of non-immune platelet refractoriness.

P749 | Abstract withdrawn

P750 | Clinical significance, feasibility, and economy of Rh antigens compatible red blood cells transfusion

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Background: Developed countries have established haemovigilance systems to assess the risk of blood transfusions, monitor irregular red blood types antibodies of patients and transfuse ABO, Rh, and Kell antigens matched red blood cells (RBCs). The Chinese haemovigilance network (CHN) was used to analyze and evaluate adverse blood transfusion reactions in 2018, but compatible transfusions with antigens other than ABO and Rh D are still not available. In China, few studies reported the clinical significance and economic cost of Rh blood group compatible transfusions.

Aims: The irregular antibodies of Rh blood group are the most common in Chinese individuals. Here we study the clinical efficacy of Rh (C, c, E, e) compatible transfusion, as well as consideration of the feasibility and economic cost of Rh compatible transfusion in current medical insurance policies.

Methods: The study included 44,530 inpatients (5499 transfused patients) and 20,735 donors from July 2018 to June 2020, and 42,153 inpatients (5287 transfused patients) and 19,836 donors from July 2020 to June 2022. Irregular antibodies were screened and

identified. In the second period, ABO and Rh phenotypes were tested, Rh antigens compatible RBCs were transfused, length of hospitalization stays (LOS), blood transfusion costs and the average use of RBCs were also analyzed.

Results: The total ratio of irregular antibody was 1.72% (186/10,786) from July 2018 to June 2022, the antibody frequency of Rh system was 43.50%, anti-Mi^a was 21.08%. After Rh phenotyping, the total probability of Rh compatible transfusion was 92.43% (18,355/19,859). In all 186 patients with irregular antibodies, 60 patients are newly developed in our hospital. There is no new development of Rh system antibody after Rh D, C, c, E, e antigens compatible transfusion from July 2020 to June 2022. The LOS and the mean RBC cost decreased. The mean cross-match experimental cost increased significantly, but this increase accounted for 3.63% and 8.27% of the increased hospital costs for hospitalized and transfused patients, respectively. The average use of RBCs for hematological patients was reduced by 2.60%.

Summary / Conclusions: The Rh antigen phenotype compatibility between donors and patients was high enough to transfuse Rh matched RBCs for more than 90% of patients. Rh compatible transfusion can diminish new irregular antibodies and save RBCs. The increased cost of Rh antigens matched transfusion was acceptable. Rh compatible transfusion is worthy to popularizing in China.

P751 | Hemostatic assessment by conventional and global coagulation assays in patients with COVID-19—an experience from a transfusion medicine center from north India

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Background: The coronavirus disease 2019 (COVID-19) has been shown to be predominantly associated with prothrombotic state. Some studies mainly from western world have highlighted the relevance of global coagulation assays for exploring the hemostatic profile in COVID-19 patients.

Aims: We aimed to see the utility of thromboelastography (TEG), a global viscoelastic coagulation assay, in assessing hemostatic profile in our COVID-19 patients as compared with conventional coagulation tests (CCT) as a part of evidence based laboratory practices.

Methods: Laboratory records and relevant clinical profiles of our hospitalized COVID-19 patients who underwent TEG testing using TEG 5000 Thromboelastograph Hemostasis Analyzer (Haemonetics, USA) in department of Transfusion Medicine over a one-year period were retrospectively studied. The reference ranges for various TEG parameters provided by manufacturer were used to define hyper and hypo-coagulable profiles.

Results: We analyzed data of 103 patients who had severe (41%), moderate (30%) or mild (29%) disease. Majority (41%) of the patients

had O blood group, followed by B (39%), A (13%) and AB (7%). The median values of platelet count was 152 (103-198) $\times 10^3/\text{mm}^3$, PT 14.9 (13.8-16.7)s, APTT 31.1 (28-36.3)s and fibrinogen 470 (327-596) mg/dL. Fifty-seven (55%) had normal platelet counts [reference range: (150-410) $\times 10^3/\text{mm}^3$], forty-four (43%) had thrombocytopenia while two had thrombocytosis. Increased INR (>1.2) was noted in 37 (36%) patients. Eighty-one (79%) patients had elevated D-dimer ($>0.5 \mu\text{g/mL}$) and seventy-five (73%) had increased FDP ($>2.5 \mu\text{g/mL}$) levels. Thirty-two (31%) had normal fibrinogen levels [reference range: 200.0-400.0 mg/dL], sixty-six (64%) had hyperfibrinogenemia while five (5%) had hypofibrinogenemia. On TEG analysis hypercoagulability was observed in 48% of them with enhanced clot strength: increased maximum amplitude (MA), clotting index (CI) and enhanced thrombin generation which were not detectable by conventional coagulation tests. Hypercoagulability was predominantly due to hyperfibrinogenemia and it persisted till 32-56 days in a subset of our recovered patients on repeat TEG analysis. Hypocoagulable TEG profile was observed in ten (9%) patients out of which, seven had platelet hypocoagulability and three had plasmatic hypocoagulability. Four patients with hypocoagulable TEG profile had clinically evident bleeding and received TEG-guided component transfusion support. Lesser recovery rate was observed in patients with hypocoagulable profile (60% mortality) compared to those with hypercoagulable and normocoagulable profiles. As compared with survivors, non-survivors showed statistically significantly lower MA and CI values.

Summary / Conclusions: A significant hypercoagulability was observed in almost half of the patients with COVID-19 which was not detectable by CCT in many patients. Hypercoagulability on TEG was observed in some patients even after clinical recovery. Major hemorrhage is an underreported complication of anticoagulated COVID-19 patients. However, it is possible that occult bleeding or hypocoagulability may be obscured by the viral procoagulant activity, which may be missed by CCT but detected as normocoagulable or hypocoagulable profile on viscoelastic tests such as TEG, and warrants modifications in thromboprophylaxis dosage. Increased mortality in our patients with hypocoagulable TEG suggests that COVID-19-associated hypocoagulability may be fatal without urgent intervention and rational hemostatic support.

P752 | Impact of granulocyte transfusions in infected neutropenic bone marrow transplant patients

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Background: Granulocyte transfusions have been used in the treatment of bacterial or fungal progressive infections in neutropenic patients for years. However, a real benefit has yet to be proven. We

aimed to analyze the impacts of granulocyte transfusions in the treatment of hematologic-oncologic and bone marrow-transplanted patients at our institution

Aims: Clinical response to transfusions and the survival rates of transfused patients were retrospectively evaluated and compared with non-transfused patients, and the occurrence of adverse transfusion reactions was analyzed

Methods: All neutropenic adult patients at the BMT Unit at the IBCC Oncology Hospital for whom granulocyte transfusion was requested from January/2015 to April/2020, in the city of São Paulo, Brazil were included. Donors received corticosteroids or corticosteroids+G-CSF, 12-16 h before donation. Collection procedures were performed using the continuous-flow apheresis machine COBE Spectra (Caridian BCT, Lakewood, CO). Transfusions were scheduled to occur until recovery from infection, neutrophils recovery, or death. Transfused patients were compared to non-transfused patients in terms of clinical response and survival to hospital discharge. Survival of patients that received concentrates from G-CSF primed donors was compared to those that received concentrates from donors mobilized with steroids alone

Results: Of the 69 adult patients included in the study, 21 were not transfused: 5 presented bone marrow recovery and 16 died before transfusion, either because it was difficult to prepare donors or because of their severe infection. 48 patients received 236 transfusions (4.9 transfusions/patient). The mean transfused dose was 0.36×10^9 granulocytes/kg. Fourteen adverse reactions were reported, all of which were mild and resolved after medication. Clinical improvement was observed in 30 of the 48 transfused patients. In the other 18 patients, their clinical condition worsened and they died. Clinical improvement had no correlation with the positivity of blood cultures, type of pathogen, underlying disease, cause of neutropenia, receiving more than four transfusions, age, weight, number of transfusions, mean dose transfused or time from the onset of infection to the first transfusion. Patients in the ICU, on ventilation, or in use of inotropes presented clinical worsening. In the group of non-transfused patients, 19 of them (90.5%) died at a median time of 13 days, and in the group of transfused patients, 29 patients (60.4%) died at a median time of 48 days. In the bivariate Cox analysis, the variables that correlated with better survival rates were: receiving granulocyte transfusions, number of transfusions, mean dose transfused, and more than four transfusions. Patients who received granulocyte concentrates from G-CSF primed donors did not have a better survival than those who received concentrates from donors mobilized with steroids alone: 27 patients died (61.4%) at a median time of 34 days compared to 2 patients (50%) at a median time of 48 days from donors mobilized with steroids ($p = 0.398$).

Summary / Conclusions: Granulocyte transfusions can be safely administered to neutropenic patients for the treatment of uncontrolled bacterial or fungal infections in the context of bone marrow transplantation. No definitive conclusions can be drawn from this analysis and further studies are warranted to evaluate the efficacy of granulocyte transfusions in the treatment of bacterial or fungal infections in neutropenic patients.

P753 | Coagulation dysfunction following burn injury as assessed by global coagulation assay—a pilot study from North India

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Background: Severe burns can cause profound alterations in hemostasis. An objective assessment of the hemostatic system is critical for effective management of burn patients. Thromboelastography (TEG), a viscoelastic global coagulation assay has not widely been used in burn trauma hence a pilot study was planned to assess the hemostatic profile in patients with thermal burn injury.

Aims: To study the hemostatic alterations in non-transfused burn patients using whole blood global coagulation assay.

Methods: Ten adult patients with severe burns involving 20%–40% total body surface area admitted to our centre in January 2024 were included in this study. Their clinical and laboratory details were recorded. Citrated blood samples were collected within 7 days of burn injuries for performing TEG and conventional coagulation tests (CCT). Analysis on TEG 5000 Thromboelastograph hemostasis machine (Haemonetics, USA) was performed on citrated blood samples within two hours of phlebotomy as per the established protocol using reference values of the manufacturer for TEG parameters. The TEG parameters studied were R, K, angle, MA, G, CI and LY30. TEG values obtained from our previous study on 10 healthy blood donors served as control. Both TEG and CCT were carried out using same blood samples.

Results: In 10 non-transfused adult patients with thermal burns (7 males and 3 females) the mean values of R (min), K (min), α (degrees), MA (mm), CI, and LY30 (%) in TEG were 4.1 ± 2.1 (min), 1.2 ± 0.4 (min), 76.3 ± 10.4 (°), 71.8 ± 8.4 (mm), 1.9 ± 3.6 , and 2.2 ± 3.0 (%), respectively. In CCT, mean values of international normalized ratio (INR), activated partial thromboplastin time (APTT), platelet count and fibrinogen were 1.6 ± 0.6 , 38.5 ± 0.7 , 93.5 ± 23.6 ($\times 10^3/\text{mm}^3$), and 383.3 ± 92.8 (mg/dl), respectively. In 4 patients on thromboprophylaxis the heparinase cups were used on TEG analysis. R value, K and α -angle values were within normal reference values whereas MA and CI parameters were found to be increased significantly in majority of the cohort on standard kaolin activated TEG. The velocity curve values MRTG, TMRTG and TG obtained from TEG were also altered from the reference values. Importantly hypercoagulable TEG showed robust thrombin generation marked by enhanced TTG values. Hyperfibrinolysis was not observed on TEG as LY30 and EPL values were not affected except in one patient. PT and APTT values showed a hypocoagulable picture in contrast to hypercoagulable TEG profile. Fibrinogen level was found to be increased in most of the patients.

Summary / Conclusions: The results from this pilot study show that hypercoagulability is present on global coagulation assay in non-transfused severe burn patients probably due to increased fibrinogen

concentration with decreased fibrinogen breakdown. Any hemostatic intervention should involve consideration of results of global coagulation tests. A detailed study involving at least 50 burn patients with serial TEG monitoring along with TEG velocity curve studies and functional fibrinogen assay is in progress which will throw more light on this topic.

P754 | Platelets suspended in plasma-only as therapy for bleeding patients with coagulation factor V inhibitors—a case report and literature review

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Background: Coagulation factor V (FV) circulates in plasma (80%) and is stored in alpha-granules of platelets (PLTs; 20%). FV inhibitors may develop following exposure to exogenous FV in individuals with congenital deficiency. Rarely, acquired FV deficiency can occur in patients secondary to various etiologies (e.g., drugs, autoimmune conditions, malignancy). Treatment of actively bleeding patients with FV inhibitors is challenging. With no FV concentrate clinically available, FV replacement can only be achieved with blood products. However, FV activity varies among the multiple types of products available. For example, FV activity is significantly lower in “thawed plasma” (i.e., frozen plasma that has been thawed and stored at 1–6°C for >24 hours) than in “freshly thawed” fresh frozen plasma (FFP) due to its short in vitro half-life. PLTs stored in plasma will inherently have greater FV activity compared to PLT additive solution (PAS).

Aims: Report a case of congenital FV deficiency with FV inhibitor and severe bleeding treated with apheresis-derived PLTs suspended in plasma-only with a concomitant literature review.

Methods: Case report and literature review of 276 articles that included reports of FV inhibitors.

Results: An elderly-man with congenital moderate FV deficiency developed a spontaneous acute on chronic subdural hematoma with midline shift. Medical history was significant for epistaxis and prior surgeries supported with FFP without bleeding complications. On admission, he received FFP, with initial laboratory results demonstrating prolonged Prothrombin Time (PT) and Activated Partial Thromboplastin Time (APTT). The prolonged clotting times did not correct following mix with normal pooled plasma, suggesting the presence of an inhibitor. FV activity level was 2% with a FV inhibitor quantified at 5 Bethesda Units (BU). Therapy was initiated with 100 mg prednisone and FEIBA (factor eight inhibitor bypassing activity) containing non-activated factors II, IX & X, and factor VIIa. Due to concern for active thrombosis, FEIBA was discontinued after 5 doses. Transfusion medicine was consulted for guidance on transfusion therapy. Due to minimal FV (i.e., <5%) in PAS PLTs, the blood bank prepared to support this patient with units of PLTs in plasma. The patient received one-half unit of apheresis PLTs in plasma every

12 hours for 6 days, and was discharged following improvement. He continued prednisone taper and plasma-only PLT transfusion 3x/week for the next month as an outpatient. A systematic literature review revealed that 30% (83/276) of published articles reported at least one case that utilized PLTs as a therapeutic source of FV. However, no case described whether platelets were resuspended in plasma or PAS.

Summary / Conclusions: This rare case of congenital FV deficiency with a FV inhibitor and severe bleeding was successfully managed using immunosuppression and PLTs suspended in plasma-only. The benefit of PLT therapy may be partially attributed to the theoretical “protective carrier effect” wherein PLTs degranulate FV at the site of injury, bypassing an inhibitor. If PLTs are considered for therapy in patients with FV inhibitors, plasma-only PLT units should be provided as units with PAS have minimal FV activity.

P755 | Does creating a maximum surgical blood component request schedule in a cardiac surgery reference center affect blood usage rates?

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Background: Preoperative blood and blood component requests play a crucial role in cardiac surgery. In this group of surgery, it is essential that reserved products are prepared and kept on standby. If any reserved products remain unused during the operation, they are withdrawn and returned to the available stock. However, these processes may lead to an increase in cross-match and transfusion rates. These, in turn, may impact clinical outcomes and resource allocation. To avoid unnecessarily retaining cross-matching and patient-specific reserved products, the Maximum Surgical Blood Component Request Schedule (MSBCRS) was established according to the respective usage rates of the centres. This practice aims to cater to clinics'

P755 - Table 1. Preoperative characteristics of patients'

	n (%) /Median (IQR)
Age	61 (54-68)
Gender, male	2315 (80.6)
BMI	27.70 (25-30.4)
Baseline Hgb	13.3 (11.9-14.5)
Baseline Hct	40.30 (36.2-43.5)
INR	1.05 (0.99-1.12)
APTT	28.8 (27-31.3)

IQR: Interquartile range (25-75), BMI: Body mass index, Hgb: Hemoglobin, Hct: Hematocrit, INR: International normalized ratio, APTT: Activated partial thromboplastin time.

demands and ensure that near-expiry products are available to other patients in need.

Aims: We conducted a study to present our centre's blood and blood component usage rates before and after the implementation of the MSBCRS.

Methods: Between January 2020 and December 2023, patients over the age of 18 who underwent isolated coronary artery bypass graft (CABG) surgery were evaluated into two periods before and after the creation of MSBCRS. Patients under 18, operated on other than CABG, undergone revision and supported by extracorporeal membrane oxygenation were excluded from the study. Patients' demographics, cross clamp time, cardio pulmonary bypass pump (CPB) time, initial and postoperative Hb, Hct values and the number of blood and blood product usage from the surgery until the first postoperative day was obtained from the daily follow-up records of the Hemovigilance Unit and Hospital Information Management System (HIMS) retrospectively.

Results: A total of 2871 patients were included in the study. 80.6% ($n = 2315$) were male and the median age was 61 (54-68) years. The rate of patients operated on in the 2020-2021 period is 37.1% ($n = 1065$), and the rate of patients operated on in the 2022-2023 period is 62.9% ($n = 1806$). Other preoperative characteristics of the patients are shown in Table 1. In the comparison of pre, intra and postoperative variables of the patients in both periods, a statistically significant difference was found between the groups in terms of age, gender, preoperative hemoglobin, hematocrit, INR and APTT levels ($p < 0.001$). There is a statistically significant difference between the groups in terms of CPB, ACC time, total ES, FFP and PPLT suspension numbers ($p < 0.05$). In blood product use, the average use of ES and FFP is higher in the 2022-2023 period, and the average use of PPLT suspension is higher in the 2020-2021 period.

Summary / Conclusions: In this study, we found that the use of ES and FFP was significantly lower in the first period, but on the contrary PPLT suspension was used less in the second period. We think that this may be due to the low number of operations performed in the first period. We believe that further prospective studies are needed to investigate the effect of creating maximum surgical blood demand schedules on blood usage.

P756 | Autoimmune hemolytic anaemia—transfusion experience in a single centre

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Background: Autoimmune haemolytic anaemia (AIHA) is a rare disease with an incidence of 1 to 3 cases per 100,000 per year in adults. Anemia caused by AIHA ranges from mild

asymptomatic anemia to severe life-threatening conditions. The efficacy of red blood cell (RBC) transfusion is debated because of speculations on the increased risk of transfusion reactions. The pretransfusion testing in these patients must be rigorous to ensure safe transfusion.

Aims: In this study, we reviewed the clinical characteristics and immunohaematological study of 25 patients diagnosed with AIHA. And we evaluated the safety and efficacy of RBC transfusions in this cohort.

Methods: Retrospective and observational study of 25 patients with new-onset AIHA from January 2017 to April 2023 in a tertiary care medical center in Spain.

Results: 25 patients were analysed with a median age of 77 years (range 42-94), 80% of whom were over 60 years old. 87% were diagnosed with warm autoimmune hemolytic anemia (wAIHA) and 12% by cold autoimmune hemolytic anemia (cAIHA). The frequency of primary AIHA (pAIHA) (52%) and secondary AIHA (sAIHA) (48%) was similar. The majority of patients had grade III anemia (44%), with a median hemoglobin level at diagnosis of 7.2g/dl. **Pretransfusions testing:** Out of the 22 patients with wAIHA, direct antiglobulin test (DAT) was positive for IgG with C3d positivity in 10 cases. DAT for patients with cAIHA was positive for C3d and the cryoagglutinins were detected in 100% of the cases. In wAIHA patients, eluate test was positive with all cells in 17 cases (77%) and specificity for Rh antigens in the remaining 5 (4 anti-e and 1 anti-D). Autologous adsorption test was optimal (removing antibody) in 19 patients (76%), of which only 6 had been transfused previously, only one of them had alloantibody. All patients were genotyped. **Transfusion at diagnosis:** 13 patients (52%) were transfused at diagnosis, with a median of 1 unit of RBC per patient. All transfused patients had grade III (54%) or IV (46%) anemia. The transfusion was considered effective when there was an increase in the Hb level of at least 0.5g/dl per unit of RBC; and it was effective in 9 patients (69.2% of transfused), with a median increase in Hb of 0.8g/dl per unit of RBC. No hemolytic transfusion reactions (HTR) were reported. **Treatment and responses:** The most common treatment was corticosteroids (84%), followed by rituximab (8%). 52% relapsed to the first line, in the transfused patients group 30,7% relapsed.

Summary / Conclusions: The clinical features are very similar to those described in the literature: higher number of wAIHA versus cAIHA, and similar frequencies of pAIHA and sAIHA. In AIHA patients, the presence of alloantibodies is usually increased (40%–60%), being the main difficulty in pretransfusions testing. In our sample, patients with optimal autologous adsorption (removal of autoantibodies/last transfusion more than three months ago), we only detected alloantibodies in 5% of them. Crossmatch was conducted in the majority of patients with autologous adsorbed serum (76%) and in all cases phenotyped blood was transfused. No HTR were reported. The efficacy of RBC transfusion is debated, in our sample it was effective in 69.2% of patients. With rigorous pretransfusions testing, we can ensure safe transfusions, improving the anemic syndrome in those patients who need it.

P757 | Abstract withdrawn

P758 | Comparative analysis of the adequacy of the use of blood components in Donostia University Hospital (DUH)

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Background: In 2015, we conducted a systematic review of the existing evidence of the transfusion recommendations using the AGREE II instrument (used for the evaluation of Clinical Practice Guidelines) and we developed an internal Guide for the Use of Blood Components for DUH (2016 edition) based on them.

Aims: To analyze and compare the adequacy of the indications of blood component transfusion in Donostia University Hospital (DUH) between the years 2015 and the period between 2019 and 2022.

Methods: 860 randomly selected transfusion episodes were reviewed in the months between April and June of 2015; differentiating platelets (PC), red blood cells (RBC) and fresh frozen plasma (FFP). Between 2016 and 2022, training in transfusion knowledge was reinforced among our hospital staff, emphasizing the importance of adapting the indications for blood components and promoting their optimal use through the Hospital Transfusion Board. In order to assess the outcome of these efforts, a total of 1792 transfusion episodes were analyzed from January 2019 to December 2022.

Results: As we can observe in Table 1, there has been a decrease in the no adequate indications for transfusion of both RBC and FFP, but not in the case of PC transfusions.

We observed that the most frequent cause of inadequacy in the case of RBC transfusion was the same both in 2015 and in the period between 2019 and 2022 and involved RBC transfusions with Hb levels between 8 and 10 mg/dl without associated risk factors or symptoms to justify it. Likewise, the most frequent cause of inappropriate use of FFP, was the same in the two time periods, this being the transfusion of FFP with INR/APTT <1.5 and without hemorrhagic symptoms or the need for an invasive procedure. In the case of PC transfusion, the number of inappropriate transfusions has remained stable between 2015 and 2019-2022. However, the most frequent

P758 - Table 1. Comparative analysis between the adequacy of the transfusions during 2015 and 2019-2022.

	Adequate	Adequate	Non Adequate	Non Adequate
	2015	2019-2022	2015	2019-2022
RBC	86%	96%	14%	4%
	542	1049	88	42
Platelets	94.8%	94%	5.21%	6%
	182	353	10	24
FFP	78.9%	97%	12.3%	3%
	30	324	106	9
Total	87.7%	95%	12.3%	5%
	754	1717	106	75

cause was different; in 2015 we noticed that the platelet number >10,000 without associated risk factors or surgical intervention was the most frequent cause of inadequate transfusion; yet between 2019-2022, it was the transfusion prior to an invasive procedure with a number of platelets greater than 50.000 (not CNS, eyeball surgery or polytrauma). We also analyzed the adequacy of transfusion in the most transfusion-intensive departments of the hospital. The department that least complied with the guidelines between 2019 and 2022 was Oncology; however, in 2015 the Anesthesiology Department was the one with the highest number of non-adequate transfusions.

Summary / Conclusions: We observed a significant decrease in the inappropriateness of the indications for RBC and PC transfusion with lower figures than the ones reported in the reviewed literature. FFP transfusion is the blood component that best meets transfusion indications as explained in our guide. PC transfusion adequacy remains stable; however, the most frequent cause of inappropriate use is different between the years 2015 and 2019-2022. Between 2019 and 2022, Oncology was the department with the highest amount of non-adequate transfusions. However a great improvement has been observed.

P759 | Requests for blood transfusion and deposition of packed red blood cells at San Pedro Hospital (Spain)

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Background: The Blood Bank of La Rioja, based at San Pedro Hospital (Logroño, La Rioja), caters to blood component demands from regional hospitals. Over 80% of Packed Red Blood Cells (PRBC) are transfused there. Despite declining donation rates, PRBC supply, especially O negative type, has risen. This calls for scrutinizing red blood cell transfusion requests and storage. La Rioja has seen decreasing blood donations, yet certain components, notably O negative PRBC, have increased.

Aims: Analysing red blood cell transfusion requests and storage.

Methods: We have analysed the distribution of PRBC in San Pedro Hospital over the last five years, taking into account the ABO and Rh group. An exhaustive examination of PRBC distribution within San Pedro Hospital over the preceding five years has been conducted, with due consideration given to the ABO and Rh group classifications. Similarly, the number and percentage of PRBC discarded at the San Pedro Hospital in relation to the number of PRBC obtained by whole blood donation and apheresis has also been reviewed during these years. In the same way, the most frequent reasons for which PRBC are withdrawn have been collected. Simultaneously, a review has been undertaken to scrutinize the number and percentage of discarded PRBC at San Pedro Hospital concerning the total PRBC obtained through both whole blood donation and apheresis methodologies during this temporal span. Furthermore, a compilation of the predominant reasons necessitating the withdrawal of PRBC has been

P759 - Table 1.

	% variation 2019-2023
PRBC Requests	+2.44
O positive	+2.73
O negative	+16.95
A positive	-1.05
A negative	+7.35
B positive	-17.96
B negative	-0.97
AB positive	-15.33
AB negative	+6.52

meticulously documented. The data has been obtained from the e-Progesa and e-BDI Plus programs.

Results: The variation of the distribution of PRBC according to ABO blood group and Rh from 2019 to 2023 is reflected in the following table:

The percentage of PRBC discarded with respect to the number of red blood cell concentrates obtained will be reflected below:

P759 - Table 2.

	% variation 2019-2023
PRBC donated	-8.89
PRBC discarded	-5.27

Summary / Conclusions: (1). In the analyzed five-year period, PRBC demand rose by 2.44%, notably for O- type, with a 16.95% distribution increase. (2). Discarded PRBC concentrates decreased by 5.27% from 2019 to 2023, indicating improved stock management.

P760 | Assessment of characteristics of recipients and patterns of use of blood and blood products at the Institute for Transfusion Medicine FBIH in 2023

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Background: Blood transfusion is still an irreplaceable method in the therapy of patients with blood and blood products. Transfusion is often used unjustifiably, especially in developing countries. This survey was designed to assess recipient characteristics and blood distribution in relation to the clinics from which claims were received.

Aims: This study was designed to assess recipient characteristics and blood distribution in relation to clinics from which claims were received.

Methods: The study was conducted from January 1 to December 29, 2023. The data were collected from the information system Renovatio, which is used in the Institute for Transfusion Medicine F BiH. The patient's age, gender, requesting departments, number and type of blood products were collected.

Results: A total of 27,927 units of blood products were issued to 3344 patients. The average age of the transfused persons was 57.25 years, and this was almost the same for male and female patients. The age range was from 0-98 years, where the ratio of male (1664) and female (1680) patients was almost equal. A request for blood was recorded from 31 clinics, namely 12,264 DE, 6977 platelet preparations, 8669 fresh frozen plasmas and 17 cryoprecipitate preparations were issued. The number of dispensed doses per patient ranged from 1 to 356 doses per patient.

Summary / Conclusions: Dominant group of patients for whom less than 5 blood products were issued, female, middle-aged. Our findings support the need for comprehensive evaluation of transfusion preparations on recipient outcomes using input/output clinical parameters hgb etc. to further understand the risks and benefits of RBC transfusion. Today, transfusion services are increasingly adopting multidisciplinary patient blood management measures to ensure fewer and more efficient transfusions to achieve clinical outcomes, including adequate hemostasis.

P761 | Evaluation of clinical usage and of red blood cell concentrates

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Background: The rational use of blood is an imperative for each blood bank and hospital.

Aims: The aim of this study is to compare clinical usage of red blood cell concentrates (RBCC) for last three years 2021-2023.

Methods: In this retrospective study we analyzed data from the department of clinical transfusion and distribution of blood using the informative system e- Delphyin.

Results: In 2021 there were 50925 donated units of blood from which 47449 RBCC were issued and 3479 (6.8%) were eliminated. Only 569 (1, 1%) RBCC expired. For usage in Clinical Center Mother Teresa (CCMT) were cross-matched 21867 (46%) RBCC from which 10116 (46.5%) were issued for different internal clinics and 11701 (53.5%) for surgery. In 2022 there were 536315 donated units of blood from which 50103 RBCC were issued and 3528 (6.6%) were eliminated. Only 563 (1%) RBCC expired. For usage in Clinical Center Mother Teresa (CCMT) were cross-matched 25209 (50%) RBCC from which 11380 (45.2%) were issued for different internal clinics and 13829 (54.8%) for surgery. In 2023 there were 56451 donated units of blood from which 53192 RBCC were issued and 3261 (5.8%) were

eliminated. Only 381 (0.7%) RBCC expired. For usage in Clinical Center Mother Teresa (CCMT) were cross-matched 24703 (46.4%) RBCC from which 13336 (45.8%) were issued for different internal clinics and 13367 (54.2%) for surgery.

Summary / Conclusions: We can conclude that the number of blood donation and issued RBCC units is growing up by the years. The distribution of RBCC to other blood banks and private hospitals is also increased. In the past the needs of blood components from surgical and internal hospitals were equal, but nowadays the surgical hospitals have increased needs for blood as a result of the new surgical procedures. Only 1% of blood is eliminated due to expiration in comparison to 6.3% of total eliminated blood units from other reasons.

P762 | Abstract withdrawn

P763 | Autologous platelet rich plasma—a study of its therapeutic applications in a tertiary care hospital in Eastern India

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Background: Platelet Rich Plasma (PRP), the term coined in 1970s to describe the plasma derived from whole blood which contained a higher count of platelet. The working definition of PRP for clinical efficacy is an increase of platelet count 3-5 times of the baseline, approximately greater than 1 million platelets/ μ l. They have been in use for last 30 years but have recently generated considerable interest in its potential in regenerative medicine. Also, the 'autologous' component of autologous PRP (A-PRP) has a better safety index compared to allogenic products. It's also more cost effective by promoting faster healing thus reducing hospital stay. Despite the promising results associated with its use, there is still a lack of standardization of preparation techniques. This study is a step towards establishing this goal.

P763 - Table 1: Comparison of laboratory values pre- and post-intervention

Laboratory value	Pre-intervention Mean(SD)	Post-intervention Mean(SD)	p value
Platelet count (in lac)	2.15 (0.63)	8.95 (7.09)	<0.001
WBC	5.86×10^3 (1.94)	1.26×10^3 (0.88)	<0.001
HCT	3.44×10^1 (0.55)	0.06×10^1 (0.03)	<0.001

P763 - Table 2: Clinical outcome observed.

Characteristics	Number	Percentages
Pain reduced	18	35.3
Improved range of movement	13	25.5
Increased growth	5	9.8
Successful implantation	5	9.8
2nd cycle of PRP given	2	3.9
No change	8	15.7

Aims: To standardize the preparation and quality of A-PRP. To evaluate the efficacy of the A-PRP in inducing desired effect of repair and regeneration to the targeted tissue cells.

Methods: A prospective observational study was conducted in the Dept of Transfusion Medicine, IMS and SUM Hospital, Bhubaneswar from Nov 20 to Sept 22. Patients who consented for this modality were recruited for the study. The PRP was prepared by the centrifugation method and the centrifuge used was Cryofuge 5500i with LH-4000W rotor and double blood bag bucket, Thermo Fisher Scientific Company (Massachusetts, US). The product was issued after clearing the internal quality control check.

Results: A total of 51 patients between the ages of 24 and 57 years participated in the study. Of them, 9% were male 6% were below 40 years Majority were 'O' group (35.3%) followed by 'B' group (2.5%), 'A' group (21.6%) and 'AB' group (15%) 1% were Rh positive. Distribution among the disease conditions were as follows: plantar fasciitis (35.3%), frozen shoulder (19.6%), primary infertility (17.6%), tennis elbow (11.8%), telogen effluvium (5.9%), osteoarthritis (3.9%), secondary infertility (3.9%) and androgenic alopecia (2%). Patients were recruited from orthopaedics, obstetrics and gynaecology and dermatology departments. The mean volume of blood collected from the patients was 117.45 ± 5.6 ml which produced a mean volume of PRP of 16.14 ± 2.69 ml.

It was found that on an average post-intervention platelet count was significantly higher (3.16 times) than the pre-intervention counts. Although the post-intervention levels were markedly reduced of WBC count (0.79 times) and HCT (0.61 times) than the pre-intervention levels.

The desired outcome was achieved in 80.4% of patients.

Summary / Conclusions: A-PRP injection is a simple, economical, and practically feasible mode of therapy. It can be used in conjunction with other modes of therapy to hasten the healing hereby reducing hospital stay. The method of preparation has a significant impact on the level of platelet recovery and activation. Standardization of its production is thus essential. The method by which PRP was produced resulted in significant changes in the hematological parameters in majority of patients.

P764 | Abstract withdrawn

Clinical transfusion— haemorrhage and massive transfusion

P765 | Abstract withdrawn

P766 | Using low titre group O whole blood for massive
transfusion activation at a medical centre in Taiwan

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Background: Traumatic hemorrhage remains a leading cause of death in civilians, and they need a rapid transfusion with large volumes of blood products to save lives. In 2017, we established massive transfusion protocols for massive bleeding patients. The first run of MTPs has a predefined ratio of RBCs, FFP, and apheresis platelet is 6:6:1. After the MTPs implementation, it has shown certain benefits but still can be improved. Because we do not have a pre-thawing procedure for fresh frozen plasma, when MTPs are triggered, it takes 30 minutes to release fresh frozen plasma for transfusion. So our emergency physician suggested using low titre group O whole blood (LTOWB) for treating trauma patients. LTOWB is being used in Europe and America but not Taiwan because our blood center does not supply LTOWB.

Aims: The aims of this study were to present a new protocol for MTPs and the following strategy after transfusion.

Methods: The AABB Standard and serological principles technical manual were applied.

Results: Since April 2023, the Department of Emergency Medicine and Clinical Pathology Center in our institution started collaborating to establish a MTPs incorporated with LTOWB units. First of all, we had to discuss with our blood supply center, the Kaohsiung Blood Center, to provide LTOWB units. Since there is no universal guideline for low titre threshold, considering the risk of hemolysis and proper blood collection units, the isohemagglutinin titres were defined as IgM anti-A and Anti-B <128 because when the titre threshold <50, according to our experiment data, around 90% donors will be excluded. All pre-storage leukoreduced group O+ whole blood products were collected and processed by the leukocyte reduction filter (PuriBlood, CO) that can be stored for up to 35 days. Secondly, LTOWB units would only be used in the first run of MTPs with 6 units (each unit is 250 c.c.). From the second run of MTPs, component therapy strategy will be taken. To distinguish LTOWB from the regular WB in stock, our blood bank system must have a record of LTOWB titres, and our system would block the blood units' issue process if the electronic system could not detect the titres data. If the LTOWB units were not transfused by day 14 after collection, they will return to the blood center and be manufactured into RL-RBCs units that could be transfused till day 35. Moreover, to follow the transfusion adverse reactions, biochemical markers of hemolysis which are LDH, haptoglobin, total bilirubin,

creatinine, and potassium levels will be measured on day 0 (the day of LTOWB transfusion), day 1, and day 2, respectively. On January 12, 2024, the new MTPs process was completed, and our inventory will include LTOWB for emergency usage.

Summary / Conclusions: To our knowledge, we are the first medical center to implement LTOWB with MTPs in our country. However, there are some issues we need to monitor carefully. Above all, the LTOWB units in America or Europe almost with a platelet-sparing filter that has a high platelet recovery; on the contrary, the platelet recovery rate of LTOWB we obtained is low because of the leukocyte reduction filter function. Though there are many LTOWB treatment experiences with low platelet recovery, we still need to pay attention to follow cautiously for our patients' treatment outcomes. Besides, as storage time increases, platelet function decreases rapidly, so blood bank staff have to manage apheresis platelet inventory effectively to prevent blood shortage or wastage.

P767 | Audit of use of massive haemorrhage protocol at Leeds
Teaching Hospitals NHS Trust

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Background: Massive Haemorrhage (MH) is a medical emergency that requires prompt management through protocol-driven multidisciplinary team approach. In Leeds Teaching Hospitals NHS Trust (LTH) the MH includes initial release of 4 units of red blood cells (RBC) and 4 units of pre-thawed Fresh frozen plasma (FFP) following by release of more RBC and FFP in a ratio of 1:1 and platelets and cryoprecipitate if hemorrhage continues. The use of MH protocol in relation to transfusion has been audited quarterly since 2015.

Aims: The objective of the audit is to improve communication between the clinical areas and the blood bank laboratory and to reduce wastage of blood components issued in the MH.

Methods: Data are collected by the blood bank laboratory staff for each time the MH is initiated in the Trust. The gathered data include site of MH, timing, samples arriving at the lab, designated communicator and order to step down the protocol. In addition, to the amount of blood products issued, transfused and disposed.

Results: MH has been activated 1320 times during an eight-year period (2015-2022). The majority of cases were trauma ($n = 734$) followed by obstetric (295) and non-trauma non obstetric patients ($n = 259$). We present the data from trauma and non trauma, non obstetric patients.

Summary / Conclusions: The number of MH in trauma and non trauma, non obstetric patients have increased significantly over the eight year period of 2015 to 2022. The vast increase in number of MH protocol activation within the context of different clinical

P767 - Table: Comparison of MH audit outcomes 2015-2022

	2015 Trauma patients	2022 Trauma patients	2015 non trauma non obstetric patients	2022 non trauma non obstetric patients
Number of patients for whom MH protocol was activated.	21	149	15	71
Number of patients transfused.	18	118	15	71
% of MH cases that had a designated communicator.	10	54	60	34
% of MH cases that the laboratory was informed to stand down.	14	34	33	28
Number of RBC issued.	298	1044	165	441
% of untransfused RBC units wasted.	9	5	5	12
Number of FFP issued	135	707	77	363
% of untransfused FFP wasted	56	29	54	51

scenarios remains a challenging issue that needs prompt management and regular review of local protocols. There is a significant number of MH protocol activations for trauma patients that are proven not to bleed which indicates that transfusion lab is included in the initial alert of Accident and Emergency. Communication between the laboratory and clinical areas as reflected by providing a designated communicator for the MH protocol and the order for the laboratory to step down has improved for trauma patients since the introduction of ongoing audits, regular meetings and drills with the Accident and Emergency team. The majority of non-transfused disposed blood components were Group O RhD negative red cells and fresh frozen plasma. Despite the increase in the overall number of MH protocol activated, the number of disposed fresh frozen plasma has declined over the most recent years and the number of disposed platelets remain low as the platelets are not included in the primary MH pack. Nevertheless, there remains a scope for improvement through methods to reduce disposal of non transfused blood components.

P768 | Emergent blood cell transfusion clinical practice of the last six years in a tertiary hospital

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Background: Red blood cells transfusion (RBCT) is an important therapeutic resource and according to several studies at least 20% are an issue in emergency room (ER). The inability to provide, in a timely manner, RBCT to a patient with a massive bleeding leads to increased mortality. Since our current blood bank is far away from the ER, to ensure a promptly available RBCT a new protocol was established allocating 4 units of uncrossmatched type O RBCs (2 type ORh positive and 2 type ORh negative) to the ER, with a written policy explaining how to use: women < 50 years-old and children should receive type O Rh- and all the others patients should receive type O Rh+ while waiting for a compatible unit. In addition, with the

P768 - Table 1 - Study Results. GI - Gastrointestinal

Characteristics, n(%)	G1	G2
Male / Female	84 (61.8) / 52 (38.2)	91 (65.4) / 48 (34.5)
Total	136	139
Cause of admission		
Trauma / GI bleeding	45 (33.1) / 59 (43.4)	43 (30.9) / 34 (23.5)
Uncrossmatched RBCs units transfused per patient		
1	43 (31.6)	72 (51.8)
2	79 (58.1)	61 (43.9)
3 or 4	14 (10.3)	6 (4.3)

P768 - Table 2 - Study Results. *according to our protocol

Characteristics, n (%)	G1	G2
Transfused uncrossmatched RBCs units		
Type ORh- / ORh+	231 (94.1) / 17 (5.9)	76 / 137
Correctly / Incorrectly*	-	148 (69.1) / 65 (30.9)
Total	248	213
Incorrectly transfused uncrossmatched RBCs units*		
Male, ORh-	-	42 (64.6)
Female, > 50 years ORh- / < 50 years ORh+	-	19 (29.2) / 4 (6.2)
Deaths		
Total	24 (17.6)	21 (14.3)

implementation of the vacuum system it's possible to send compatible units in a more rapid and efficient way.

Aims: To evaluate the compliance to these transfusion practices and to assess whether there is a benefit on having uncrossmatched RBCs in the ER.

Methods: We conducted an observational study of 275 patients admitted to our hospital that required emergency RBCT. Patients were divided in 2 groups according to the date they were admitted in ER - before and after our protocol was activated: group 1 (G1) from 2017 to 2019 and group 2 (G2) from 2020 to 2022.

Results: We obtained data from 275 patients. Table 1 shows the baseline characteristics, the most common underlying conditions and the number of uncrossmatched RBCs transfused in each group. The number of RBCs incorrectly selected according to our protocol is shown in table 2. Three female patients aged <50 years (mean age 45 ± 6 years) were incorrectly transfused; however, two of them were Rh+ and the one who was Rh- died, making it impossible to know if she developed anti-Rh antibodies.

Summary / Conclusions: We conclude the number of uncrossmatched RBCT was lower in G2. Additionally, male patients transfused with type O Rh- RBCs were the most frequent deviation from our internal protocol. We can speculate that this may be due to lack of knowledge/adherence of physicians to emergency transfusion protocol. In the ER it may not be possible to know patients' age before starting

treatment, which may explain why 3 women (mean age 45 ± 6 years) were transfused with type O Rh+. Due to our study findings our blood bank service briefed the ER director regarding the significance of a theoretical and practical training in emergency RBCT, as lack of awareness of the risks and unnecessary costs of inappropriate use of blood products may lead to avoidable errors.

P769 | Blood on the frontlines—uncovering the vital role of massive transfusion protocols in critical care

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Background: In the context of major trauma, the Massive Transfusion Protocol (MTP) has been a critical intervention, significantly reducing transfusion time, mortality rates, and the rejection of blood components. Implemented in our center since 2008 with updates in 2014 and 2023, this study provides a comprehensive **Transfusion Service** perspective, focusing on 2023 when systematic data collection was established

Aims: This work explores activation patterns across hospital units, assessing the MTP's compliance, alloimmunizations (ALLI), impact on

P769 - Table 1.

Preparation transfusional panel in adults			
1st transfusional pack (TP)	2-4 RBC O- (or Isogroup)	Min O'	Tranexamic 10-20mg/kg (Viscoelastic guiding)
2nd TP	6RBC/1Pool	M: 10'	Fibrinogen 2gr
3th 4th... TP	6RBC/4FFP/1Pool	M: 20'	Fibrinogen 2gr
Notify The Blood Bank Regarding The Protocol'S Continuation			

P769 - Table 2: Results.

Leading Cause	Gender	ABO	Blood Prod Sent/Back	Deat 24 h
Traffic	M:10 F:6	O+ (33)	RBC 10.6 ± 5.7 / 3.7 ± 3	Y:21 No: 62
Pricip (Autolytic)	M:9(6) F:9 (2)	A+ (23)	FFP 3.6 ± 2.9 / 0.52 ± 1.1	Death 30D
GI	M:8 F:4	B+ (7)	Platelets 1.5 ± 1.9 / 0.13 ± 0.3	Y:10 No:52
Intrahospital	M:8 F:11	AB+ (4)	Median Trans Packs: 2.1	
Stab/Gun Wound	M:8 F:2	O- (4)	Medical Identified	
		A- (8)	Yes: 78 No: 5	
		B- (3)		

minimizing blood products wastage and mortality rates at 24 h and 30 days. Our analysis offers unique insights into the ongoing evolution and effectiveness of the MTP, highlighting its pivotal role in optimizing patient outcomes.

Methods: Single-centre study, retrospective conducted in 2023. Statistical analyses are presented in terms of medians, percentages, and correlation assessments. The study delves into previous medical studies, blood product deviations, and the number of protocol steps taken.

Results: In 2023, 83 MTP were activated. Activations occurred in various settings: 35% in pre-hospital (27), 63% in the trauma intensive care unit (TICU) (53), 16% in surgical services (13), 10% in both the medical intensive care unit (ICU) and the cardiac ICU (5 each). 2 activations were in obstetrics, and 5 in paediatrics. The median age was 46, with 13 individuals under 30. Among the 53 MTP activations in males, traffic-related incidents were the principal cause, while for females 30 MTP activations, intrahospital events predominated. Other causes are detailed in the table. The median duration of MTP was 39 min (IQR: 5–50 min), with a standard deviation of 26 min. Hemoderivatives distribution comprised 884 units of Red Blood Cells (35% returned: 310), 130 units of Platelets (8% returned: 11), and 298 units of Fresh Frozen Plasma (14% returned: 43). The overall rejection rate for blood products was (18) 1%. Among Rh-negative patients (15%), 5 received Rh-positive red blood cells. The two survivors received 7 and 8 RBC units with no allo anti-D. In 56 cases (67%) were not previously studied and no blood transfusion reactions occurred. In total ALLI occurred in 1 patient with anti-K + anti-Lua. The global mortality rate was (31) 37%, with 25% and 32% mortality rates at 24 h and 30 days, respectively.

Summary / Conclusions: The MTP ensures standardized care, reducing rejection of blood products to just 1% out of 1312 distributed. With a primary impact observed in critical care units, particularly the trauma unit, there is a clear need for widespread protocol knowledge in these areas and operating rooms. Despite of 67% unknown patients, no haemolytic reactions or relevant alloimmunizations were observed among survivors including Rh-negative individuals, mortality rates similar than reported. "Quilten, Transfusion Revue Medicine, 2018".

P770 | A decade of experience in prehospital transfusion in Castilla La Mancha—innovation and management in red blood cell packets, fibrinogen and calcium

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Background: Haemorrhage is a leading cause of preventable death for population suffering accidents or bleeding injuries in regions with low

population density where healthcare services may not cover all population in remote areas. Resuscitation using blood products and limited infusion of normal saline improves survival of critical bleeding patients. HEMS program in Castilla-La Mancha, which includes physicians and nurses, was the first out of hospital Emergency Service in Spain that provided packed red blood cells (pRBC) transfusion where the accident or the pathology happens without delaying the transport to the referral Hospital. This program has been developed jointly between the haematologists of the Blood Bank of Ciudad Real and Gigante 2, Emergency Prehospital Service of Castilla-La Mancha. Calcium and Fibrinogen concentrate had been added to many others blood transfusion protocols around the world as a complement for prevention of coagulopathy.

Aims: To describe the process of out of hospital pRBC transfusion in the HEMS of Castilla-La Mancha over the last 10 years. The protocol for out of hospital blood transfusion was developed according to medical indications criteria and safety, monitoring and tracking items established by national law. In June of 2023 this procedure added human fibrinogen concentrate and calcium to improve coagulopathy level.

Methods: Retrospective observational study including outpatients from June 2014 to December 2023. The medical helicopter (EC 145T2) was provided with two pRBC O Rh(D) negative, and two grams of fibrinogen. Also calcium gluconate. The Shock Index was selected for the indication of transfusion according to the literature reviewed and as a rate simple to obtain out-of-hospital. To achieve the feasibility and preservation of the pRBC it was established a prospective monitoring of volume, haematocrit, haemoglobine, leucocytes, coulter, hemolysis and microbiological culture. The control hematologic analyses were performed immediately and 35 days after collection.

Results: One hundred and sixteen pRBC transfusions were administered to 76 patients during out-of-hospital advanced medical assistance. The mean age of the patients was 49 years (range: 12–90 years). 59 out of 76 patients had polytrauma, 8 patients digestive hemorrhage, 2 patients gun accident, 2 knife accident and 5 patients had vascular pathology. 12 women and 64 men with an average age of 49 years to 90 years. Neither post-transfusional reactions nor adverse effects were found. All patients arrived alive at the corresponding referral Hospital. In 2023 Four Patients were administered 2 gr of fibrinogen and three of them received also gluconate calcium, 10%/10cc, 1 dose. RBC units are changed every 35 days to allow the use of the not-used units in the Hospital after achieving their optimal status.

Summary / Conclusions: Our ten years experience shows that the process designed for the out-of-hospital packed red blood cells transfusion in the HEMS of Castilla-La Mancha can be considered useful and safe for the treatment for critically ill patients. Fibrinogen and Calcium Gluconate concentrates added as a complement of pRBC transfusion protocol have been introduced as a way to prevent and treat coagulopathy in severe trauma bleeding patients. The procedure of collection, conservation, tracking and tests to transport blood units and fibrinogen has demonstrated to keep both, the standard conditions and properties of the product.

P771 | Platelet transfusion in a deficient IgA patient with active bleeding—case report

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Background: Common variable immunodeficiency (CVID) is a rare primary immunodeficiency, characterized by an impaired B cells differentiation, resulting in a deficient immunoglobulin (Ig) production (low serum IgG and low levels of IgA and / or IgM) and dysregulated immune response. Patients with low levels of serum IgA may experience transfusion reaction when requiring blood components. In an emergent situation of active bleeding the management of these patients can be challenging.

Aims: To report the challenge of managing an acute severe bleeding in a patient with CVID and thrombocytopenia, who had a previous severe allergic reaction to platelet transfusion.

Methods: We present the case of a 40-year-old man who was diagnosed in the past with CVID conditioning IgA deficiency (IgA < 5mg/dL) treated with intravenous Ig containing low concentration of IgA (<25mcg/mL), and congenital thrombocytopenia (platelet count < 40 × 10⁹/L; gene RUNX1 mutation). In 2012, prior to a cervical swollen lymph node biopsy, the patient was transfused with platelets and experienced an anaphylactic reaction, which was reversed with corticosteroid administration. Due to his clinical condition the patient was frequently hospitalized with severe respiratory tract infections and hemoptysis due to bronchiectasis. Since he had a previous allergic reaction, apheresis platelet from known IgA deficient donors were given whenever the patient required a platelet transfusion. As IgA deficient platelets were not immediately available, during one of his recent hospitalizations we selected random apheresis platelet units for dosing IgA levels, although these levels were higher than those of the intravenous Ig (76.5 and 45.6 mg/dL). During the last episode, the patient was admitted to the emergency room with active bleeding causing airway obstruction (hemoglobin 8.6 g/dL, platelet count 14 × 10⁹/L). As IgA deficient platelets were not available, 4 mg of recombinant activated coagulation Factor VII concentrate (rFVIIa) were administered. As active bleeding persisted, a life-saving non-deficient IgA platelets transfusion was required, prior to an arteriography (with previous prophylactic corticosteroid and clemastine administration).

Results: The patient was stabilized and remained in the intensive care unit. Five days later, he presented a platelet count < 10 × 10⁹/L; however, a watchful waiting strategy was adopted as he was not bleeding and IgA-deficient platelets were not available. The patient remained stable for a few days until he had a catastrophic massive hemoptysis with airway obstruction, which led to his death.

Summary / Conclusions: Platelet transfusion plays an important therapeutic role in symptomatic thrombocytopenia and in bleeding

prophylaxis in severely thrombocytopenic asymptomatic patients. However, it can be challenging in IgA-deficient patients with active bleeding as they may experience an allergic reaction when receiving non-IgA-deficient platelets. Sometimes there are no readily available IgA deficient donors due to the condition prevalence. In life-saving situations, when IgA deficient platelets are not available a transfusion of non-IgA-deficient platelets may be necessary with prior prophylactic corticoid administration. In these situations, off-label use of rFVIIa may be helpful.

P772 | Transfusion strategy in massive hemorrhage

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Background: Massive haemorrhage is a serious clinic state which if left untreated, can be life-threatening. Current transfusion strategy recommends the early combination of fresh frozen plasma and platelet concentrates with packed red blood cells.

Aims: The aim of our study was to analyse particularities of the request of blood products destined for massive bleeding patients and revealing any non conformities.

Methods: We conducted a retrospective descriptive study realized over two monthes (September and October 2023) concerning the urgent requests for massive blood products transfusion received by the military blood transfusion center to evaluate the compliance of the prescriptions and to describe, according to the requesting services, the nature and the quantity of the blood products prescribed.

Results: We identified 178 requests for massive haemorrhage. 63% of prescriptions come from civilians clinics. The departments with the highest demand were: anaesthesia and intensive care (44%), general surgery (12%), gynaecology (5%) and urology (3.5%). A life-threatening emergency was mentioned in 13% of cases. The sex ratio of patients was 1.4, with an average age of 54. Request unconformities consisted in non mentionnig pre-transfusion haemoglobin. Infact only 17% of requests from military hospitals and 79% of those from private clinics mentioned pre-transfusion haemoglobin with an average of 6.8 gr/dl. Moreover, the early association of plasma transfusion was observed in only 22% of cases and platelet transfusion in 3% of cases.

Summary / Conclusions: Haemorrhagic shock must be treated without delay. It is in this context that the Transfusion Center should be able to satisfy quantitatively and qualitatively urgent request by making available haemorrhage packs aiming to standardising the massive transfusion strategy.

Clinical transfusion—adverse events, including TRALI

P773 | Adverse events during preoperative autologous blood donation and its usage in obstetrical patients—a multicenter, retrospective study in Japan

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Background: To evaluate the safety of preoperative autologous blood donation (PAD) and its transfusion is important, especially for young pregnant donors, in the era of an extremely low incidence of viral transmission.

Aims: Adverse Events (AEs) at all steps from donation to transfusion of PAD were collected, and the benefit and safety of PAD and its transfusion were evaluated in obstetrical patients.

Methods: A questionnaire was sent to hospitals, and safety information from donation to transfusion of 2378 obstetric PAD donors was collected and analyzed. Wilcoxon rank sum test was used to compare statistical difference between continuous variables. Bowker test was used to analyze association between VVR between at 1st donation and second donation. To determine risk factors of VVR multivariate logistic regression analysis was used.

Results: A total of 1664 patients received autologous blood only (autologous group), 146 patients received both autologous and allogeneic blood (allogeneic group), and 568 patients received no transfusions (no transfusion group); 91.9% of patients avoided allogeneic transfusion. The vasovagal reflex (VVR) occurred in 63 of 2378 patients (2.6%). Patients who experienced VVR at first donation was associated with VVR at second donation ($p = 0.0077$). Age and multiple babies were detected as statistically significant factors for VVR with multivariate logistic regression analysis. All VVR events were mild or moderate in severity. AEs other than VVR at donation developed in 114 patients (4.8%), and they were the main components of all AEs that were unique for obstetrical patients. The 49 mother-related AEs were as follows: 20 patients developed supine hypotensive syndrome; 10 felt sick (nausea, discomfort, headache); and 5 had warning bleeding. Four experienced puncture site complications. Remaining every symptom was one case each. All nine baby-related AEs were cases of decreased heart rate. Eight of them recovered spontaneously or after change of the mother's body position or fluid infusion. Transfusion reactions (TRs) occurred in 29 patients (1.2%). Surprisingly, the number of patients who had TRs per unit was not substantially different between the autologous (0.29 / 100 units) and the allogeneic group (0.39 / 100 units). In autologous group hypertension and fibrile reaction were frequent seen, meanwhile allergic reaction was frequent in allogeneic group, suggested that the pattern of the various symptoms with TRs was quite different between both groups. There is no reported viral transmission. In summary 201 of 2378 patients (8.5%) experienced some AEs during PAD and its transfusion, although most AEs were mild. Even in no transfusion group 8.1% experienced some AEs.

Summary / Conclusions: Considering that most AEs occurred from donation and were specific to obstetrical patients, PAD for obstetrical patients was not very beneficial. Moreover, similar TRs rate between autologous group and allogeneic group could not show priority of autologous blood. PAD and its transfusion should be performed under a sophisticated transfusion management system.

P774 | ABO incompatibility and component irradiation are independently associated with platelet transfusion reaction rate

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Background: Platelet components are characterized as ABO identical (ABOid), minor incompatible (ABOmin), major incompatible (ABOmaj), and bidirectionally incompatible (ABObi). Unit compatibility allocation is based on inventory management to avoid product wastage balanced against the risk of reduced transfusion efficacy and adverse patient outcomes. The impact of platelet compatibility in association with product irradiation (iSDP) or pathogen reduction (prSDP) is unknown.

P774 - Table 1. Reactions related to transfusion as a proportion of total transfusions.

Unit type	Compatibility			Total
	ABOid	ABOmin	ABOmaj	
nSDP	7/3131 (0.22%)	2/648 (0.31%)	10/826 (1.21%)	19/4647 (0.41%)
prSDP	5/1057 (0.47%)	3/228 (1.32%)	3/252 (1.19%)	11/1552 (0.71%)
iSDP	89/10166 (0.88%)	18/2053 (0.88%)	36/2568 (1.40%)	148/15024 (0.99%)
Total	101/14354 (0.70%)	23/2929 (0.79%)	49/3646 (1.34%)	178/21223 (0.84%)

ABOid correlated to 0/42, 0/15, 5/237 (2.11%), and 5/294 (1.70%) for nSDP, prSDP, iSDP, and total transfusions, respectively.

P774 - Table 2. Logistic regression analysis.

		Univariate, Odds ratio (95% CI)
Compatibility	ABOid vs ABOmin	2.19 (0.83, 5.80)
	ABOmin vs ABOid	1.12 (0.71, 1.76)
	ABOmaj vs ABOmin	1.72 (1.05, 2.83)*
	ABOid vs ABOmaj	2.44 (0.99, 6.04)
	ABOmin vs ABOmaj	1.27 (0.50, 3.21)
	ABOmaj vs ABOid	1.92 (1.36, 2.71)*
Unit type	iSDP vs prSDP	1.39 (0.75, 2.58)
	prSDP vs nSDP	1.74 (0.83, 3.66)
	iSDP vs nSDP	2.42 (1.50, 3.91)*

* Maintained significance in multivariate analysis.

Aims: This study aims to determine the combined and independent impact of platelet compatibility and irradiation or pathogen reduction on transfusion reaction rate.

Methods: Retrospective review of all adult platelet transfusions from 2020 to 2022, including all reported adverse reaction evaluations, was performed with logistic regression to evaluate the significance of ABO compatibility and product treatment on reaction rate.

Results: Out of 21330 transfusions to 3450 patients, 285 (1.33%) reactions were reported and 178 (0.83%) were diagnosed as related to transfusion: 1 acute hemolytic, 1 acute lung injury, 3 transmitted infections, 4 hypotensive, 8 circulatory overload, 59 febrile nonhemolytic, and 102 allergic. Twelve reactions were graded as severe: 1 acute lung injury, 2 circulatory overload, 9 allergic. The compatibility of transfusion was 67.7% ABOid, 13.8% ABOmin, 17.2% ABOmaj, and 1.4% ABObi. Non-treated platelets (nSDP), iSDP, and prSDP were transfused 21.8%, 70.9%, and 7.3% of cases, respectively.

Summary / Conclusions: The results demonstrate a trend of increasing risk of reaction associated with the presence of major incompatibility and product treatment, particularly irradiation, which may be at least partially reflective of the population morbidity. The distribution of transfusions across compatibility and product treatment category was independent, but the effect on reaction rate was compounded by subcategory. This effect was not proportional so that the nominal risk of an nSDPmaj was greater than iSDPid. Interestingly, ABOmin showed no comparative impact for iSDP transfusions, and the only acute

hemolytic reaction was non-severe with an O-Rh negative iSDP transfused to an A-Rh positive patient. The increased risk for prSDPmin may be attributable to the use of platelet additive solution, although the total number is small. Further research may expand on these associations to support their use as an adjunct consideration in choosing product compatibility, given individual patient product treatment needs.

P775 | Abstract withdrawn

P776 | Pulmonary complications of transfusion in the haemovigilance system of Hospital Infanta Sofia (Madrid) from 2019 to 2023

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Background: Transfusion-associated circulatory overload (TACO) and transfusion-related acute lung injury (TRALI) are serious adverse transfusion reactions. TACO is the most common respiratory adverse event associated with transfusion and is the leading cause of transfusion-related mortality according to both the UK Serious Hazards of Transfusion (SHOT) and the US Federal Drug Administration (FDA).

Aims: The aim of this study was to analyze the pulmonary complications reported to the haemovigilance system of Hospital Universitario Infanta Sofia (Madrid) during the last five years and identify patients risk factors to try to prevent TACO.

Methods: Retrospective analysis of all adverse reactions reported to the haemovigilance system to identify pulmonary complications between 2019 and 2023. We analyze age, sex, product transfused and in the cases of TACO the level of haemoglobin prior to transfusion, previous history of congestive heart failure, chronic obstructive pulmonary disease and renal impairment.

Results: During this five year period a total of 27132 red blood cells, 2783 units of platelets and 1555 units of fresh frozen plasma were issued. A total of 102 adverse reactions were reported in this period in the haemovigilance system, 35 of them (34.3%) were recorded as pulmonary complications of which 2 DAT (5.7%), 7 TRALI (20%) and 26 TACO (74.2). In the TRALI patients 57% were males with median age of 67 (range 50-93 years). The

product administered was multicomponent in 1 case, platelets in 2 cases and red blood cells in 4 cases. In the TACO patients 54% were females with median age of 81 (range 60-95 years). The product administered was red blood cells in 90% of cases (range 1-3). The median haemoglobin prior to transfusion was 7.6 g/dl (range 4.1-9 g/dl). 15.3% of TACO patients had previous history of COPD, 35% of congestive heart failure and 57% had creatinine clearance <30 ml/min, median 23 ml/min (range 6-86 ml/min). All the TACO cases were detected because the active surveillance of transfusion process by Haemovigilance Nurse.

Summary / Conclusions: The real TACO incidence is difficult to ascertain due to under-recognition and under-reporting. In our hospital all cases of TACO were detected by Haemovigilance Nurse during the review of the medical chart that we carry with all transfused patients. In our study we detect some risk factors for TACO as: female sex, older age, chronic heart failure and renal impairment. We advocate for the widespread use of pretransfusion checklist to detect patients at risk. Clinical decision support systems incorporating hemodynamic parameters or creatinine clearance could trigger recommendations for diuretic administration or alternatives to transfusion in patients at risk.

P777 | Exposure to cryoprecipitate, but not extracellular vesicles isolated from cryoprecipitate, mediates a small reduction in endothelial cell viability in a laboratory model of TRALI

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Background: Despite the introduction of risk-reduction strategies (e.g. male donors for clinical plasma), transfusion-related acute lung injury (TRALI) remains one of the more frequent causes of transfusion-related morbidity and mortality. Cryoprecipitate is used for patients with fibrinogen deficiency or dysfibrinogenaemia. In Australia, as it is derived from fresh frozen plasma (FFP) prepared from male donors, there should be minimal risk of TRALI. However, cryoprecipitate contains extracellular vesicles (EVs), and previous studies have shown that EVs from red cell and platelet units can cause TRALI in laboratory and mouse models.

Aims: To use an in vitro model of human pulmonary microvascular endothelial cell (HLMVEC) cytotoxicity to assess the risk of TRALI from cryoprecipitate and EVs isolated from cryoprecipitate.

Methods: EV isolation and enumeration: Cryoprecipitate units ($n = 5$) were thawed, pooled, aliquoted and re-frozen. Aliquots of the pooled cryoprecipitate were thawed, centrifuged twice, and underwent size-exclusion chromatography (Izon qEVoriginal 70 nm; Izon automated fraction collector). Collected fractions were concentrated to 0.1 mL (Amicon Ultra-15 filter). The presence of EVs in the collected fractions

was confirmed by western blot (CD9, CD81, and FLOT1) and cryogenic transmission electron microscopy (cryo-TEM), and EVs were enumerated and sized by nanoparticle tracking analysis. HLMVEC cytotoxicity assay: For each experiment ($n = 9$), confluent HLMVECs were treated with 2 µg/mL E. coli lipopolysaccharide (LPS) and cultured for 6 hours (37°C, 5% CO₂). Freshly isolated neutrophils were added (1:10 neutrophil:HLMVEC ratio). HLMVECs and neutrophils were either left untreated or were treated for 30 min with either the pooled cryoprecipitate (1.25% or 2.5% v/v approximating 2 or 4 units of transfused cryoprecipitate respectively) or the isolated EVs (equivalent numbers to those added for 1.25% or 2.5% v/v pooled cryoprecipitate). HLMVECs underwent trypan blue staining, 3-5 fields per well were acquired with Olympus microscope with 10X objective, and viable HLMVECs were identified by ImageJ analysis. **Statistical analyses:** For each experiment, HLMVEC viability was normalised to the LPS-only control well. Data were analyzed with repeated measures analysis of variance (ANOVA, one-way for HLMVEC cytotoxicity followed by Tukey's post hoc test). $p < 0.05$ was considered significant.

Results: The successful isolation of EVs was confirmed by presence of CD9, CD81, and FLOT1 proteins by western blot, as well as observation of circular particles with a membrane bilayer upon cryo-TEM. The isolated EV fraction contained 1.12×10^{10} particles/mL with an average mode size of 216nm compared to 6.75×10^{10} particles/mL and 171 nm in the pooled cryoprecipitate. In the TRALI model, exposure to 2.5% cryoprecipitate resulted in a small but significant reduction in HLMVEC viability compared to the LPS-only control (91% vs. 100% viability; $p = 0.0016$). In contrast, exposure to 1.25% cryoprecipitate or either concentration of the isolated EVs did not reduce HLMVEC viability compared to the LPS-only control.

Summary / Conclusions: In this two-hit laboratory model of TRALI, exposure to a concentration of cryoprecipitate (2.5%), approximately equivalent to transfusing 4 units, resulted in a small reduction in HLMVEC viability. However, exposure to a lower concentration of cryoprecipitate (1.25%) or the equivalent number of EVs, did not impact HLMVEC viability.

P778 | Abstract withdrawn

P779 | Significance of HLA-antibody detection in febrile non-haemolytic transfusion reaction reported in Singapore General Hospital

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Background: Febrile non-haemolytic blood transfusion reactions (FNHTR) are mainly associated with anti-HLA antibodies. FNHTR may occur without a measured fever if chills and/or rigors are the only presenting symptom.

Aims: We study the detection rate of anti-HLA antibody (IgG) among patients reported for FNHTR at Singapore General Hospital (SGH) in

Singapore, its relationship to the presenting symptoms and the subtypes of anti-HLA antibody Class I and Class II.

Methods: A total of 112 cases of voluntarily reported FNHTR to the Blood Bank at SGH over a 4-year period from January 2020 to December 2023 were studied retrospectively to identify their presenting symptoms such as fever, chills/rigors alone or both fever and chills/rigors. They consisted of 73 patients receiving red cell units and 39 patients receiving other products (platelets 29 units and frozen plasma 10 units). Red cells were all buffy coat removed and platelets were all leuco-reduced units. FNHTR is defined as occurring within 4 hours of transfusion and involving either a fever of at least 38 degrees Celsius and a change of at least 1 degree Celsius from the pretransfusion temperature, or chills and/or rigors. Detection of anti-HLA antibody (IgG) is by flow cytometry on blood samples collected for FNHTR and reported as Class I or Class II detected if the panel reactive antibody (PRA) is >3%. Class I anti-HLA antibodies bind both T and B cells, while antibodies against Class II antigens usually bind only B cells.

Results: Of the 112 cases of red cells and other products with symptoms of FNHTR received for anti-HLA antibody testing, 67 (60%) were detected positive with Class I or II or both. On further categorisation of these cases based on symptoms, 42 had fever only, 37 had chills/rigors only and 33 had fever and chills/rigors. Positive HLA-antibody was detected on 23 out of 42 (55%) for fever only, 26 out of 37 (70%) for chills/rigors only and 18 out of 33 (55%) for both fever and chills/rigors. This result showed that FNHTR can occur without a measured fever if chills and/or rigors are the only presenting symptom. In fact, chills/rigors only had the highest anti-HLA antibody detection rate of 70% compared to the other 2 categories of symptoms with 55% each. The anti-HLA antibody positive detection rate for cases of red cells was 46 out of 73 (63%) with 63% being Class I, 9% Class II and 28% both Class I and II. For cases of other products, the anti-HLA positive detection rate was 21 out of 39 (54%) with 52% being Class I, 10% Class II and 38% both Class I and II.

Summary / Conclusions: This study highlights the importance of identifying and reporting the symptom of chills/rigors only, even without fever, in a blood transfusion reaction which will give the highest detection rate of positive anti-HLA antibody in confirming FNHTR. Cases of red cells and other products transfusion with positive HLA-antibody were mostly due to Class I while those with other products transfusion had higher combined Class I and II than the red cell transfusion. Further studies are required to elucidate its underlying pathophysiology.

P780 | Immunization rate in RhD-negative patients transfused with RhD-positive red cell concentrates

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Background: In relation to the management of red cell concentrates (RCC) stocks in blood banks, in situations of shortage, transfusion of RHD-positive RCC to RHD-negative recipients can be considered when the risk of immunization is relatively low, e.g. elderly patients. The Blood and Tissues Bank of Aragon (Spain) had supplied 38,000 RCC in 2023 to 19 hospitals. The RHD antigen is the most immunogenic of the non-ABO blood group antigens. However, the risk of Anti-D immunization depends on patients' individual factors (age, immunocompetence, associated pathology, inflammatory status, genetic predisposition) and blood component characteristics (red blood cell count and other circumstances).

Aims: The aim of this study is to determine the immunization rate among RHD-negative recipients transfused with RHD-positive RCC and to assess the characteristics of patients who have developed Anti-D antibodies.

Methods: A retrospective analysis of RHD-negative recipients transfused with RHD-positive red blood cell concentrates during 2022 was performed in our transfusion network. Age, gender, number of RCC received, underlying pathology, possible Anti-D immunization in cases requiring new transfusion needs in 2022 and 2023 and the possible concomitant appearance of other anti-erythrocyte antibodies were recorded.

Results: A total of 210 patients were transfused (mean age 85.33 years, range 52-104), 112 men (mean 83.03 years, 52-98), and 98 women (mean 88.27 years, 65-104). A total of 612 RCC were administered, with an average of 2.91 per recipient (range 1-70). Anti-D immunization was detected in 20 patients (9.5%). The additional presence of Anti-C +E was identified in two of them, and Anti-c+S in one. The presence of Anti-D was not detected in 67 cases (31.9%). In 123 patients (58.6%) an immunization control was not performed because no new transfusion was required in the period 2022-2023. In the 87 patients who underwent anti-D immunization control, Anti-D was detected in 22.98% of cases. The immunized patients have an average age of 84.6 years (range 59-104), with 15 males and 5 females. They have received an average of 2.05 red blood cell concentrates (range 1-4). Ten patients had neoplastic disease, eight had haemorrhagic gastrointestinal disease and two had cardiovascular pathology. Eight patients had associated chronic kidney disease.

Summary / Conclusions: In advanced age RHD negative patients, who had been transfused with RHD positive RCC, we found an immunization rate of 22.98% of the cases in which a follow-up had been performed. It should also be taken into account that 58.6% of the cases did not require a new transfusion, so that Anti-D immunization was of clinical significance in only 9.5% of the patients.

P781 | Basophil activation test using basophils from children with positive peanut- or buckwheat-specific immunoglobulin E and donor serum after ingesting the corresponding foods

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Background: There have been instances of peanut-allergic patients experiencing allergic symptoms following blood transfusions from donors who had consumed peanuts. This raises concerns about the potential induction of allergic reactions in transfusion recipients with food allergies, depending on the donor's intake of the corresponding food prior to blood donation. A prior report from our team highlighted the activation of basophils in children with food allergies by the serum of donors who consumed the corresponding food. This phenomenon is particularly prominent in cases with high levels of allergen-specific immunoglobulin E (IgE) and holds promise in understanding the pathogenesis of allergic transfusion reactions. However, the analysis in our previous study only encompassed three foods (egg, milk, and wheat).

Aims: The objective is to analyze a basophil activation test (BAT) using basophils from children positive for food-specific IgE (excluding egg, milk, and wheat) and donor sera post-ingestion of the corresponding foods, and validate the occurrence of a similar phenomenon observed in our earlier study.

Methods: Healthy volunteers consumed either 125 g of peanuts or 350 g of buckwheat noodles. Blood samples were collected before and after consumption, and serum was isolated. BAT was conducted using the obtained sera and peripheral blood from children positive for peanut- or buckwheat-specific IgE.

Results: Among the participants, ten tested positive for peanut-specific IgE and nine for buckwheat-specific IgE. One case with peanut-specific IgE displayed a positive BAT result in serum collected 8 h post-peanut ingestion compared to pre-ingestion. Conversely, three cases with buckwheat-specific IgE showed positive BAT results in serum collected 2 or 4 h after consuming buckwheat noodles.

Summary / Conclusions: In children sensitized to peanuts or buckwheat, basophils exhibited activation in the blood after ingestion of these foods. However, even in patients with a documented history of anaphylaxis, BAT did not consistently yield positive results. This variability may stem from differences in sensitivity among cases and variations in the specific allergens targeted in each case. Additionally, further studies are needed to determine the actual amount of food-derived allergens present in donor blood.

P782 | Clinical profile of patients with severe transfusion reactions in Baptist Hospital Mutengene, Cameroon

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Background: Blood transfusions like any other medical procedure is not risk-free. Hemovigilance seeks to capture undesirable outcomes in the transfusion chain from donor to recipient. Thus, they have been categorized into various severity taking into account several parameters such as; requiring treatment, in-patient hospitalization, disability, life threatening, or death. Severe transfusion reactions incorporate life-threatening or death. To mitigate these, understanding the pattern of occurrence among patients in our context is important.

Aims: We aimed to describe the clinical characteristics of patients who had severe transfusion reactions at Baptist Hospital Mutengene, Cameroon. A secondary outcome was to evaluate the proportion of inappropriate transfusions among these events.

Methods: This was a retrospective observational study from January 2022 to December 2023. Data collected from the patient consisted of in-patient ward, gender, age, history of transfusion, indication for transfusion, pre-transfusion hemoglobin, type of blood component, blood group, blood volume, and clinical diagnosis. This was entered and analyzed in Epi Info version 7. A p -value < 0.05 was considered statistically significant. The guideline of clinical use of blood 2021 by WHO and ISBT was used to define the appropriateness of transfusion. Definitions for the various severe transfusion reactions and imputability scores were aligned with that of ISBT.

Results: There were 45 severe transfusion reactions (1.2%) out of 3754 transfusion events. Whole blood transfusions were used in all the transfusion reactions registered. The recipients were predominantly males (61.3%) and had a mean age of 33.1 ± 26.7 years old. 25% had a prior blood transfusion before the reaction and the mean pre-transfusion hemoglobin was 7.36 ± 2.4 g/dL. TRALI (44.4%), TACO (17.8%), and hemolytic transfusion reactions (acute and delayed, 13.3%) were the most common severe transfusion reactions recorded. We found a 28.8% mortality rate wherein 50% of all TACO and 25% of TRALI cases. The internal medicine department (42.2%) and children's ward (28.9%) had the highest frequency as against maternity (0%). We noticed 10 cases were inappropriately transfused and 3 were due to clerical errors (right blood to wrong patient). In addition, 35.5% of the cases had an imputability score of 4 (certain).

Summary / Conclusions: TRALI and TACO were the leading causes of morbidity and mortality and necessitates urgent interventions such as leukocyte-reduced blood components. Transfusion education is important to improve decision-making in transfusion and respect of transfusion protocol, as they have fatal consequences. Moreover, clinicians should be more vigilant with prescriptions in the internal medicine and children's wards to enhance safety.

P783 | Transfusion reactions in IgA deficient patients—red flags or red herrings?

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Background: IgA deficiency is found in approximately 1:700 of the UK population. Varying degrees of reactions to blood products including anaphylaxis have been reported in those patients and have been theorized to be due to anti-IgA antibodies which makes transfusion advice challenging.

Aims: To review transfusion management in IgA deficient patients, based on clinical evidence from three reported cases of transfusion reactions.

Methods: We report three cases of transfusion reactions due to possible IgA deficiency referred to the Welsh Blood Service during 2023.

Case 1: A 76-year-old female with multiple co-morbidities and a historic record of IgA deficiency, but no anti-IgA antibodies or record of a transfusion reaction. The patient had multiple transfusions of **washed red cells** due to concerns regarding her IgA deficiency. Following re-evaluation, she received a blood transfusion of **standard (unwashed) red cells**, with no adverse effects.

Case 2: A 39-year-old female requiring a blood transfusion following a post-partum haemorrhage, and previously found to be IgA deficient (<0.05g/L). The patient was reported to have suffered a reaction following only 10ml of transfused red cells. The reported reaction was more of a febrile than an anaphylactic /allergic response. Due to a low Hb, a further two units of **standard red cells** were transfused with no adverse reaction. Additional testing confirmed the patient to have IgA deficiency (<0.05g/l) with Anti-IgA antibodies.

Case 3: A 66-year-old male who received red cell transfusion following a Coronary Artery Bypass Graft, developed a profound hypotensive event within a few minutes of the first AND second red cell transfusions. Subsequently he was transfused with **washed red cells** to a post transfusion Hb of 122g/dl despite Patient Blood Management (PBM)

advice. No reaction was reported with washed red cells. Additional laboratory tests confirmed IgA deficiency with anti-IgA antibodies.

Results: Cases 1 and 2 could both be described as 'red herrings' as both patients safely received standard red cell transfusion without reaction. Case 3 suggests anti-IgA antibodies as the potential cause of transfusion reaction.

Summary / Conclusions: Many cases of transfusion reaction due to suspected IgA deficiency are complicated, with unclear history, incomplete test results and conflicting previous advice. This makes transfusion management of IgA deficient patients a challenging task, despite clear National and local guidelines. Utilising a PBM approach will reduce the need for blood transfusion, further reducing the risk of a transfusion reaction in these patients.

P784 | Concordance in the severity of hemolytic transfusion reactions and hemolytic disease of the fetus/newborn across antibody-antigen specificities

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Background: Antibodies to red blood cell (RBC) antigens have the potential to cause both hemolytic transfusion reactions (HTR) and hemolytic disease of the fetus and newborn (HDFN). Antigen characteristics which might predict for the development of HTR and/or HDFN are not fully understood. Manifestation with one adverse outcome (HTR or HDFN) may raise the question of the odds of experiencing the other (HDFN or HTR respectively) on antigen re-confrontation. Herein we explore published concordances in antibodies to specific antigen targets for the occurrence and severity of associated HTR versus HDFN.

Aims: For antibodies specific to a target red cell antigen, evaluate published the concordance of HTR and HDFN.

P784 - Table 1: Number of antigens in each cross-category of reaction severity. Antigen counts are provided separately for Low (<1%), Moderate (1-95%), and High (>95%) prevalence antigens.

Acute or Delayed Hemolytic Reaction	Hemolytic Disease of the Fetus and Newborn			
	None	Mild	Moderate	Severe
None	Low: 1 Moderate: 3 High: 6	Moderate: 1	Low: 3	Low: 3 Moderate: 1 High: 1
Mild	Low: 2 Moderate: 3 High: 8	Moderate: 3 High: 3	-	Moderate: 1 High: 2
Moderate	Moderate: 7	Moderate: 1 High: 3	Low: 1 Moderate: 2 High: 1	Low: 2 Moderate: 2 High: 4
Severe	High: 2	Moderate: 1 High: 2	Low: 1 Moderate: 3 High: 2	Low: 3 Moderate: 3 High: 2

Methods: A natural language processing algorithm was used to scrape antigen information pages at BloodAntigens.com (Lane WJ, Lancet Haematology, 2018) for antigen data using R version 4.2.2 and the stringr package. Manual spot checks were performed for accuracy. Antibodies to specific antigens were cross-tabulated according to reaction severity. Heatmap visualization and correlations were performed using the ggplot2 and dplyr packages.

Results: The database contained 216 unique red cell antigens of which 86 (40%) were a low prevalence under 1%, 114 (52%) were moderate prevalence between 1 and 95%, and the remaining 16 (8%) were high prevalence. In our dataset, 86 antigens (40%) had data available for both HTR and HDFN. In cross-tabulation analysis, 31 antigens (36%) showed discordance, such that 22 antigens (26%) were reported for HTR but not HDFN, while 9 antigens (10%) were reported for HDFN but not HTR (Table 1). For the remaining antigens, clinical information was unavailable or unknown in the database.

Summary / Conclusions: We provide a summary of the dual propensity for HTR and HDFN for published antibodies with this information. Our findings might inform the counselling of those with child-bearing potential after HTR, and reinforce matching paradigms for RBC transfusion after HDFN. Further research is needed to validate and inform concordance odds and to explain the biology of non-intuitive discordances.

P785 | Appropriateness of management of acute adverse transfusion reactions at the bedside

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Background: Transfusion reactions may be difficult to diagnose and manage as they can present with non-specific overlapping symptoms and may have life threatening consequences. All cases of suspected reactions should prompt immediate discontinuation of the transfusion and treatment guided by the predominant sign and symptom till diagnosis is reached. It is thus important to recognise the predominant signs and symptoms so that appropriate management is provided. There is scant literature covering the bedside response of transfusion reaction and appropriateness of the response. The present study aimed to learn response of the bedside staff towards transfusion reactions and appropriateness of the treatment provided.

Aims: To assess the appropriateness of management of acute adverse transfusion reactions (AATR) at the bedside.

Methods: Retrospective observational study conducted in Department of Transfusion Medicine of tertiary care institute from north India. Transfusion instances (where blood component was issued & transfused) and AATR reported over period of one year were included. TR details including bedside management were obtained from Transfusion Reaction Reporting Form (TRRF) and transfusion reaction work-up registers. Transfusion reaction rates (per 10000 blood component) were calculated and component wise, age wise and gender wise differences were analysed using chi-square test. A *p*-value of <0.05 was considered significant. Transfusion reaction

management guidelines as recommended by the Hemovigilance Program of India (HvPI) were used to assess appropriateness of the treatment given. Any additional recommendations from AABB and BSH were also included and considered appropriate.

Results: A total 108889 blood components issued and transfused during study period that included 30777 packed red blood cell (PRBC), 16793 leukoreduced PRBCs, 42693 platelet concentrates (PC-WB), 15379 fresh frozen plasma (FFP), 474 cryoprecipitates, and 2774 single donor platelets (SDP). The overall rate of AATR was 6.6 per 10000 issued and transfused blood products. Statistically higher rate of AATR (*p* < 0.001) was observed with PRBC (11.9 per 10000 blood products) as compared to PC and FFP (0.7 vs 3.2 per 10000 of blood products respectively). Statistically higher rate of AATR was observed when blood component was transfused in female patients as compared to male patients (8.6 vs 4.3 per 10000 blood products *p* < 0.05). Rate of AATR among paediatric and adult recipients was 6.6 vs 5.7 per 10000. Most common AATR was Allergic transfusion reaction (2.2 per 10000 blood products). Almost all adverse transfusion events were managed by immediate discontinuation of transfusion along with medical management which includes Inj. Pheniramine Maleate I.V. (antihistaminic) and Inj. Hydrocortisone Sodium Succinate 100 mg (corticosteroid) which was assessed to be inappropriate. Management done in only 7.6% was deemed appropriate as per the published recommendation.

Summary / Conclusions: The AATR rate in the present study was comparable with the national rate reported by HvPI. PRBC (cellular) components and transfusion in female patients was associated with higher rates of AATR. The study highlights the lack of knowledge of the bedside staff regarding immediate management of AATR. Training programs covering symptom based management of AATR at bedside may help bridge this gap and increase the transfusion safety. The study was limited by the fact that the TR reporting was passive and mild reactions may not have been reported.

P786 | Audit on transfusion related adverse events in Colombo South Teaching Hospital, Sri Lanka

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Background: Transfusion therapy is a critical component of modern healthcare, providing a lifeline for patients in need of blood or blood products. Currently there is growing recognition of adverse events associated with blood transfusions which vary from mild reactions to serious life-threatening complications resulting in morbidity and mortality. These adverse events could be acute, occurring within 24 h of the transfusion or delayed, which occur 24 h after blood and blood product transfusion. Rational use of blood and blood products has been shown to reduce these adverse events while improving patient outcome. Therefore, it's the responsibility of the clinical team to properly report the adverse events related to transfusion and to involve

the Transfusion Medicine team for appropriate management of the patient.

Aims: This audit aims to determine the types and frequency of transfusion reactions, to assess the patterns of management by the clinical teams and to identify the pre transfusion thresholds for blood and blood product transfusion

Methods: This audit was conducted in Colombo South Teaching Hospital from August 2023 to January 2024. Data was obtained from transfusion related adverse event forms, blood grouping and cross-match requests and bed head tickets.

Results: During the six-month period a total of 10,429 blood and blood products were issued with 93 (0.89%) transfusion reactions reported. All reactions reported were acute reactions. Among the affected patients, 59 (63.4%) were females and 34 (36.6%) were males with a mean age of 53.1 years (Standard deviation (SD) 19.83). Among the transfused products red cell concentrates, 87.1% were responsible for the highest number of reactions followed by fresh frozen plasma (FFP) 7.5%, platelet concentrates 3.2% and cryo-supernatant plasma (CSP) 2.2%. Mean hemoglobin (Hb) level among the patients who were transfused with red cell concentrates was 7.28 g/dL (SD 1.42) with 22 (27.1%) patients being transfused to a hemoglobin of more than 8g/dL. Among the reported transfusion reactions, commonest was febrile non haemolytic transfusion reactions (FNHTR) 46 (49.5%) followed by minor allergy 31 (33.3%), anaphylaxis 5 (5.4%), transfusion associated circulatory overload (TACO) 5 (5.4%) and 1 (1.1%) transfusion associated dyspnoea (TAD). 2 ABO incompatible transfusion reactions occurred in the given period. Out of the 93 reactions reported 3 were not related to transfusion. 20 (35.1%) patients without allergy/anaphylaxis received intravenous chlorpheniramine and hydrocortisone. Increase in FNHTR could be attributed to the lack of universal leukodepletion facilities in the country.

Summary / Conclusions: This audit highlights the importance of reporting transfusion related adverse events as a part of the haemovigilance system which will provide feedback to the clinical team. It will increase staff awareness on the types of transfusion reactions, the susceptible patient population and appropriate management of transfusion related adverse events. Therefore, it is recommended to report both acute and delayed transfusion reactions, avoidance of unnecessary administration of chlorpheniramine and hydrocortisone and encourage evidence-based blood and blood product transfusion while adhering to patient blood management protocols.

P787 | Abstract withdrawn

P788 | Incidence of febrile non haemolytic reaction within transfused patients

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Background: The administration of blood products is most commonly linked to transfusion reactions, which encompass a spectrum of adverse

events ranging from mild to severe, potentially life-threatening occurrences. Febrile Nonhemolytic Transfusion Reactions (FNHTRs) stand out as the predominant acute transfusion reactions in terms of frequency.

Aims: This review aims to showcase the occurrence of FNHTRs during transfusions and to notify healthcare professionals about this specific transfusion-related occurrence in a healthcare facility of the third level in a low-income economic country.

Methods: Observational study on the incidence of Febrile Nonhemolytic Transfusion Reactions from 2007 to 2023. Data were collected from patients who underwent transfusions with labile blood products during this period within the Hemovigilance Center at La Rabta Hospital.

Results: Adverse reactions were recorded among 312 patients. Within them, 60% (187 patients) of FNHTRs were suspected. Out of all FNHTRs under suspicion, 31% were validated following an etiological inquiry, while 69% were classified under other diagnoses. Febrile Non-hemolytic Reactions were observed in 38% of platelet transfusions, 51% of red blood cell concentrate transfusions, and 11% of fresh frozen plasma transfusions. The incidence of FNHTRs differs depending on the type of platelet product used. It is higher (0.43% of transfusions) with nonleukoreduced products compared to a lower range of 0.1% with leukoreduced platelet products. Nonhemolytic febrile reactions also vary based on the leukoreduction of red blood cell concentrates; 0.27% for non-leukoreduced versus 0.09% for leukoreduced products. 46.6% of FNHTRs occurred during the transfusion, while 53.4% manifested between 30 min and 2 h after the transfusion. Women with a reproductive background (5.3% of the FNHTRs observed) and those who underwent numerous blood transfusions (1.8% of the FNHTRs observed) had a higher likelihood of encountering FNHTR.

Summary / Conclusions: FNHTR boasts the highest occurrence among all transfusion reactions, imposing significant challenges on both patients and healthcare systems. Leukoreduction of blood products before transfusion significantly reduces nonhemolytic febrile reactions. Understanding the importance of being vigilant regarding febrile non-hemolytic reactions is essential to promptly identify, manage appropriately, and enhance patient safety throughout transfusion procedures.

P789 | Alloantibody production in thalassemia patients is still a worrying issue

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Background: Although transfusion is a crucial treatment in most of patients, there are some serious complications which more affected chronic recipients for instance whom with thalassemia major and intermedia. One of risky complications is alloimmunization that reduce patients' quality of life. So, identifying alloimmunization prevalence and their phenotyping is helpful for early patient's management.

Aims: The aim of this study was evaluating the frequency of alloantibody production in thalassemia patients with transfusion dependency.

Methods: The present study was conducted on 100 transfusion-dependent thalassemia major patients who had more than 20 times transfusion events. Antibody screening and identification tests were performed using the tube method.

Results: The studied patients were 52% female and 48% male. The most prevalent detected alloantibodies were Anti-K (47%), Anti-E (33%), Anti-C (15%), Anti-D (13%), Anti-c (8%), Anti-e (5%), Anti-Kpa (4%), Anti-S (3%), Anti-Fya (2%), Anti-Jka (2%), Anti-k (1%), Anti-Jkb (1%), respectively. Secondary analysis showed that 11% of patients had only one alloantibody, 32% had two alloantibodies, and 7% had equal or more than 3 alloantibodies.

Summary / Conclusions: According to our study, despite advance progressions in transfusion medicine procedures, services, and patients' management, there is a high prevalence of and Kell and E alloantibodies. So, patient's phenotype and genotyping at the initiation of chronic transfusions are seriously recommended, as well as studies regarding critical alloimmunization predictors to effective patients' management to overcome this challenging complication.

P790 | Overview of the adverse transfusion reactions in the Institute for Transfusion Medicine of Republic of North Macedonia

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Background: Although blood transfusions are generally safe because of the strict transfusion safety measures, recipients of blood or blood components can experience transfusion related adverse reactions. In order to improve the quality of the whole transfusion chain, every undesirable event or reaction of the blood donation and transfusion must be recognized, reported, analyzed and corrective and preventive measures should be taken as part of the haemovigilance.

Aims: The aim of this study was to analyze the reported adverse transfusion reactions (SAR) in the Quality Assurance and Quality Control (QAQC) Department of the Institute for Transfusion Medicine of Republic of North Macedonia – Skopje (ITM).

Methods: This is observational retrospective analysis of the serious adverse reactions that were reported to the QAQC department for the period from 2007 till 2023.

Results: In the last seventeen years there were reported total of 33 serious adverse reactions due to transfused blood components: in 2007 there were 2 reported adverse transfusion reactions (Cryoprecipitate and Fresh Frozen Plasma (FFP)), in 2008- 1(FFP), in 2009 - 1, in 2010 -1(FFP), 2011 - 3 (2 (RBCSAGM and 1 FVIII conc.), in 2012 - 1 (Cryoprecipitate), in 2013- 2 (RBC-SAGM and Cryoprecipitate), in 2014 - 2 (LD RBC-SAGM and Cryoprecipitate), in 2017 - 2 (RBC-SAGM), in 2018 - 4 (1-FFP and 3-RBC-SAGM), in 2019 - 3 (FFP), in 2020 - 4 (1-Platetelet concentrate, 3 RBC-SAGM), in 2021 - 1 (RBC-SAGM), in 2022 - 4 (RBC-SAGM) and in 2023 - 2 (RBC-SAGM)). The most common signs and symptoms that were reported

in the special forms for reporting SAR were mild allergic and febrile non-haemolytical transfusion reactions that included fever, chills, increased body temperature, urticaria, rash and vomiting, just a few of them were moderate and severe with headache, dyspnoea, hypotension and tachycardia. There was no respiratory distress nor mortality associated with blood transfusion in the last 17 years.

Summary / Conclusions: Obviously the SAR are under-reported in ITM, that can be due to various reasons. Monitoring the SAR gives us data and helps us to understand the effectiveness of the transfusion safety measures. They can be used as a quality indicator and together with the clinical strategies can reduce the probability of reactions and improve patient outcomes. Hospital transfusion committees have extremely important role in functional haemovigilance which is integral part of the total quality system in the whole transfusion chain.

P791 | Delayed type hyperhaemolysis syndrome in a patient with sickle cell disease—a case report

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Background: Sickle cell disease (SCD) is an inherited haemoglobinopathy with chronic haemolytic anaemia due to defective β -globin synthesis. Blood transfusion can be lifesaving and reduces disability in the management of SCD. However, it can cause complications, including allo-immunisation, iron overload, delayed hemolytic transfusion reaction (DHTR) and hyperhaemolysis syndrome (HHS).

Aims: Case report. An 18 year old male is a diagnosed patient with HbSS SCD. He was on hydroxyurea and folic acid and was transfusion dependent. He had a history of DHTR in 2012 and found to have anti-c and anti-Jk^b. This time he presented with features of anaemia and painful sickle crisis to a local hospital with haemoglobin (Hb) of 50g/L and was given red cell transfusions. Despite this he was complaining of worsening anaemia, fever and dark red colour urine and Hb was dropped to 64g/L. His clinical condition was further deteriorated and was transferred to a tertiary care center for ventilator support and further management. Direct agglutinin test (DAT) was positive with both anti IgG and anti C3d specificity and a new antibody, anti-S, was detected in addition to anti-c and anti-Jk^b. ABO group specific, Rh and Kell matched, Jk^b and S antigens negative, IAT crossmatch compatible red cell unit was transfused. However, patient continued to have low Hb and with elevated LDH and reticulocytopenia, HHS was suspected. Further two red cell transfusions were given following IV immunoglobulin (IVIg) 1g/kg for 2 days and IV Methylprednisolone 500mg/day for 3 days. With these measures his Hb improved to 103g/L.

Methods:

Results: HHS is a rare, potentially life-threatening hemolytic transfusion reaction characterized by a lower Hb than pre-transfusion. Hemolytic features with reticulocytopenia favors the diagnosis of HHS. There are two types of HHS, acute and delayed. Delayed type HHS is the most likely diagnosis in this patient as a new antibody was identified 8 days after the initial transfusion with a positive DAT. Differentiation

between DHTR and delayed type HHS is quite challenging. Awareness and early recognition of HHS is important to reduce morbidity and mortality. Transfusion of antigen negative crossmatch compatible blood may not prevent the occurrence of HHS. Management of these patients depends on severity of anaemia and rate of hemolysis. Transfusions should be avoided as much as possible to minimize the hyperhaemolysis. If RCC transfusions are required in rapid severe hemolysis as in this patient, transfusion along with IVIG and steroid is beneficial.

Summary / Conclusions: Even though HHS is a rare complication, anticipation, clinical suspicion and appropriate investigations are needed for early diagnosis and better patient outcome. Red cell transfusion should be minimized and given with IVIG and steroid cover if needed.

Clinical transfusion—haemovigilance and patient safety

P792 | ABO incompatible red cell transfusion events and near misses in the United Kingdom; prevention is better than cure

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Background: Serious Hazards of Transfusion (SHOT) is the UK independent haemovigilance scheme. Serious adverse reactions and errors related to transfusion of blood components are reported to SHOT via an electronic confidential reporting system. ABO incompatible (ABOi) red blood cell (RBC) transfusions continue to be reported in the UK. ABOi can be fatal and must be prevented. Where ABOi are detected prior to transfusion these are reported to SHOT as near miss events and provide essential learning potential.

Aims: To review the number of ABOi RBC transfusions and ABOi RBC near miss events in the 6-year period 2017-2022 to identify weaknesses in the transfusion pathway leading to ABOi, and the barriers in place which prevented ABOi.

Methods: Cases accepted by SHOT from 2017 to 2022 were analysed to determine how ABOi RBC transfusions had occurred, and where weaknesses lay during the transfusion pathway. Barriers and preventative measures were identified from ABOi red cell near miss events during the same period.

Results: There were 21 ABOi RBC transfusions during this period, of which two led to patient death, and four resulted in major morbidity in the patient. Primary errors in the clinical area accounted for 19/21, and primary laboratory errors for 2/21. There were multiple errors during the transfusion pathway in 10/21 cases. In total 31 errors occurred in the 21 cases. Clinical errors occurred during administration (17/21), collection (10/21) and sample taking (1/21). The main influencing factor in these errors was incomplete patient identification (PID) processes leading to wrong patient transfused, wrong unit collected, unit delivered to wrong location, and a sampling error which resulted in a wrong blood in tube

event (WBIT). Electronic blood management systems (EBMS) were available in 2/21 cases and pre-administration checklists were used in 4/21 cases, but all failed to prevent ABOi. In 17/21 cases it was reported that the unit was not checked against the patient's wristband. In 2/21 cases units were checked at the nurse's station, rather than at the patient-side. Laboratory errors occurred during testing (2/21) and component selection (1/21). The errors included issuing units to incorrect patient, issuing incorrect ABO group, and ABO misinterpretation during patient sample testing. There were 415 incorrect blood component transfused – wrong component transfused (IBCT-WCT) near miss events during this period, of which 63 could have resulted in an ABOi RBC transfusion. These errors were due to 59/63 clinical errors and 4/63 laboratory errors. Of these 27/63 were detected using information technology (IT) and 36/63 by staff using a pre-administration checking procedure.

Summary / Conclusions: Latent holes during the transfusion pathway can lead to cumulative errors resulting in patient harm. Lack of robust PID processes have the potential to impact on patient safety throughout the whole transfusion pathway but have the largest impact in clinical areas and have resulted in patient death due to ABOi. Assumptions, cognitive bias, and multitasking have been detailed as influencing factors. Preventative measures such as pre-administration checklists and implementation of IT systems such as EBMS can have significant impact on reducing the occurrence of ABOi RBC, with these actions preventing three times the number of ABOi during the 6-year period. These measures must be robust, and not just a tick box exercise, as errors continue to occur where these preventative measures are in place.

P793 | Are we hemovigilant? Blood transfusion reaction documentation and investigation at a tertiary hospital in Sub-Saharan Africa (SSA)

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Background: Blood transfusion (BT) is a critical component of patient management that improves the patient clinical outcomes. However, BT may cause acute or delayed transfusion reactions. To ensure hemovigilance, it is crucial that the reaction incident be accurately documented and a timely Blood Transfusion Reaction (BTR) investigation done. This information improves transfusion practices, minimizes adverse reactions, and enhances patient safety along the hemovigilance chain. A thorough investigation of reactions helps identify errors, enable corrective measures, and prevent future incidents. The ultimate goal of hemovigilance is continuous quality improvement of the transfusion chain through corrective and preventive actions to improve patient safety and outcomes, enhance donor safety, and reduce wastage.

Aims: To investigate transfusion documentation and transfusion reaction investigation practices. To obtain baseline data for Hemovigilance quality improvement (QI) initiatives.

Methods: We conducted a retrospective review of all blood requests made and transfusions done among all the patients at AIC Kijabe Hospital from January 2022 to July 2023. Data was collected from the

MOH Blood Transfusion Register (MBTR) and was compiled into an Excel sheet. We also reviewed patient files from the Hospital Management and Information System (HMIS), to check for documentation of transfusion by either the treating clinician or nurse.

Results: Between January 2022 and July 2023, 2677 blood requests were registered, with 2671 samples (99.8%) deemed compatible. The remaining 6 samples (0.2%) were incompatible hence excluded from the transfusion process. 228 (8.5%) of the requested blood products had no rationale for non-collection documented. Out of 2443 collected blood products, 87 (3.6%) were returned, with 8 units (9.2%) unused and 8 (9.2%) indicating BTR. For 71 (81.6%) of returned products, the underlying cause for return was not ascertained, as documentation was absent in both the HMIS and the Adverse Transfusion Reaction Form (ATRF). Standardized documentation procedures, including ATRF and laboratory investigations, were not consistently conducted across all returned samples, including those with Blood Transfusion Reactions (BTR) indicated.

Summary / Conclusions: Blood product documentation, transfusion reaction documentation, and BTR investigation at AICKH are suboptimal, therefore the standard acceptable hemovigilance practice requires improvement.

Next Steps: To minimize this variability, and improve efficiency, we have initiated a Continuous Quality Improvement (CQI) hemovigilance program. We have adopted the Six-Sigma DMAIC model to strengthen our existing processes, targeting a 95% efficiency in BTR documentation and investigation in one year, with at least 30% improvement every quarter. Current interventions

P794 | The effect of the suspected acute transfusion reaction flow chart in reporting and identifying reactions

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Background: Blood transfusion is a life-saving procedure but also has serious side effects. A transfusion reaction is defined as an undesirable response by the patient to infused blood and blood components. Identifying the reactions that may occur during blood transfusion is an important issue in order to provide the correct intervention.

Aims: Acute transfusion reactions share common signs and symptoms that make classification difficult at onset. Systematic flow charts and guidelines should be used when identifying and responding to transfusion reactions. This study was conducted at Koç University Hospital to detect Acute Transfusion reactions and prepare a flow chart for correct intervention.

Methods: Koç University Hospital The creation of the Approach to the Patient with Suspicious Acute Transfusion Reaction flow chart was put on the agenda by the Transfusion Committee in 2017. The reaction section has been edited in the Blood Transfusion Record and Observation form and all signs and symptoms that raise suspicion of reaction have been added. Literature research was carried out and in line with

the National Hemovigilance Guide, "BCSH Guide for the Investigation and Management of Acute Transfusion Reactions, Guidelines for the Administration of Blood and Blood Components and Australian Red Cross Blood Service, Acute Transfusion Reactions Poster were used." flow scheme has been created. In 2018, a pilot study was carried out in Hematology and KIT services, which have high blood usage, and arrangements were made. It started to be used in November 2018 after training was provided throughout the hospital.

Results: It was observed that nurses' awareness and reporting increased during patient follow-up during transfusion with the use of the approach flow chart to the patient with suspected acute transfusion reaction. While transfusion reactions before the flow chart were evaluated retrospectively, suspected reactions were reported immediately after the use of the flow chart. When a transfusion reaction was suspected, correct diagnosis and standard intervention were provided to each patient. There were 123 suspected reactions between January and September 2019, a 100% increase compared to January-September 2018. Depending on the product used, the probability of a suspected reaction has increased by 25% to 0.98. When the reactions detected in line with the National Hemovigilance Guide were categorized, it was seen that TACO and Anaphylactic reactions did not occur in 2018, while TACO and Anaphylactic reactions were encountered in 4 cases and 2 cases in 2019.

Summary / Conclusions: Correct identification and treatment of transfusion reactions It is effective in reducing transfusion-related morbidity and mortality. When the 5-year data was evaluated, a total of 81,142 blood component transfusions were determined and the prevalence of transfusion-related adverse reactions was found to be 69/10,000 blood components. While the prevalence of reaction development was 61/10,000 in 2018, it increased to 86/10,000 in 2019. In the study conducted by Kar et al., 61,636 units of product were used in two years and the reaction frequency was found to be 0.09%. In the study conducted by Bal et al., the frequency of reaction development was 108/100,000. When compared with other studies, it has been observed that in our hospital, with the use of the flow chart, awareness was raised and the number of reaction reports increased and serious reactions were detected.

P795 | Incidence and severity of transfusion-associated adverse events using an active haemovigilance programme with 24h follow-up. A second period study

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Background: A prior study in our centre revealed that an active haemovigilance (HV) programme with 24h follow-up increases the incidence of transfusion-associated adverse events (TAAEs) 14 times,

P795 - Table 1.

	All*	Transfusions for one event	Severe and deaths*	Transfusions for one severe or death
Confirmed TAAEs	346; 47.86 (42.83-52.89)	209 (189-233)	71; 9.82 (7.54-12.11)	1018 (826-1327)
FAHR	215; 29.74 (25.77-33.71)	336 (297-388)	6; 0.83 (0.17-1.49)	12048 (6693-60281)
PULMONARY	102; 14.11 (11.37-16.85)	709 (594-879)	64; 8.85 (6.69-11.02)	1130 (907-1496)
TACO	77; 10.65 (8.27-13.03)	939 (767-1209)	49; 6.78 (4.88-8.68)	1475 (1153-2049)
TRALI	13; 1.80 (0.82-2.78)	5561 (3603-12183)	10; 1.38 (0.53-2.24)	7229 (4463-19012)
TAD	12; 1.66 (0.72-2.60)	6024 (3847-13873)	5; 0.69 (0.09-1.30)	14458 (7705-117075)
IBCT-WCT	5; 0.69 (0.09-1.30)	14458 (7705-117075)		
Hyperhaemolysis	1; 0.14 (-0.13-0.41)	72289 (24422-75302)	1; 0.14 (-0.13-0.41)	72289 (24422-75302)
UCT	23; 3.18 (1.88-4.48)	3143 (2231-5315)		

*Cases, Rates per 10,000 (CI95%). FAHR: Febrile, Allergic and Hypotensive Reactions. Digestive: Nausea, Vomiting and Diarrhoea. TRALI Transfusion Related Lung Injury. TAD Transfusion-associated Dyspnoea.

compared to a traditional passive HV model. The incidence of TAAEs and mortality with this new model was 57.3 (95% CI, 50.5–64.2) and 1.1 (95% CI, 0.13–2.01) per 10,000 transfusions. (Bueno et al. Transfusion, 2023).

Aims: The aim of this study is to calculate the incidence of TAAEs and severity over a second time period, to validate our active HV programme.

Methods: We monitored a total of 72,289 transfusions in 10,445 patients between January 2020 and December 2023. A trained HV nurse reviewed patients' electronic medical history up until 24 h after each transfusion and recorded any event that might be related. Subsequently, an HV haematologist reviewed the records and assigned a definition, severity and imputability according to the 2024 SHOT reporting categories. Near misses were not analyzed in this study. We used a χ^2 squared test to compare incidences.

Results: Haemovigilance nurses labelled 539 transfusions with at least one suspicious transfusion-related symptom (TRS), of which 149 (27.6%) were ruled out by the blood bank medical review. Finally, 346 TAAEs and five deaths were confirmed. (See Table 1). Mortality incidence was 0.69 (95% CI, 0.09–1.30) per 10,000 transfusions, or one death for every 14,458 transfusions (95% CI, 7705–117075). Compared to our previous study, the incidence of TAAEs was slightly lower ($p = 0.03$) but there were no statistically significant differences in mortality rates. (P : ns). *Uncommon and New Complications of Transfusion* (UCT) were mainly gastrointestinal. We also reported five *Incorrect Blood Component Transfused-Wrong component transfused* (IBCT-WCT), due to identification errors at the time of transfusion and one death because of hyperhaemolysis. *Pulmonary* events occurred well after transfusion, and were mainly severe; (64 out 102; 62.7%) Four out five deaths (80%) were due to pulmonary TAAEs; mainly *Transfusion associated circulatory overload* TACOs.

Summary / Conclusions: Ours study confirms the previous TAAEs incidences in our hospital and validates our active HV programme.

Pulmonary events, especially TACOs remain the most worrying event related to transfusion. External validation of our system in other centres would be desirable to better define the actual risk of transfusion.

P796 | Kleihauer testing—how good do we need to be?

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Background: Kleihauer testing is recommended in New Zealand ("NZ") following birth or potential sensitising events after 20 weeks' gestation to detect large fetomaternal bleed and guide doses of RhD immunoglobulin to prevent anti-D alloimmunisation. To review clinical practice and improve patient safety, the national Transfusion Clinical Nurse Specialist team and lead Transfusion Medicine Specialist re-audited RhD Immunoglobulin Use and Kleihauer Testing in New Zealand.

Aims: To determine the level of Kleihauer testing being completed and RhD Immunoglobulin use and dosing in RhD negative women who gave birth to an RhD positive, or unknown baby, compared with results obtained from a previous clinical audit of RhD Immunoglobulin Use in NZ undertaken by the NZ Blood Service ("NZBS") in 2009.

Methods: Data was collected from the nine largest hospitals, who covered 74.9% of births in NZ at the time. All data around births was collected from the Ministry of Health's National Maternity Collection, who provided a list of hospital identifiers of mothers and babies who had given birth between 1 July 2018 and 30 June 2019. Blood Groups and Kleihauer results were obtained from NZBS records and hospital laboratories. Hospital Kleihauer policies were reviewed and whether flow cytometry was performed. The lead Transfusion Medicine Specialist obtained data from the Ministry

while the Clinical Nurse Specialists obtained laboratory data from their hospitals.

Results: Kleihauer testing remains the principal test used across all NZ hospitals audited, with only two offering HbF flow cytometry only where the Kleihauer test indicated a bleed greater than 2 or 2.4 mLs. 10% of the 40,405 women who gave birth over the course of a year were RhD negative. 68 (1.8% of RhD negative women) already had an anti-D antibody and were excluded from analysis. Two-thirds of RhD negative women had a baby that was RhD positive or RhD unknown. Kleihauer testing was performed in 76% of these, compared with 44% reported in 2009. Across this and the 2009 audit, 3 in one thousand births gave a positive Kleihauer result requiring further RhD Immunoglobulin. Extrapolating from this, potentially one or two women in this audit were underdosed with RhD Immunoglobulin due to lack of Kleihauer testing. 87 RhD negative women who had RhD positive/unknown babies did not receive RhD Immunoglobulin at all at birth and were unprotected from sensitisation. The remaining 96% of women received it perinatally consistent with national recommendations. Despite the high rate of RhD Immunoglobulin administration, the 87 women not receiving RhD Immunoglobulin compares badly with 1-2 potentially sensitised from not having a Kleihauer test. All audited hospitals now have policies in place requiring Kleihauer testing for RhD negative women giving birth to RhD positive or RhD unknown babies.

Summary / Conclusions: Results show a significant improvement in Kleihauer testing especially those sites with previously low rates reported in 2009. The lack of RhD Immunoglobulin administration represents a significantly higher risk for the formation of anti-D antibodies than lack of Kleihauer testing.

P797 | ABO incompatibility red cell transfusions review of Serious Hazards of Transfusion (SHOT) over the last 10 years

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Background: ABO-incompatible (ABOi) red blood cell (RBC) transfusions have the potential to cause severe clinical consequences including patient death. Bedside Electronic Transfusion Checks (BETC) have been introduced in different countries to prevent these serious events at the bedside: however, the benefits of such systems for patients have not been quantified previously. As part of implementing BETC at four hospitals at Barts Health Trust in London, we aim to tackle this gap by quantifying the benefits of BETC for the patients and to achieve this, we need to understand the burden associated with ABOi from RBC transfusion.

Aims: The overall aim of this review is to identify and quantify harms (i.e. morbidity and mortality) associated with ABOi RBC transfusions, so we can extrapolate the benefits for patients.

Methods: Published data from cases submitted to UK SHOT haemovigilance reports were collected for 10 years from 2013 to 2022. Data were extracted by two independent reviewers and collated into a Microsoft Excel database for further analysis. The data was analysed to determine the number of reports and the rate of mortality/morbidity associated with ABOi RBC transfusion.

Results: During the 10-year review period, there were a total of 17,549,938 RBCs issued throughout the UK, with 50 (or 1 in 300,000 RBC transfusions) of these being identified as ABOi RBC transfusions. Of the 50 cases identified, 5 (10%) resulted in patient death and 13 (26%) in major morbidity for the patients. Major morbidity was classified as admission to a critical care unit (6 [46.2%]), red cell exchange (1 [7.7%]), renal failure (1 [7.7%]), intravascular haemolysis (1 [7.7%]), and in 4 (30.8%) cases there were no further details outlined on how major morbidity was classified. Of the 50 ABOi RBC transfusions 41 (82.0%) were associated with clinical errors and 9 (18.0%) were associated with laboratory-based errors. Administration of the component (21 [51.2%]) was identified as the most common clinical step where errors were made, followed by blood collection (16 [39.0%]) and sample collection (4 [9.8%]). Of the laboratory-based errors, component selection (6 [66.7%]) was identified as the most common error followed by sample testing (3 [33.3%]).

Summary / Conclusions: Ten years of data collection showed that ABOi RBC transfusions are rare events, however, are associated with significant short-term morbidity and mortality. There was a noticeable lack of standardisation in the earlier SHOT report highlighted by the fact that the reporting technique for patient outcomes changed from year to year. Data on the long-term outcome of these cases is lacking. National haemovigilance systems across the world should look to harmonise and standardise the short-term and long-term outcome data collection for ABOi RBC transfusions, so we can better understand the burden of these events on patients.

P798 | Safety of amotosalen/UVA platelets and plasma transfused in routine clinical use—real world evidence from 2 large European transfusion medicine centers, 2019–2022

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Background: Pathogen reduced (PR) platelet (PLT) and plasma (PLS) components were approved in Europe in 2002 and 2006, respectively. Cerus, the manufacturer of the amotosalen/UVA technology (INTERCEPT® Blood System) conducts voluntary periodic hemovigilance (HV) studies with centers using the technology to collect real world evidence (RWE) on the safety of INTERCEPT treated

P798 - Table 1.

	Site	Graz	Warsaw	Total
Platelets	Subjects	134	165	299
	Units	1054	1286	234
	TRs	2	3	5
	%TR / units	0.19%	0.23%	0.21%
Plasma	Subjects	7	52	59
	Units	462	719	1181
	TRs	0	2	2
	%TR / units	0.00%	0.28%	0.17%

components transfused in routine clinical use. Previous sponsored HV studies collected safety data on 21,548 INTERCEPT PLTs in 4765 patients and 57,428 INTERCEPT PLS in 9813 patients in 11 countries between 2003-16. A new round of data collection began in 2019. Data collected through November 2022 are presented.

Aims: To document the nature, frequency and severity of transfusion reactions (TRs) in patients transfused with INTERCEPT PLTs and/or PLS in routine clinical practice and compare these findings with historical data.

Methods: Two hospitals in Graz, Austria and Warsaw, Poland participated in this observational, non-interventional, single-arm study. Data related to the use of INTERCEPT PLTs and PLS were collected prospectively from routine hospital and blood bank data systems for all patients transfused in selected wards during defined surveillance periods. De-identified patient data were stratified by sex, age and clinical diagnosis. TRs were defined according to ISBT terms and identified by physicians using hospital reporting systems. Descriptive statistics were calculated for each site.

Results: A total of 2340 PLT and 1181 PLS components were transfused to 299 and 59 patients, respectively. Apheresis accounted for 44% of PLTs transfused in Graz and 88.1% in Warsaw. All PLS components in Graz were from apheresis; >97% of PLS in Warsaw were derived from whole blood. All PLTs in Graz and 60% of PLTs in Warsaw were suspended in platelet additive solution (SSP+); the remainder of PLTs in Warsaw were stored in 100% plasma. All PLTs were approved for 7-day storage. Both sites transfused >75% of PLTs on days 2-5 post-collection; ~3%-4% of PLTs were transfused on day 7. Approximately 1.4% of PLTs in Warsaw were cryopreserved and transfused after day 7. Per patient PLT utilization was similar in both sites (mean: 8, range: 1-58). Five PLT TRs were reported (2 febrile non-hemolytic TRs [FNHTR] in Graz; 1 FNHTR and 2 allergic TRs in Warsaw). The per PLT TR rate was 0.21% for both sites. Two PLS TRs (allergic and unclassified) were reported in Warsaw (0.17% per PLS rate). All TRs were non-serious and moderate in severity; all patients recovered without complications. Rates were comparable to historical rates for PR and non-PR components at the participating hospitals and lower than rates (~1%) observed in prior HV studies.

Summary / Conclusions: Cross-sectional HV studies allow hospitals and medical device manufacturers to reaffirm safety profiles with RWE. Cerus aims to complete the study in 2024 with an additional ~2000 PLT and/or PLS units (total >5000) in the final dataset.

P799 | Blood transfusion safety in patients treated with anti-CD38 antibodies - efficacy of a pretransfusion protocol based on a neutralizing reagent, in a tertiary hospital

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Background: Anti-CD38 monoclonal antibodies have become very important in the treatment of plasma cell dyscrasias. However, they can produce an interference in pretransfusion testing causing panagglutination in the indirect antiglobulin test (IAT). We can solve this problem with a variety of methods like dithiothreitol (DTT) on reagent red blood cells (RBCs), which is considered the gold standard despite being time-consuming and a cumbersome technique. DaraEx is an anti-CD38 neutralizing agent that overcomes this interference in a simple, fast and efficacious method. Our group has already validated a DaraEx-IAT protocol with 100% concordance with the DTT-IAT technique and a shorter turnaround time.

Aims: To study the usefulness of our pretransfusion protocol to guarantee safe blood transfusion of patients treated with anti-CD38 monoclonal antibodies in our hospital using DaraEx as the first choice reagent to solve the expected interference.

Methods: We conducted a descriptive, retrospective 6-year study (2018-2023) analyzing our pretransfusion study protocol in patients treated with anti-CD38 monoclonal antibodies which consists in:

1. IAT and extended erythrocyte antigen typing prior to treatment
2. Inserting a warning in the transfusion file of each patient regarding expected interference
3. If transfusion was required, solved IAT

P799 - Table I

Patients with Dartumumab (n = 93)	
2018-2020	11 (11.83%)
2021	11 (11.83%)
2022	25 (27.08%)
2023	46 (47.92%)
Sex (n = 93)	
Men	41 (44.09%)
Women	52 (55.91%)
Age (mean, years)	72.28

P799 - Table 2

Transfusion requirements (n = 93)	
Yes	13 (13.98%)
No	80 (86.03%)
IAT result while on treatment with anti-CD38 Ab (n = 36 IAT)	
Positive	36 (100%)
Negative	0 (0%)
IAT interference resolution (n = 36 IAT)	
With DaraEx (in 12 patients)	26 (72.23%)
With DTT (in 1 patient)	10 (27.78%)
Incidents during/after transfusion (n = 13 patients)	
Yes	0 (0%)
No	13 (100%)
Alloimmunization after transfusion (n = 13 patients)	
Yes	0 (0%)
No	13 (100%)

interference with DaraEx-IAT 4. If interference was not resolved, DTT-IAT technique was used.

Results: In the studied period, 93 patients treated with anti-CD38 were analyzed (Table 1). All of them had a negative IAT prior to the initiation of anti-CD38 and 93.55% underwent extended erythrocyte antigen typing. Of the 93 patients, 13.98% (n = 13) required transfusion (mean: 2.5 RBCs concentrates; range: 1-10 units over the studied period) and a total of 36 pretransfusion tests that required interference resolution were performed (72.23% DaraEx-IAT; 27.78% DTT-IAT). Only one patient, a Poem's syndrome with renal failure who required 10 pretransfusion studies, required DTT-IAT to clear up the interference with an approximate 1-h delay in RBC units release compared to those patients with negative DaraEx-IAT. There were no transfusion related adverse events and after a minimum 4-month follow-up no RBC alloimmunization has been detected. (Table 2).

Summary / Conclusions: In our centre, few patients treated with anti-CD38 antibodies require transfusion. The transfusion of Rh-Kell compatible RBC concentrates with a negative DaraEx-IAT (or DTT-IAT) in these patients appears to be safe and cost-effective (no alloimmunization, no transfusion-related adverse events). DaraEx-IAT quickly and easily resolves the interference in most patients. We only find an exception in the resolution of the interference with DaraEx-IAT which reinforces the need to maintain DTT-IAT at the laboratory. The reasons for this exception remain unknown and require further study.

P800 | Universal pre-storage leukoreduction in RBC transfusions leads to significant reduction in febrile reactions identified through body temperature monitoring of hospital information system

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Background: Febrile reaction is the most common adverse reaction to blood transfusions. It can be mistaken for severe reactions, leading to misdiagnosis and requiring extra medications and resources. Part of febrile transfusion reactions can be prevented when transfusing with prestorage leukoreduction RBCs.

Aims: To evaluate the effectiveness of universal prestorage leukoreduction (UPL) in RBC transfusions in reducing febrile reactions, as implemented since July 2017 in a tertiary medical center.

Methods: Patients received blood transfusions in a tertiary medical center in Taiwan during 2013-2020 were included in this analysis. Only 3.7% of RBC transfusions used prestorage leukoreduction RBCs before July 2017. Characteristics of patients, transfusion details, and vital sign assessment data during blood transfusion were retrieved from the hospital information system and linked. Febrile reactions were identified if there was an increase in body temperature reaching 38°C, with a rise equal to or greater than 1°C within 6 h after the start of transfusion. The incidence rate and its 95% of confidence interval of febrile reaction was estimated. The t-test for proportion was used to compare the incidence rate before and after the implementation of UPL, and the chi-square test was used to evaluate differences in incidence among subgroups.

Results: A total of 62,401 transfusions performed with vital sign assessment during 2013-2020 were included in this analysis, and 910 of them identified febrile reaction from their vital sign assessment data. The overall incidence rate of febrile reaction was significantly decreased by 60.9% after implementing UPL, from 2.30% (95% CI: 2.12-2.49) to 0.90% (95% CI: 0.81-1.00) (p-value < 0.0001). Before UPL implementation, female patients had a significantly higher incidence rate (2.70%, 95% CI: 2.42-3.02) than male patients (1.96%, 95% CI: 1.74-2.22) (p = 0.0006), but the difference disappeared afterward (Female: 0.91%, 95% CI: 0.77-1.07; Male: 0.90%, 95% CI: 0.78-1.04; p = 0.99). The incidence rate exhibited an inverted U-shaped pattern and varied significantly across age groups. It peaked in patients aged 20-39 years old both before (4.64%) and after implementing UPL (1.43%), and was at its lowest in patients aged 60 years or older both before (1.82%) and after implementing UPL (0.78%). A significant 57-69% decrease was observed across all age groups. The incidence rate of RBC transfusions, performed with or without other blood components, significantly decreased from 2.66% (95% CI: 2.44-2.89) to 0.95% (95% CI: 0.85-1.07) (p < 0.0001) after implementing UPL. If patients were transfused with RBC only, platelets only, and fresh frozen plasma (FFP) only, the incidence rates were 2.34%, 0.97%, and 1.08%, respectively, before implementing UPL; afterward, the rates reduced to 0.77%, 0.64%, and

0.73%, respectively. No significant reduction was observed when patients transfused with platelets only and FFP only. The overall incidence rate of febrile reactions identified through the hospital information system was significantly higher (1.46%) than the rate reported through the hemovigilance system (0.05%) ($p < 0.0001$).

Summary / Conclusions: The incidence of febrile reactions reduced significantly by 60.9% after implementing UPL in RBC transfusions. Slightly reductions were also observed in platelets and FFP transfusions. Febrile reactions can be efficiently identified through the hospital information system.

P801 | Effectiveness of leucodepletion filters in reducing adverse transfusion reactions in multi-transfused beta thalassaemia major patients

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Background: Leukoreduction involves the removal of white blood cells from cellular components to reduce the risk of adverse transfusion reactions most notably febrile nonhaemolytic transfusion reactions (FNHTRs). Leukoreduction also has the ability to stop leukocyte metabolites like histamine and cytokines from being released. These metabolites enhance the rate of red cell haemolysis and are responsible for adverse reactions. Currently, leukoreduction is achieved by using Leukocyte Reduction Filters (LRFs) that trap leukocytes while permitting the desired blood product to pass through. The processes of barrier retention and cell adsorption onto the filter membrane enable leukoreduction by filtration. LRFs may be used at the bedside, in a hospital, or in a blood centre at the time of collection.

Aims: Currently, no LRFs are used at the study site due to financial constraints. Hence, the current study was designed to assess the efficacy of LRFs in reducing adverse transfusion reactions in multi-transfused beta thalassaemia major patients.

Methods: This prospective study was performed at the Thalassaemia Centre of the Divisional Headquarters Teaching Hospital, Mirpur, AJK, Pakistan. Over a 2-month period, 70 units of red cell concentrates were transfused to 70 beta-thalassaemia major patients (aged 11–17 years) with a history of FNHTRs. No pre-medications (Solu-Cortef or Avil) were administered to observe the effectiveness of the LRFs. In 35 patients (Group-1), PuriBlood bed-side leukocyte filters (PuriBlood Medical Co., Ltd., Taiwan) supplied by Alpha Evolution Enterprises Pvt. Ltd., Pakistan, were used. In Group-2 patients, non-leukoreduced blood was routinely transfused. PuriBlood LRFs aim to eliminate > 99.99% (4-log reduction) of leukocytes with less than 8% red cell loss. Written consent was

obtained from patients (or their parents) before the start of transfusions and the use of LRFs. The data were analysed using SPSS version 25.0.

Results: The age of transfused red cells ranged from 4 to 19 days. Adverse transfusion reactions occurred in 52.85% ($n = 37$) of transfusions. Transfusion reactions were significantly reduced ($p < 0.05$) in Group-1 patients receiving PuriBlood-filtered red cells (8.57%; 3/35), which had earlier reported a high incidence of FNHTRs. In Group-2 patients, the transfusion reactions were much higher (97.14%; 34/35). The median onset and duration of the reaction were two hours (range 20 min–17 h) and four hours (range 1/2–23 h), respectively. About 82.85% ($n = 29$) of the reactions occurred during transfusion. None of the of the transfusions were discontinued.

Summary / Conclusions: The results showed that a significant reduction occurs in FNHTRs when bedside LRFs are used as compared to non-leukoreduced blood. Hence, leukoreduction of blood components in thalassaemia patients can be useful in preventing transfusion reactions. The leucofilters use needs to be encouraged and regulated by the government. This is an ongoing study, and LRFs will be routinely used on thalassaemia patients to assess the efficacy of leucodepletion and correlate it with the economic benefits of employing leukoreduced blood components, such as a decrease in FNHTRs and a prolongation of transfusion interval.

P802 | Analysis of transfusion reactions reported to the Korean Haemovigilance system

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Background: Korean national haemovigilance system was launched in 2007 and has been operated by the Korean Society of Blood Transfusion supported by the Korean Ministry of Health and Welfare since August 2010. We developed the on-line reporting system in 2011.

Aims: We intended to analyze the transfusion reactions reported to the Korean Haemovigilance System from 2011 to 2023.

Methods: The classification of transfusion reactions was revised in 2017, and the following reactions are currently being reported: acute hemolytic transfusion reaction (AHTR), delayed transfusion reaction (DHTR), delayed serologic transfusion reaction (DSTR), febrile non-hemolytic transfusion reaction (FNHTR), allergic reaction/anaphylactic reaction, hypotensive transfusion reaction, transfusion-related acute lung injury (TRALI), transfusion-associated circulatory overload (TACO), transfusion-associated dyspnea (TAD), post-transfusion purpura (PTP), transfusion-associated graft vs. host disease (TA-GVHD), and transfusion-transmitted infection (TTI). The number, severity, and related blood products by types of transfusion reactions and by year are analyzed.

Results: Total 33,742 transfusion reactions were reported during this period. Of these, 33,713 (99.9% of the total) occurred regardless of incidents, and there were 29 cases (0.01%) in which incidents actually

led to transfusion reactions (complete incidents). FNHTR (19,563, 57.9%) and allergic reactions (9629, 28.5%) accounted for the majority (86.4%) of all transfusion reactions. Six hundred and nineteen cases (1.8%) of TAD, 342 cases (1.0%) of hypotensive reaction, 111 cases (0.3%) of TACO, 49 cases (0.1%) TRALI, 45 cases (0.1%) of AHTR, 20 cases (0.06%) of DHTR, 20 cases (0.06%) of DSTR, 4 cases (0.01%) of TTI and 3 cases (0.01%) of PTP were reported. Three thousand three hundred and sixty-seven cases (10.0%) were classified miscellaneous. A total of 29,143 cases were given information on the severity of transfusion reactions, with 26,124 mild cases (89.6%), 2,785 moderate cases (9.6%), 227 severe cases (0.8%), and 7 death cases (0.02%). TRALI (87.5%), TACO (73.1%), AHTR (64.4%), Hypotensive reactions (49.6%), and DHTR (40.0%) had a high proportion of moderate to severe severity, while DSTR (94.7%), FNHTR (93.4%), miscellaneous (90.9%), and allergic reaction (86.4%) showed mostly mild severity. The number of cases by blood product is in the order of red blood cells (RBC, 69.4%), single donor platelets (SDP, 10.7%), platelet concentrates (PC, 10.2%), fresh frozen plasma (FFP, 8.9%), cryoprecipitates (CRYO, 0.1%) and others (0.6%). The number of transfusion reactions compared to the amount of blood products transfused is one case per 641 units of SDP, one case per 650 units of RBC, one case per 2,030 units of FFP, one case per 4332 units of PC, one case per 17,453 units of CRYO, and one case per 1169 units overall.

Summary / Conclusions: Mild FNHTR and allergic reactions account for the majority of reports to the Korean Haemovigilance System. However, reports of more severe TRALI, TACO, AHTR, and hypotensive reactions are also gradually increasing. Continuous reporting and monitoring of transfusion reactions were possible through the continuous operation of the national blood surveillance system. It is expected that it will help establish transfusion-related policies, improve work levels, and develop various guidelines in the future.

P803 | Abstract withdrawn

P804 | Near misses—an improvement tool for transfusion safety

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Background: A near miss event (NM) refers to any error along the transfusion chain, that in the case of not have been detected could represent an additional risk, in some cases with a fatal outcome, for the patient. Its early detection is essential in order to establish preventive and corrective measures. In Spain, NM have to be declared to the *Sistema Nacional para la Seguridad Transfusional (SISNST)*, through the regional blood transfusion centre, using a standard form. In order to improve the quality of the registered information and its subsequent analysis, the form was modified (MF) and distributed to blood centres and hospitals in 2023 for its implementation, by the SISNST. The MF includes new items (mistakes attributable to transportation and Transfusión Centre), more specific and expanded description of pre-existing ones, qualitative information about analysis of the root causes, communication to the hospital transfusion committee and adoption or not of preventive/corrective actions.

Aims: To determine the impact of MF obtaining relevant information to minimize risks.

Methods: A descriptive study was carried out analysing the data of the reported NM by hospitals in our autonomous community (Madrid) between May and December 2023, after the implementation of MF.

P804 - Table.

Question	Answer	%
Staff area that detected NM	Transfusion service staff	64%
Place of NM detection	Laboratory	59%
Errors filling out MF or during sample extraction	This is another type of error	33%
Prescription errors	The special characteristics of the component are not indicated	35%
Transfusion service errors	Reception of the request and/or samples was not carried out correctly	33%
Handling and storage errors	The red blood cell concentrate has remained outside the refrigerator longer than appropriate	49%
Errors in the patient's bedside at the time of transfusion	The agreement between the patient's data and the bag label was not verified	44%
Transfusion center distribution errors and transportation service errors	Other type of error	67%
Staff responsible of the error	Hospital nurse	61%
Was the staff who made the mistake the usual staff?	Yes	89%
Time when the error occurred	Daytime	84%
Communication of the NM to the transfusion committee	No	65%
Has the cause(s) of the NM been analysed?	YES	62%
Corrective measures were adopted once the NM was analyzed	Yes	71%

Results: During the period of study 895 NM were reported, versus 312 NM in the same period of 2022. TABLE II summarizes the answers to the modified questionnaire questions that obtained the highest response percentages.

Summary / Conclusions: According to the obtained results MF has been used more frequently than the previous form (+186%). It has allowed to detect the critical points that could represent an increase of the risk, in our transfusion process and also evidence that the majority of NMs have been detected by the transfusion service staff, in the laboratory and have been committed by usual hospital nurses during daytime hours. It has allowed as to reinforce initial and continuing training programs for staff and the role of transfusion committees and transfusion safety systems.

P805 | Safety blood transfusion—analysis of the impact of the Haemovigilance Nurse in the hospital network of the Community of Madrid (Spain)

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Background: Haemovigilance systems are tasked with detecting, recording, and analyzing information pertaining to all adverse effects of transfusions. As laid down in the European Union Blood legislation, mandate the reporting of adverse reactions and serious adverse events related to the quality and safety of blood and its components, while the clinical use falling under the responsibility of Member States. In Spain, the Ministry of Health publishes an annual Haemovigilance report, which includes the adverse reactions, administration errors and "near misses" blood transfusion-related. According to published literature, the Haemovigilance Nurse (HN) or "Transfusion Officer" is a crucial figure in enhancing transfusion safety, and demands the highest level of specialized education, training, interpersonal skills, and management abilities to serve as a referent on blood transfusion. This role has been developed in Madrid since 2015 and this study demonstrates its impact on the visibility of transfusion risks.

Aims: Describing the functions of the Haemovigilance Nurse (HN) in hospitals where the figure has been implemented. Analyzing their impact on the reporting of adverse effects detected throughout the transfusion process.

Methods: Retrospective, descriptive, comparative, longitudinal, and multicenter study. Functions of HN: Educate Clinical Staff on best transfusion practice. Audit the transfusion practice throughout the hospital. Review and establish guidelines and procedures. Investigate and report serious transfusion reactions and events to the Spanish

P805 - Table 1: Haemovigilance in the Community of Madrid.

Year	Near-Miss Incidents (N)	Adverse reactions (N)	Administration Errors (N)	Notification Rate (per 10,000)
2012	22	222	16	9.27
2013	37	210	13	9.25
2014	26	164	14	7.18
2015	75	208	23	9.44
2016	70	184	13	8.62
2017	112	322	16	14.56
2018	119	379	23	17.16
2019	216	401	26	21.14
2020	47	302	28	13.08
2021	326	382	32	25.27
2022	470	444	53	33.97

The increase in the reporting rate was statistically significant (p -value <0.0025) (95% CI), as was the detection of Near-miss incidents (p -value <0.001) (95% CI) and administration errors (p value <0.004). No significant relationship was found in the evolution of adverse reactions.

Data from hospitals that incorporated HN in 2023, reveals a very significant increase in the reporting rate of "near misses"; from 0 to 39.17 per10,000 (Hospital1, transfusion>5000); from 2.31 to 55.31 (Hospital 2, transfusion>25,000) and 97 to 100.65 (Hospital 3, transfusion >25,000).

National Haemovigilance System. Data were collected from Haemovigilance reports published from 2012 to 2022. Comparative analysis was conducted using the independent samples t-test, applied to the arithmetic means before and after 2015, when the Haemovigilance Nurse's activities began in Madrid Hospital Network. In addition, "near-miss" data was collected from hospitals that implemented the figure in 2023. The average number of near misses reported up to 2022 was compared with the rate in 2023.

Results:

Summary / Conclusions: The results indicate that the HN's activity has an impact on increasing the incident and near-miss incident notification rate, revealing the true extent of risks associated with transfusion, that could be under-reported. Detecting and analyzing near-miss incidents allow for addressing preventable risks in transfusion and focusing preventive measures effectively. The HN contributes to an improvement in transfusion safety.

P806 | Advancing patient care—comprehensive review of home blood transfusions at Burgos University Hospital

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Background: Home-based blood transfusion has been employed worldwide in recent decades and is currently experiencing significant growth,

expected to continue in the coming years. This approach aims to decentralize hospital care with the goal of improving patient autonomy and quality of life. This practice has become increasingly relevant due to various factors such as increased life expectancy, the prevalence of chronic diseases necessitating periodic transfusions, and the exploration of alternatives to minimize the risk of nosocomial infections.

Aims: To determine the prevalence of home blood component transfusion, the units transfused, and the characteristics of patients in the Home Hospitalization Service of the University Hospital of Burgos.

Methods: A descriptive cross-sectional study was conducted on patients transfused in the Home Hospitalization Unit of the University Hospital of Burgos from 1/1/21, to 12/31/23. The total number of patients admitted to HAD during the periods 2021-2022 and 2023 was 922, 1005, and 1062 patients, respectively. **Inclusion criteria:** Patients with any pathology requiring transfusion of red blood cell concentrates, platelets, or plasma during their stay in HAD and who were under scheduled follow-up or interconsultation from another service.

Results: During the period from 1/1/21, to 12/31/23, there were 488 transfusion events in a total of 157 patients, with a median age of 89 years (range: 26-100), of which 55.41% were women. The prevalence of transfusion events in patients admitted to HAD during the period 2021-2022 and 2023 was 16.32%. The most transfused blood component was red blood cell concentrate, accounting for 94.47%, followed by platelet concentrates at 3.07%, irradiated red blood cell concentrates at 2.87%, and fresh frozen plasma at 0.41%. The overall mean number of transfusion events was 3 units per patient. During the periods of 2021 and 2022, the mean remained at 3 units per patient, while in 2023, the mean decreased to 2.8 units per patient. The type of patient who received transfusions most frequently were patients with iron-deficiency anemia (33%) and multifactorial chronic anemia (26%), with the highest proportion in the year 2021 at 38%, followed by onco-hematological patients at 22%, and oncological patients at 19%. During the period of 2021, the most frequent sources of origin were Primary Care and geriatric residences. According to our records since 2009, there has been a steadily increasing demand for home transfusions, attributed in part to demographic changes in the patient population, improvements in survival rates, aging, increased conditions requiring chronic care, as well as reducing the risk of nosocomial infections, especially during the COVID-19 pandemic. During 2021, only 2 mild transfusion reactions were recorded. In contrast, no adverse reactions were identified in the year 2022, while in 2023, 2 mild transfusion reactions were observed again. The most frequent adverse event was non-hemolytic febrile transfusion reaction. All mild reactions were efficiently managed at the individual's home according to existing protocols for the management of transfusion-related complications.

Summary / Conclusions: The prevalence of transfusion events in patients admitted to HAD during the periods 2021-2022 and 2023 was 16.32%. The majority of patients were polymorbid and elderly with onco-hematological conditions. Transfusions have been a safe procedure in the patient's home, with few adverse effects.

P807 | Audit of blood transfusion incidents in the province of Girona, Catalonia, Spain

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Background: The objective of haemovigilance is the continuous quality and safety improvement by the reporting of incidents. In the blood transfusion area we could define: (1). Near misses: error detected and solved before the transfusion. (2). Blood transfusion incidents (BTI): error detected after transfusion. BTI are potentially harmful to patients or even fatal, and could cause overtransfusion in Transfusion Services.

Aims: The aim of this audit is to describe the transfusion incidents in our area, and analyse the data to conduct corrective and preventive action (CAPA) plans.

Methods: We conducted an audit of all the incidents notified in the province of Girona, seven reporting centres from 2018 to 2023. BTI were divided in four main categories: Inappropriate transfusion (Inadequate indication or dosage, delayed or omitted). Manipulation errors (transfusion >4h, transfusion <1h, transfusion mixed with other medication). Component requirements errors (Irradiation not respected, phenotype not respected). Blood component errors (ABO mismatch, component destined to other patient or wrong component solicited).

P807 - Table 1. Blood components transfused in 2023 and Notifications reported from 2018 to 2023

Center	Blood Components transfused 2023	Notifications reported (%)
Girona	8111	71 (68%)
Institut Català d'Oncologia	4176	19 (18%)
Maresme-Selva	3260	4 (4%)
Figueres	3157	3 (3%)
Santa Caterina	2417	4 (4%)
Olot	1015	2 (2%)
Campdevàdol	512	1 (1%)

P807 - Table 2. Distribution of BTI by component type.

Blood Transfusion Errors	Packed Red Blood Cells	Platelets	Plasma
Inadequate indication or dosage	23 (29%)	20 (91%)	3 (100%)
Component Requirement	18 (23%)	1 (4.5%)	-
Manipulation	11 (14%)	-	-
Delayed transfusion	11 (14%)	-	-
Blood Component Error	10 (12%)	1 (4.5%)	-
Omitted	6 (8%)	-	-
Total	79 (100%)	22 (100%)	3 (100%)

Results: From 2018 to 2023, 104 BTI were reported. Notifications by centre and transfusion burden is showed in Table I. Notifications by year 2018 11 (11%) 2019 8 (8%) 2020 6 (6%), 2021 12 (11%) 2022 27 (26%) 2023 40 (38%).

The most frequent BTI was inappropriate transfusion 63 (60%), of which 46 were inadequate indication or dosage, 11 delayed transfusion and 6 omitted transfusions. There were 19 (18%) component requirement errors reported, not respecting the irradiation 9 cases, followed by not respecting the red blood cells phenotype in 5 cases, and 5 cases of transfusion of not fractioned component. Blood component errors reported 11 (11%) included wrong ABO component selection but fortuitously with ABO compatibility 9 cases, one case of ABO mismatch transfusion, and one case of mistake at the blood transfusion solicitude. Manipulation errors reported 11 (11%) 10 cases of slow transfusion, and one case of transfusion of red blood cells with iodine contrast. All the plasma BTI were related to wrong dosage, that led to under therapeutic transfusion. The most frequent BTI related with platelet was inadequate indication, that resulted in overtransfusion. Table 2 resumes the BTI by component type.

Summary / Conclusions: BTI are underreported. Trained personal is key to detect and notify them, in order to develop CAPA plans. The increase of notifications over the years could be related to the performed platelet transfusion audit during 2022. The most common error was the inadequate indication of blood transfusion, in this way educational interventions and periodical training should be carried out to improve the transfusion knowledge of practitioners. Formation in the blood transfusion indication should be provided to practitioners to ensure a correct transfusion.

P808 | Mis-transfusion captured by real time hemovigilance

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Background: Transfusion-related errors (TRE) are unfortunate events that occur due to system failures and are experienced by 3.7% of hospitalized patients. TREs are considered critical medical errors due to the potential for significant morbidity and mortality. Safeguards, often implemented after failure mode effect analysis (FMEA), are practices known to protect against harm. Real-time hemovigilance (RTHV) surveillance programs capture data surrounding transfusion-related adverse events (TRAE) ensuring performance of FMEAs and implementation of deference barriers.

Aims: We aim to highlight the ability of our RTHV system in detecting TRE and demonstrate the added value of the system to

P808 - Table 1.

15:43	1
17:46	2
18:30	1
21:30	1
00:30	0

ensure retrospective corrective interventions occur after system failures.

Methods: Our institution is a 678-bed hospital and ambulatory center that dispenses approximately 100,000 blood products annually. TRAEs are detected using our RTHV system. This system consists of a transfusion digital dashboard with an integrated TRAE score, specialty-trained transfusion nurses, and transfusion medicine advanced practice providers (TMAPP). Our RTHV system enables healthcare providers to intervene promptly and mitigate potential harm to patients. Patient-centered TRAE scores are computed and continuously updated during and up to 12 h post-transfusion. Aberrant scores trigger a patient evaluation and, if a TRAE is suspected, a diagnostic workup is performed. For medically urgent TRAEs, a transfusion medicine physician (TMP) is consulted for real-time management.

Results: Our RTHV system detected a hypotensive event related to the transfusion of one unit of red blood cells. The patient's TRAE score increased from one to two, prompting the deployment of a TMAPP. Physical exam was remarkable for only hypotension, no fever was observed. Pre-medication with Tylenol was documented in the electronic health medical record. The post-transfusion workup DAT was positive for C3 initially and IgG subsequently. Elution studies eluted an anti-Jka antibody. Retrospective review of the pre-transfusion testing demonstrated the presence of an anti-Jka antibody. The dispensed unit typed positive for the Jka antigen. Final review of the case by a TMP resulted in the diagnosis of a hemolytic transfusion reaction

Summary / Conclusions: Informatics systems eliminate the underreporting of TRAEs and function as a defense barrier. Data captured by RTHV systems prompted policy revisions to ensure potentially avoidable TRE do not recur.

P809 | Improving near misses reporting in transfusion services—the role of database enhancement and professional training

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Background: Haemovigilance plays a pivotal role in ensuring the quality, traceability and safety of blood components and their transfusion.

P809 - Table 1. Characteristics of transfusion incidents and transfusion rates. CMT: cross-matching tests. NM: near misses.

Year	2020	2021	2022	2023
Global transfusion incidents	27	25	14	168
Adverse events	19	16	6	7
Administration errors	2	2	0	1
Near misses	6	7	5	160
Incidents not related to sample extraction for CMT	0	2	1	4
Incidents related to sample extraction for CMT	6	5	4	156
Inaccurate demographic data	3	1	3	69
Sample labelling issues	0	0	0	83
Demographic data and labelling issues	0	1	0	3
Wrong blood right patient labelling	3	3	1	1
Transfusion requests	29,864	31,184	31,655	33,866
CMT NM reporting rate	2.01	1.60	1.26	46.06
CMT NM reporting rate percentage change	-	-20.2	-21.2	3545.4
Transfusions	30,370	28,826	29,276	28,927
Transfusion error reporting rate	8.89	8.67	4.78	58.08
Transfusion error reporting rate percentage change	-	-2.4	-44.9	1114.5

In this context, enhancing the documentation of near misses (NM) is crucial, particularly given the challenges posed by personnel turnover, time constraints, lack of information, or inherent complexities, which may result in underreporting the true frequency of such events.

Aims: The objective of this work was to improve the registration of NM in a database by providing periodic training to staff on the registration process.

Methods: This descriptive longitudinal study, conducted between 2020 and 2023 at a tertiary hospital, comprising both retrospective and prospective phases. Initially, we collected data on incidents related to sample extraction for cross-matching tests (CMT), reported between 2020 and 2022. An improvement action was implemented from 2023 onward, aiming to enhance both the database and the training of the professionals. On one hand, rejection reasons were categorized in the database to facilitate registration: inaccurate demographic data, sample labelling issues, demographic data and labelling issues and wrong blood-right patient labelling. On the other hand, periodic training meetings were conducted with blood bank personnel. These sessions included training on the database, informing about the systematic inspection of tubes upon reception, raising awareness about the impact and usefulness of NM reporting, and any routine-related queries that arose.

Results: The year CMT near miss rate was 2.01, 1.6 and 1.26 between the years 2020 and 2022 respectively, with inaccurate demographic data being the main CMT near miss. After implementing the improvement action, rate was 46.06 during 2023, resulting in a 3545.4% increase compared to the previous year, with the main CMT near miss being the sample labelling issue. This also resulted in an increase in the overall incident notification rate,

raising the Transfusion error reporting rate by 1114.5% compared to the previous year (Table 1).

Summary / Conclusions: It can be asserted that the implemented improvement action has greatly enhanced the reporting of NM and transfusion errors. The next step is to analyze which transfusion requesting areas are the most recurrent in the collected NM to carry out another improvement action consisting of periodically training professionals from those areas, aiming to reduce the number of NM. It is evident that professional training is essential and enables the improvement of transfusion service quality.

P810 | Analysis of transfusion related serious adverse reactions; Polish experience 2015–2022

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Background: Transfusion of blood components may sporadically be associated with the risk of serious adverse reactions (SARs), both related and unrelated to the errors at various stages of the blood transfusion chain.

Aims: The study aim was to assess the number as well as the type of SARs in Poland in the 2015 - 2022 period.

Methods: Retrospective analysis of the data related to SAR cases forwarded to the Institute of Hematology and Transfusion Medicine (IHTM) by Polish Blood Transfusion Centers and reported to the

European Commission pursuant to directive 2002/98/EC and commission directive 2005/61/EC.

Results: In 2015 - 2022, there were reports of a total of 394 SARs with 1-3 imputability levels, including: 243 cases related to red blood cell concentrate (RBC), i.e. 2,61/100 000 units issued, 91 cases related to fresh frozen plasma (FFP), i.e. 3,91/100 000 units issued, 60 cases related to platelet concentrate (PC), i.e. 6,01/100 000 units issued. The highest number of SARs (76) was reported in 2018, the lowest (21) in 2016. For RBC transfusions, the most frequent SARs were transfusion-associated circulatory overload (TACO) - 97 cases (including 1 fatality); transfusion related acute lung injury (TRALI) - 47 cases. Cases of immunological hemolysis were less numerous - 26 but included 2 fatalities. The most frequent FFP-related SARs were TACO - 121 cases, anaphylaxis/hypersensitivity - 94, TRALI - 77; no fatalities were reported. The most frequent PC-related SARs were anaphylaxis/hypersensitivity - 25 cases, TACO - 13 cases, TRALI - 7; no fatalities were reported.

Summary / Conclusions: Serious adverse reactions per number of units issued were relatively most numerous for PCs, and the least numerous for RBCs. All reported fatalities were related to RBCs transfusions. The most frequent SARs were TACO, TRALI and anaphylaxis/hypersensitivity. The main reason for transfusion-related fatalities were human errors (2 acute hemolytic reactions due to transfusion of incompatible RBCs). This may indicate the need for additional training for personnel involved in blood transfusion.

P811 | The importance of deep interview by physician in donor selection to ensure low-risk donors—a case report of malaria investigation at Jakarta Blood Centre after detection in patient—2023

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Background: Indonesia is one of the countries with several regions that are malaria-endemic, including in Maluku, East Nusa Tenggara, Sulawesi, Papua, West Papua, and some parts of Kalimantan and Sumatra. Meanwhile, the DKI Jakarta Province has been declared malaria-free since 2019. Malaria can be transmitted through blood transfusion, but in Indonesia, regulations only require screening tests for four infectious diseases: Hepatitis B, Hepatitis C, HIV, Syphilis. In the donor selection stage, it is hoped that high-risk malaria donors can be identified during interviews by the examining physician. Previously, donors would fill out a form answering several questions about their personal data and health history. There is only one question on the form related to malaria: "Have you ever suffered from malaria before?" Of course, the honesty of the donor in answering this question is crucial as it can influence the physician's decision in determining donors at risk of malaria. Jakarta Blood Centre received reports of two malaria cases from the Jakarta Provincial Health Office. These patients were detected with malaria at the hospital. Both patients were known from their medical records to have undergone treatment with blood transfusions.

Aims: To investigate the cause of malaria cases in patients, who have been transfused with blood originated from the Jakarta Blood Centre.

Methods: Checking data in the Blood Transfusion Information System (BTIS) of the Jakarta Blood Centre, blood bags that have been transfused to Patient X and Patient Y. In the blood flow section BTIS will be able to obtain all information related to the blood. And then the Jakarta Blood Centre confirmation team traced the listed contact numbers, then conducted detailed interviews specifically for health histories that may lead to infection or exposure to malaria.

Results: The results of investigation are presented in the table below

Summary / Conclusions: This further reinforces the belief that the donor selection process during the physician-donor interview stage

P811 -Table 1.

Investigation result		
Information	Patient X	Patient Y
Onset of symptom	23 December 2021	28 October 2022
Malaria diagnosis	4 January 2022	9 November 2022
History of blood transfusion	YES	YES
Amount of blood transfusion	7 bags of PRC	5 bags of PRC
Donors reachable	5 people	4 people
Donors at high-risk for Malaria	Yes, 1 person	Yes, 1 person
History of living in or traveling to Malaria endemic area	Lived in Papua for work from 2014 to 2021	Traveled to Papua in February 2021
History of Malaria	Yes, hospitalized in November 2021 due to Malaria	Yes, hospitalized in April 2021 due to Malaria
History of blood donation	9 December 2021	7 October 2022

From the investigation results above, it was found that the blood transfused to Patient X and Patient Y, one of which came from a high-risk malaria donor as seen from their residency history, travel history, and previous treatment for malaria. This information should have been obtained during interview in donor selection stage, but unfortunately this was not done.

requires special attention and caution regarding the possibility of high-risk donors for diseases that can be transmitted through blood transfusion, especially malaria, due to the lack of malaria screening tests.

P812 | An assessment of radio frequency identification based system for tracking and traceability of red cell units in a quaternary care hospital based blood centre

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Background: Radio frequency identification (RFID) can be a key enabler for enhancing productivity and safety of the blood product supply chain. Radio frequency identification can help overcome a number of common challenges and process inefficiencies associated with identification and tracking of blood products. The rationale of the study was to assess the utility of RFID based inventory and traceability in our blood centre.

Aims: To assess the utility of RFID based system for inventory management and bedside monitoring.

Methods: The study was a part of patient safety imitative under quality improvement program. It was based on Plan-Do-Study-Act. The RFID (Biolog ID systems) based inventory as well as bedside monitoring was incorporated at blood centre and at bed side of the patient. A total of 1200 Red cell units were assessed. They were divided in to two groups: Group I 600 red cell units without RFID tags which were processed and issued manually (data used for observations) and Group II another 600 red cell units which processed and issued with RFID system. The parameter assessed were turnaround time, transfusion follow up form, Inventory verification, mismatch transfusion and hemolysis in red cells. The ethical permission of the study was taken from the institute ethical board.

Results: The time taken at the issue counter with RFID based blood bags was significantly lower vs manual issue of blood bags (4.3 ± 1.2 mins vs 6.2 ± 2.1 min.) The collection of transfused empty blood bag & transfusion follow-up, was 96% (without RFID) vs 100% (with RFID). At the bed side, time of start and end of transfusion was tracked in 99.7% of episodes with RFID tags vs 87% in red cells used RFID. The time taken for physical verifications was 64 mins vs auto generation after RFID. There was no hemolysis reported. No wrong identification of patient and blood mismatch was observed. Similarly no mismatch blood transfusion reported from ward/ot/icu. The average time for a blood unit out from storage was lesser in RFID bags (9 mins vs 11 mins).

Summary / Conclusions: RFID based system provide us with critical/ expiry notification to enable blood issue as per FIFO. There was limited exposure of the units to temperature resulting in minimal effect the quality of blood. Collection of Transfusion Collection Form along with empty blood bag can be attained 100% within stipulated time. RFID based system helped in optimization of transfusion process and

manpower with fast and contact less communication of real time monitoring and traceability

P813 | Haemovigilance—Macedonian experience and plans for future developments

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Background: Blood transfusion is safe and effective medical treatment if used appropriately, but it can be associated with a few adverse effects either immediate or delayed. Haemovigilance is a surveillance system and is dealing with the safety of the blood transfusion chain, from vein to vein. Every definition for haemovigilance includes recognition, reporting, analyzing and taking actions on undesirable consequences of blood donation and transfusion in order to improve quality of the whole transfusion chain.

Aims: Aim of this study is to show the experience of the Institute for Transfusion medicine of Republic of North Macedonia, the current situation and the plans for future developments of the haemovigilance in our country.

Methods: The Institute for Transfusion Medicine of Republic of North Macedonia (ITM) is working according the actual national legislative, the Law for safety in blood supply (Official Gazette, No 110 from September 2007), which is based on and is harmonized with the First European Blood Directive (“mother” Directive) 2002/98/EC for safe and quality collection, processing and testing of blood, storage and distribution of the blood components and its consequent by-laws harmonized with the rest three “daughter” European Blood Directives. The form and content of the documentation in the process of the use of blood, written forms and procedure for reporting of serious adverse events and reactions from used blood and blood components is prescribed in the Rulebook (Official Gazette, No 87 from July 2010) which is harmonized with the Directive 2005/61/EC. In accordance with this Rulebook, there are forms for reporting serious transfusion reactions that are partially filled out by a small number of hospitals.

Results: ITM has appointed doctors specialists in transfusion medicine, as external members in the hospital transfusion committees. The process of introducing the legally existing forms into the electronic healthcare system “mojtermin” has begun, which must be filled out by clinical doctors who order blood and blood component transfusions for their patients, with precise data on how many units and which blood components have been transfused and if there was serious adverse reactions and events.

Summary / Conclusions: Although many elements exist, haemovigilance as a national and functional system does not exist completely in R.N. Macedonia. There is no competent authority as part of the

Ministry of Health of R.N. Macedonia as a key element to verify the consistency and clarity of the collected information related to serious adverse reactions and events. There is continuous need to work on establishing and maintaining fully functional haemovigilance system, that should involve all relevant stakeholders and should be coordinated between the Institute for Transfusion Medicine, hospital staff, hospital transfusion committees and the national health authorities by establishing competent authority in R.N. Macedonia; and the following prerequisites are necessary: legal framework, national guide for haemovigilance, agreed definitions, use of standardized reports, guaranteed continuous funding of the haemovigilance program, functional hospital transfusion committees, introduced system of corrective and preventive actions, established culture of professionalism, anticipated international cooperation and establishing correct awareness and alert system to ensure that the transfusion policies, standards and guidelines will be followed.

P814 | Abstract withdrawn

P815 | Monitoring and analysing the effect of quality interventions to improve bedside transfusion practices

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Background: Blood transfusion process includes a complex series of events from ordering the blood, blood transportation to the destined location, correct patient identification, rightly administering the blood, monitoring and documenting the transfusion and adverse events, if any. Adequate documentation is important for each step of transfusion and is considered as an essential part of transfusion practice. However, the documentation of bedside transfusion practice is often overlooked and inadequate in developing countries.

Aims: To analyze the process of recording transfusion monitoring at bedside and the effect of implementation of safety interventions by reviewing the feedback forms at our centre.

Methods: This is a prospective, observational, single centre cohort study conducted in the department of Transfusion medicine in a tertiary care centre from Jan 2020 to Dec 2023 for 4 years. The study period was divided into two phases. Phase 1 2020-2021 and phase 2 2022-2023. For each blood transfusion episode, documentation of the transfusion event is done in the transfusion monitoring record which is received by the blood centre as transfusion feedback form. Parameters studied for the following in both phases for correct patient identification, number of forms received; completeness of the filling of the form; Turnaround times (TAT1): time from receiving issue slip in the blood centre to issue of the reserved blood component; TAT2: Time calculated from issue of blood component to start of blood transfusion (beyond 30 min -delayed transfusions); TAT3: Time from start to end of blood transfusion. (Maximum permissible time 4 h

from issue) along with vitals monitoring. Quality interventions include continuous call backs to the bedside for the feedback; better temperature controlled transport carriers with integrated software, using dedicated personnel; trainings for the nursing and duty medical officers. Data is captured in Microsoft excel and analyzed using spss software 20.

Results: During the study period, for a total of 35,571 transfusion episodes (1-4 components per episode); the mean transfusion feedbacks received showed statistically significant ($p < 0.001$) improvement from 83.02% in phase 1 to 97% in phase 2 along with other parameters like delayed transfusions and extended transfusions.

Summary / Conclusions: Quality and safety interventions helped us to reduce the turnaround times; better technology for traceability; timely transfusions, better staff compliance in bedside transfusion care, thereby better patient monitoring and care.

Clinical transfusion—alternatives to blood transfusion

P816 | Abstract withdrawn

P817 | Martial therapy—better management of the patient in the Emergency Department (ED)

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Background: Iron deficiency anemia is a common finding in emergency department patients and intravenous iron may be the therapy of choice in many cases. Incorrect management of these patients is often observed, and is the cause of a high and unjustified number of visits to the Emergency Department, despite the fact that this condition is preventable and curable, without resorting to transfusion therapy.

Aims: The purpose of this study is to evaluate the effectiveness of surgeries for the management of iron-deficiency patients through a shared protocol between the Transfusion Service and the Emergency Department to reduce hospital stays in the Emergency Department and evaluate the decrease in transfused patients, processed units, and improve the transfusion appropriateness.

Methods: The patients studied were suffering from iron deficiency anemia, referred to the Emergency Department because they were symptomatic with these values: Hb ≤ 6 , MCV ≤ 80 fl, TSAT $\leq 20\%$, Ferritin ≤ 30 ng/ml and RET %. The study was conducted in the two-year period 2022/2023. Iron therapy was performed with ferric carboxymaltose (Ferinject, FCM) 50 mg for 4 monthly infusions (1 infusion per week). The management of these patients was performed through

training meetings and weekly audits to ensure the correct use of blood and implement iron therapy.

Results:

P817 - Table 1.

Year	Total access emergency department	Transfused red blood units	Infused iron units
2022	236	183	581
2023	470	284	1323

Summary / Conclusions: This management has demonstrated a saving of approximately 290 units of red blood cells in 2022 and approximately 660 units for the year 2023 (1 patient on average was transfused with 2 units of red blood cells). Transfusion therapy was used only in critical cases, with Hb <6 g/dL and hemodynamically unstable. Taking charge of the chronic patient through the establishment of a martial therapy clinic has made it possible not only to avoid inappropriate transfusions, but to reduce access to the Emergency Department, making pathways more fluid for the acute patient. Ultimately, educational initiatives and collaboration between the transfusionist, the doctor in the emergency department and the doctors in the area can lead to safer and more targeted therapies and better management of the blood resource.

P818 | Abstract withdrawn

Clinical transfusion—Patient Blood Management (PBM)

P819 | Investigating blood product prescribing patterns among healthcare workers in Uganda

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Background: Effective blood product use is critical in Uganda's unique healthcare context. Despite medical advancements enabling tailored blood product use, anecdotal reports suggest a preference for whole blood and packed red blood cells (PRBCs) among healthcare workers, potentially due to knowledge gaps and systemic barriers. This study sought to investigate the current prescribing patterns, and healthcare workers' (HCWs) knowledge of blood product indications, identifying influencing factors, and perceived barriers to blood product prescription.

Aims: To determine the blood products frequently prescribed by healthcare workers in Uganda by patients' age and diagnosis. To assess the level of knowledge regarding the indications of blood products among

healthcare workers in Uganda. To determine the factors influencing knowledge regarding the indications of blood products among healthcare workers in Uganda. To determine factors healthcare workers in Uganda perceive as barriers to prescribing specific blood products

Methods: An online cross-sectional study was conducted among HCWs involved in blood product prescription in Uganda. Ethical approval was obtained from Mulago National Referral Hospital's Research and Ethics Committee. Data were collected through a self-administered pretested questionnaire in form of Google forms distributed on WhatsApp platforms and emails. Data analysis was performed in Microsoft Excel and STATA employing a logistic regression model to determine the factors associated with good knowledge ($p < 0.05$).

Results: Of the 306 participants, 213 (69.6%) were male with a mean age of 31 [SD = 6] years. Most participants were degree holders ($n = 237$, 77.5%) and completed their highest qualification in the past 3 years. Only 176 (57.5%) have ever attended a training on blood transfusion and 163 (53.3%) frequently prescribe blood to their patients. Most HCWs prescribe whole blood and PRBCs, and up to 124 (40.5%), 141 (46.1%), and 255 (83.3%) have never prescribed platelets, fresh frozen plasma (FFPs), and Cryoprecipitates respectively. Mean knowledge score was 5.3 (SD = 2.0). Diploma and certificate holders had lower knowledge (OR: 0.3, $p = 0.037$), while specialized officers showed higher knowledge (OR: 2.4, $p = 0.009$) compared to general practitioners. Frequent blood prescribers were more knowledgeable (OR: 2.1, $p = 0.039$). A negative correlation existed between knowledge scores and duration (in years) post-qualification and last training. Main barriers included lack of on-job training and facilities lacking specific blood products.

Summary / Conclusions: This study reveals gaps in HCWs' knowledge and prescribing patterns regarding blood products in Uganda. Suboptimal use of specific blood components suggests a need for targeted training interventions. Diploma and certificate holders, and those with limited prescribing experience, should be prioritized for educational initiatives. Addressing identified barriers, such as on-job training and ensuring consistent availability of specific blood products, is crucial for improving blood transfusion practices in Uganda. These findings contribute valuable insights to enhance blood transfusion services in Uganda.

P820 | Sanguine synergy—navigating the impact of patient blood management strategies on obstetric patients in a tertiary care setting

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Background: Patient Blood Management (PBM) is a proactive, evidence-based, multidisciplinary approach with improved patient safety and clinical outcome being the primary focus through the management of anaemia, optimization of haemostasis, iatrogenic blood

loss minimization and reduction of allogeneic blood transfusion while achieving waste reduction, cost control and quality improvement along the process. A PBM programme usually involves strategies to optimise blood utilisation preoperatively, during surgery and while the patient recovers. Since antenatal patients demonstrate diverse physiological changes, this study planned to assess the impact of management of nutritional anaemia on the principles of patient blood management and the outcomes associated with it among Obstetric patients.

Aims: To analyse the first pillar of Patient Blood Management, which is optimisation of the patient's own red cell mass, in correcting antenatal/ postnatal anaemia in Obstetric patients by the use of Haematinics and red cell transfusion and compare outcomes in patients managed either by red cell transfusion or Haematinics only or those with both Haematinics and red cell transfusion.

Methods: All antenatal in-patients with a period of gestation equivalent to 14 weeks and beyond presenting with nutritional anaemia were enrolled. The subjects were stratified into three distinct management strategy groups: Group 1 - Patients administered with PRBCs exclusively; Group 2 - Patients treated solely with haematinics; Group 3 - Patients subjected to a combination of both PRBCs and haematinics. Comprehensive pre- and post-intervention data, encompassing parameters such as hemoglobin levels, type of anemia, frequency of PRBC transfusions, complications, and various other metrics, were meticulously recorded. Statistical analyses, utilizing appropriate tools, were subsequently employed to make nuanced comparisons across each of the aforementioned management groups.

Results: The study spanned a duration of 15 months, commencing in May 2021 at the zenith of the second wave of the COVID-19 pandemic in India. A comprehensive cohort of 226 patients was enrolled. The severity of anaemia exhibited a direct correlation with the gravida status of the patient. Notably, a statistically significant weak negative correlation ($p = 0.014$) was discerned between age and serum ferritin levels. For patients managed exclusively with PRBC, the transfused units per patient averaged 2.58, whereas those who received both PRBC and haematinics received 2.08 units per patient. Noteworthy is the finding that complications were significantly reduced in the group receiving haematinics exclusively ($p = 0.0012$). Furthermore, a significant association ($p < 0.001$) was observed between sepsis and patients subjected to any form of transfusion. While the length of hospital stay did not exhibit significant differences across all three management groups, it was markedly prolonged among patients with anaemia compared to other antenatal patients admitted to our hospital.

Summary / Conclusions: In conclusion, strict adherence to the first pillar of patient blood management emerges as a pivotal determinant in enhancing the overall health of the patient. This commitment not only reduces the incidence of transfusions but also mitigates the occurrence of complications. To optimize the efficacy of patient blood management strategies, early targeting of this specific patient group is recommended, allowing for the proactive augmentation of their red cell mass.

P821 | Abstract withdrawn

P822 | Preoperative anaemia management reduces RBC use in cardiac surgery

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Background: Cardiopulmonary bypass surgery (CPB) dilutes haemoglobin (Hb) and haemostatic factors, thus increasing the risk of bleeding and perioperative red blood cell (RBC) transfusion. Preoperative Hb of 130 g/L⁻¹ minimises the likelihood of transfusion for CPB patients (Cavalli BJA 2023). Preoperative anaemia management (PAM) was implemented at The Prince Charles Hospital (TPCH) Brisbane, Australia in July 2014 using WHO anaemia thresholds as optimisation targets (Hb 130 g/L⁻¹ for males, 120 g/L⁻¹ for females).

Aims: To investigate if PAM reduced the average RBC units transfused per cardiac surgery patient.

Methods: Single site retrospective study of cardiac surgery patients at TPCH. Inclusion criteria: patients aged ≥ 18 years undergoing CPB between 1 June 2014 and 31 Dec 2018. PAM pathways were categorised in four ways to account for patient assessment and Hb optimisation. Patients were divided into three groups based on their initial Hb (g/L⁻¹) prior to any PAM: <120 , 120 to 129, and ≥ 130 . The Bellman Equation was used to calculate the average intraoperative, ICU, and ward RBC units transfused per patient.

Results: Data on 1017 female (Table 1) and 2668 male (Table 2) cardiac surgery episodes were analysed. Female median age (IQR) was 66 (55-74) and 66 (57-73) years for males. Irrespective of sex or PAM pathway, patients with an initial Hb < 120 g/L⁻¹ received more RBC

P822 - Table 1. Average RBC Units transfused per female cardiac surgery patient (n = number of patients).

Initial Hb (g/L ⁻¹)	PAM Status	Intra-op (n)	Post-op ICU (n)	Post-op Ward (n)
<120	AO	1.55 (51)	0.60 (52)	0.17 (46)
	ANO	1.63 (27)	0.96 (25)	0.16 (25)
	NANO	2.03 (188)	1.93 (187)	0.46 (181)
120 to 129	AO	0.15 (26)	0.12 (26)	0.04 (26)
	ANO	0.34 (68)	0.44 (71)	0.03 (65)
	NANO	0.75 (139)	0.60 (138)	0.12 (133)
≥ 130	AO	0.15 (62)	0.24 (62)	0.02 (62)
	ANO	0.32 (171)	0.49 (176)	0.08 (170)
	NANO	0.46 (240)	0.72 (244)	0.08 (232)

P822 - Table 2. Average RBC Units transfused per male cardiac surgery patient (n = number of patients)

Initial Hb (g/L ⁻¹)	PAM Status	Intra-op (n)	Post-op ICU (n)	Post-op Ward (n)
< 120	AO	1.71 (49)	2.34 (50)	0.30 (47)
	ANO	1.89 (28)	1.74 (27)	0.46 (26)
	NANO	2.06 (297)	2.59 (293)	0.34 (266)
120 to 129	AO	0.70 (50)	1.14 (50)	0.20 (49)
	ANO	0.58 (40)	1.10 (39)	0.23 (39)
	NANO	0.57 (198)	1.01 (201)	0.24 (198)
≥ 130	AO	0.08 (142)	0.35 (144)	0.09 (141)
	ANO	0.16 (572)	0.41 (582)	0.07 (569)
	NANO	0.21 (1208)	0.50 (1225)	0.11 (1199)

units per patient. Males received more RBC units per patient postoperatively than intraoperatively. Females 'assessed and optimised through PAM' (AO) had lower average RBC units per patient regardless of initial Hb levels. The average units transfused for 'not PAM assessed nor optimised' (NANO) females with an initial Hb 120 to 129 g/L⁻¹, was nearly double those females with initial Hb >130 g/L⁻¹. However, the average units transfused was the same for both these groups of 'AO' and 'PAM assessed and not optimised' (ANO) females.

Summary / Conclusions: PAM 'assessed and optimised' females had reduced average RBC units transfused per patient. However, the benefits of PAM pathways for males remain unclear. Further cost / benefit analysis is required to understand any economic advantage of increasing PAM optimisation targets to 130 g/L⁻¹ for all cardiac surgery patients.

P823 | Challenging iron deficiency diagnostic cut-off—exploration with big data approach

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Background: Treating suboptimal iron store and iron deficiency is one of the most important elements in optimising erythropoiesis in patient blood management. Serum ferritin is commonly used for diagnosis of iron deficiency. However, the optimal decision cutoff for diagnosing iron deficiency is still debated. The World Health Organization (WHO) defines iron deficiency using cutoff levels <15 ng/ml for adults and <12 ng/ml for children. The Royal College of Pathologists Australasia recommends a cut-off of <20 ng/ml (< 45 pmol/L) for diagnosing paediatric iron deficiency and <30 ng/ml (<68 pmol/L) for diagnosing adult iron deficiency.

Aims: We aimed to study the relationship between the ferritin level and the red cell parameters using a big data approach to evaluate the optimal ferritin level required for erythropoiesis.

P823 - Table 1.

CBC Parameters	Threshold ferritin value (ng/ml)			
	Young Children (age <5 years)	Older Children (age 5 to <13 years)	Adolescents (age 13 to <18 years)	Adults (age 18 to <65 years)
Hb	15	20	23	29
Hct	19	20	27	34
MCV	28	22	41	44
MCH	28	22	36	38
MCHC	16	24	25	25
RDW	26	27	38	36

Methods: The complete blood count (CBC) and the ferritin test results of patients aged <65 years in the New Territories West Cluster in Hong Kong between July 2019 and December 2023 were retrieved from the Laboratory Information System. The analyzers used were Beckman Coulter DxH 800 haematology analyzer for CBC and Abbott Alinity analyzer for ferritin test. Same patient results after the first episode were excluded. CBC and ferritin test results on the same date from the same patient were merged for analysis. Cases with ferritin ≤60 ng/ml were selected for analysis. The data were analyzed separately in 4 groups: young children (age <5 years), older children (age 5 to <13 years), adolescents (age 13 to <18 years) and adults (age 18 to <65 years). The relationship between red cell parameters and ferritin were analysed by an asymptotic regression model in R software. Outliers were excluded by residual analysis. Asymptotes were defined as the ferritin level at which further increase of ferritin by 1 ng/ml would not result in a change of red cell parameter value by 0.01.

Results: In the study period, there were 195,461 CBC and 87,189 ferritin results for the adult group and 35,918 CBC and 2787 ferritin results for the paediatric and adolescent group. After data merging, eventually 5036 and 537 merged results were available in the adult group and the paediatric and adolescent group respectively. In all groups, the red cell parameters showed a positive correlation (negative correlation for RDW) with ferritin until a threshold value of ferritin where the red cell parameters plateaued despite a further increase in ferritin value. In general, the threshold ferritin values for MCV, MCH and RDW are higher than those for Hb, Hct and MCHC. The threshold ferritin values for optimal levels of red cell parameters for each group is summarised in the Table below.

Summary / Conclusions: Our study suggests that iron-restricted erythropoiesis starts to occur before reaching the established recommended cutoffs of iron deficiency. MCV, MCH and RDW are more sensitive to iron deficiency than Hb, Hct and MCHC, and are early indicators of iron-restricted erythropoiesis. Identification of early iron deficiency by using erythropoiesis-based ferritin cutoff values could have potential implications for enhancing the management of iron deficiency and anaemia in the patient blood management perspective.

P824 | Increased transfusion of packed red blood cells, length of ECMO support and older age adversely affect the outcome of COVID-19 patients with acute respiratory distress syndrome

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Background: The COVID-19 pandemic was associated with dramatic growth in the number of patients with infection-related acute respiratory distress syndrome (ARDS). In ARDS patients, venovenous ECMO support was reported to decrease the death rate relative to the use of mechanical ventilation, while substantially increasing the need of blood products.

Aims: The present study aimed to evaluate a potential association of the amount of transfused blood products with intensive care unit (ICU) mortality among ECMO-supported COVID-19 patients, since these results could be applicable to a broader range of patient populations suffering from other infection-related ARDS.

Methods: Data were retrieved from patient electronic medical records and blood bank registries of three Israeli medical centers.

Results: The study included 102 patients [median age 49 (range 17-73) years]. The median duration of ECMO support was 20.5 (4-240) days and the median amount of transfused packed red blood cell (PRBC) units equated to 12.5 (0-197). Patients were categorized in the following three groups according to the total number of PRBC units received throughout the ECMO support period: <5, 5-15, and ≥16 units. The ICU mortality rates in these groups amounted to 14.3%, 25.7% and 60.9%, respectively, with an odds ratio (OR) between the last and first groups of 9.33 (95% CI 2.4-36.3). In bivariate analysis, associations between the following parameters and the elevated ICU mortality rate were demonstrated: ECMO support ≥38 versus <38 days (63.6% vs 27.5%; $p < 0.001$), age ≥50 versus <50 years (56% vs 23%; $p < 0.001$) and the number of transfused PRBC ≥16 versus <5 units (60.9% vs 14.3%; $p = 0.001$). The multivariate analysis identified age ≥50 years as a significant independent factor for increased ICU mortality (OR = 4.47; 95%CI 1.74-11.49). Transfusion of ≥16 PRBC units in combination with ECMO support for ≥38 days (OR = 14.18; 95% CI 3.11-64.77) were found to be associated with the highest mortality risk. Sepsis was the most frequent cause of death throughout the ECMO support period as well as during ICU hospitalization.

Summary / Conclusions: The results of the current study demonstrated that three quarters of ECMO-supported COVID-19 patients <50 years old and about half of older patients were successfully discharged from the ICU. Age ≥50 years was found to be a significant independent factor for increased ICU mortality. Likewise, in both age groups, a high number of transfused PRBC units in combination with long ECMO support were associated with elevated ICU mortality. These findings suggest that the utilization of approaches aiming to reduce the amount of drawn blood in ICU patients, while preserving their hemoglobin level, and thus diminishing the need for allogeneic blood transfusion could be advantageous.

P825 | A 5-year retrospective database review of anaemia prevalence and associated laboratory biomarkers in a central South African setting

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Background: Anaemia is a prevalent condition that affects populations in all income brackets. The World Health Organisation (WHO) identifies young children and pregnant women as the primary groups affected, with iron deficiency being a major contributor, accounting for nearly half of global anaemia cases. Various factors, including HIV, TB, cancer, micronutrient deficiencies and other acute and chronic illnesses can all reduce blood haemoglobin. Despite extensive research, the prevalence and associations between anaemia and certain biomarkers in adult medical patients have been less clearly defined.

Aims: This study aimed to investigate the prevalence and biochemical parameters associated with anaemia seen in medical in-patients, from archival laboratory data.

Methods: A retrospective descriptive study included patients 12 years and older, admitted to the Internal Medicine wards at Pelonomi Regional Academic Hospital, Bloemfontein, South Africa. Laboratory data were extracted from the National Health Laboratory Service (NHLS) corporate data warehouse database, after approval from the NHLS Academic Affairs and Research Management System. The analysis encompassed various parameters, including index haemoglobin, parameters of the full blood counts, nutritional markers, dysglycaemia indicators, estimated glomerular filtration rate, albumin levels, inflammation markers, and the status of HIV and TB, in patients with anaemia during a 5-year study period of 2018-2022. We used the WHO diagnostic criteria for anaemia and severity in males and non-pregnant females. Only patients with a haemoglobin done on admission were included in this study.

Results: Of the 1282 patients included in the study, 1007 were anaemic (78.5%). Females aged 15 to 65 years constituted the highest contributor (46.8%). The mean haemoglobin of males and females were 10.82 g/dL (SD ±3.4) and 9.71 g/dL (±2.8) respectively. Moderate

anaemia was most prevalent (45.2%), followed by severe and mild (33.9% and 20.9% respectively) with normocytic anaemia being the most common morphology noted between both sexes. The prevalence of anaemia in HIV-positive patients was 74.6% and The prevalence of TB was 13.7%. A minority of males (26.0%) and females (28.1%) were investigated for anaemia. Among these, females had a prevalence of iron deficiency anaemia of 4.3% and males a prevalence of 2.0%. The prevalence of anaemia of inflammation was similar between the sexes at 17.1%.

Summary / Conclusions: The findings from our study emphasise significant concerns regarding the prevalence and investigation of anaemia at Pelonomi Regional Academic Hospital, surpassing findings in similar patient profiles in higher socio-economic countries. Most patients were anaemic, with a significant contribution from females of childbearing age. The mean haemoglobin was lower in females, suggesting a higher burden of anaemia in this group. The high prevalence of anaemia in HIV-positive patients indicates a potential association between these conditions. Despite this, the study reveals a concerning trend of inadequate investigation, with only a minority assessed for iron deficiency, and highlights potential implications for transfusions in these patients. The study emphasises the need for improved investigative and management strategies for anaemia, particularly in a high-risk, adult, co-morbid population.

P826 | Optimizing preoperative hemoglobin levels—the role of blood management consultation

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Background: Preoperative anemia management is critical for optimizing surgical outcomes. Patient Blood Management (PBM) strategies have emerged as valuable tools to enhance hemoglobin levels before surgery.

Aims: This study aims to assess the impact of PBM consultation based on hemoglobin levels and variations in transfusion rates among different patient subgroups undergoing different surgeries.

Methods: A retrospective analysis was conducted on 300 patients referred to PBM consultation between 2021 and 2023, sorted into subgroups based on age, initial hemoglobin levels, and type of surgery. Data on hemoglobin levels, perioperative transfusion rates, and treatments received were collected and analyzed.

Results: PBM consultation resulted in a significant increase in hemoglobin levels across all patient subgroups. Patients aged 75 years or older demonstrated a moderate increase of 1.2 g/dL in hemoglobin levels post PBM consultation. Those with initial hemoglobin levels below 10 g/dL experienced a substantial increase of 3.5 g/dL post PBM consultation. Surgical subgroups showed variations in hemoglobin response to PBM, with non-oncological, non-orthopedic patients exhibiting the highest average increase of 3.6 g/dL. Transfusion rates slightly varied among subgroups, with the highest rates observed in patients undergoing other oncological surgeries (16.66%). *Detailed data are shown in Table 1*

Summary / Conclusions: This study highlights the effectiveness of PBM consultation improving preoperative hemoglobin levels, with varying responses among different patient subgroups. These findings emphasize the importance of tailored approaches to preoperative anemia management based on patient demographics and surgical characteristics.

P827 | Abstract withdrawn

P828 | Abstract withdrawn

P826 - Table 1.

Population	Age (y)	Initial Hb (g/dL)	Pre-Surgery Hb (g/dL)	Treatment
Overall Population (300)	68	10.4	12.2	ESA-51%, IV iron-95%, Transfusion rate-12.33%
Age>75 (106)	82	10.4	11.6	ESA-55%, IV iron-94%, Transfusion rate-12.26%
Initial Hb<10 (100)	66	8.7	12.3	ESA-70%, IV iron-97%, Transfusion rate-15%
Neo-Digestive Surgery (132)	73	10.5	11.9	ESA-70%, IV iron-97%, Transfusion rate-15%
Neo-Gynecologic Surgery (21)	60	10.8	11.8	ESA-48%, IV iron-95%, Transfusion rate-14.28%
Neo-Urinary Surgery (43)	70	10.6	11.9	ESA-56%, IV iron-98%, Transfusion rate-9.30%
Other Oncological Surgeries (24)	64	10.5	11.9	ESA-33%, IV iron-79%, Transfusion rate-16.66%
Orthopedic Surgery (14)	70	10.2	11.7	ESA-36%, IV iron-100%, Transfusion rate-7.14%
Other Surgeries (66)	63	9.8	13.5	ESA-53%, IV iron-80%, Transfusion rate-13.63%

ESA: Erythropoiesis-Stimulating Agents.

P829 | Are we going backwards—increase in overnight transfusion post pandemic

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Background: Non-essential overnight transfusion interrupts the recipient's sleep and also that of neighbouring patients. Transfusing overnight can also expose the patient to avoidable risk factors such as inadequate observation and monitoring, related to the reduced lighting and lower staffing levels. NZBS conducted an audit of overnight transfusion in 2011 and recommendations were implemented in strengthening blood policy to avoid overnight transfusion and maximise the opportunity to transfuse during the day in haemodynamically stable patients in non-high acuity units. Three years on since the Covid 19 pandemic was declared, Auckland Blood Bank has noticed a higher number of blood requests arriving in the evening. Feedback from ward nursing teams at Auckland City Hospital is that blood transfusion is being prescribed later in the day.

Aims: To see the percentage of the red cell units administered overnight between the hours of 8pm and 8am post pandemic, compared with pre pandemic levels and with NZBS' 2011 multi-site audit.

Methods: The transfusion nurse specialist undertook a retrospective audit of red cell issues at Auckland Blood Bank, New Zealand's busiest blood bank, during the audit months in 2019 and in 2023. The total number of red cell transfusions of the audit period was provided by NZBS' data analyst.

Results: Overnight transfusion post pandemic has indeed increased (13.9% vs 17.8%, $p = 0.021$). During March and April in 2019, total 1731 units were transfused with 206 units overnight. The number of units transfused overnight during the same months in 2023 was 294 out of 1636 units. However, both audit periods had more than doubled or nearly tripled compared to the NZBS' multisite audit in 2011 where the overnight transfusion in Auckland City Hospital was 6% ($p < 0.00001$ when compared with 2019 and 2023).

Summary / Conclusions: This audit has shown a significant increase in the amount of overnight transfusion compared to the 2011 audit. Although the recommendations in 2011 have been implemented, it does not appear hospital adhering to the policy, increasing safety risk in patients.

P830 | Assessment of single-unit red cell transfusion policy implementation in a tertiary care hospital in Catalonia—relevance of ongoing training

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Background: Patient Blood Management (PBM) programs aim for comprehensive, multidisciplinary, and multimodal management of

patients likely to receive blood transfusions. Numerous studies have demonstrated that implementing PBM policies leads to reduced unnecessary transfusions and improved clinical outcomes for patients. In the field of transfusion medicine, PBM recommendations advocate for prescribing a single unit of red blood cells (RBC) concentrate per transfusion for non-bleeding patients. It is imperative to reassess the need for further transfusion before prescribing additional units, moving away from the previous practice of prescribing a fixed dose of two units per transfusion. The dissemination of pocket-sized informative materials can serve as a useful reminder to establish these new practices. This study, conducted in a tertiary hospital in Catalonia, evaluates adherence to the updated recommendations by analysing the prescription patterns of red blood cells concentrates before and after an educational campaign. Specifically, we assess the impact of providing pocket-sized informational materials to healthcare providers responsible for prescribing transfusions.

Aims: This study evaluates the effectiveness of implementing a single-unit red blood cell (RBC) transfusion policy in a 350-bed tertiary hospital in Catalonia, both before and after the implementation of an informative campaign conducted between December 2019 and March 2020. The study aims to determine whether continuing training initiatives contribute to sustained adherence to the single-unit transfusion policy.

Methods: We conducted a retrospective analysis using the hospital's clinical workstation management tool (SAP-BO) to assess the percentage (%) of single-unit RBC transfusion requests for non-actively bleeding patients, comparing data from 2016-2018 (5750 requests) to 2020-2022 (9828 requests). Evaluation was performed both overall and by hospital service.

Results: A total of 15578 RBC transfusion requests were analysed, with 5750 from 2016-2018 and 9828 from 2020-2022. The percentage of single-unit RBC transfusion showed improvement post-intervention. Haematology, ICU and oncology were the top prescribing services, with percentages exceeding 60%.

Summary / Conclusions: Small interventions, such as disseminating pocket-sized informative material, can effectively shift entrenched practices like prescribing RBC transfusions in pairs. Haematologists play a crucial role in promoting and monitoring these recommendations, which are already gaining traction in other hospitals in Catalonia.

P830 - Table 1. Characteristics of transfusion incidents and transfusion rates. CMT: cross-matching tests. NM: near misses.

Year	Num. RBC	Num. of requests for RBC	Num. 1 RBC	% 1 RBC
2016	521	305	127	41.64
2017	4459	2635	1070	40.61
2018	4767	2810	1302	46.33
2019	Intervention			
2020	5094	3230	1761	54.52
2021	5411	3517	2011	57.18
2022	4614	3081	1888	61.28

P831 | Abstract withdrawn**P832 | Preoperative anaemia pathway—ensuring equitable patient management across Wales**K Towell¹, S Ditcham¹, J Gregory¹, C Jones¹, C Evans²¹Blood Health Team, Welsh Blood Service (Velindre University NHS Trust), Llantrisant, ²Cardiothoracic, Cardiff and Vale University NHS Trust, Cardiff, United Kingdom

Background: Variation in preoperative anaemia services across Wales led to excessive testing, multiple appointments, and delayed treatment. An All-Wales Perioperative Anaemia Pathway was developed in agreement with NHS Wales Health Boards (HBs) and issued by the Blood Health National Oversight Group (BHNOG) in 2021. The pathway promotes patient blood management by reducing variation in preoperative optimisation of anaemia and reducing avoidable perioperative transfusions. In 2022, Velindre NHS Trust and BHNOG secured funding from Value Based Health Care (VBHC) to support a programme to implement the pathway.

Aims: Collaborative working with preoperative assessment clinic (POAC) colleagues to implement the All-Wales Preoperative Anaemia Pathway to ensure equitable patient management for major elective surgery.

Methods: - Assess current anaemia management through engagement and benchmarking audits with HB stakeholders performing major surgery. Using surveys and data outlined by Commissioning for Quality and Innovation (CQUIN) definitions to evidence pre-implementation anaemia management. Development of a preoperative assessment screen (POAS) into national laboratory information systems (LIMS), aligned with the All-Wales Pathway and built upon reflex testing. Data dashboard for ongoing benchmarking agreed by stakeholders. Recurring funding for HBs to support anaemia management.

Results: Benchmarking audits in April 2023 determined 62.5% HB pathways aligned with the All-Wales pathway. HBs are working towards 100% alignment. Baseline data 2020 indicated 51.4% patients with Hb<130g/l had iron assessment to determine if iron deficiency was the cause. This increased to 68.5% (January 2024), as evidenced by the dashboard. POAS implemented in all HBs. HBs 3 and 5 were first to utilise POAS and demonstrate the most significant uplift in iron assessment in patients identified as anaemic. Evidence for the other HBs is yet to be captured due to later implementation. Impact of anaemia on length of stay (LoS), readmission and 1 year mortality evidenced in dashboard. Ongoing activity to link transfusion/IV iron data to evidence impact on patient outcomes and change of practice such as increased anaemia treatment with iron and reduction in avoidable transfusions.

Summary / Conclusions: To reduce avoidable transfusions, improve preoperative optimisation and improve patient outcomes following major elective surgery, an effective anaemia management pathway is crucial. A national pathway ensures anaemia management is standardised and equitable. VBHC funding has

provided necessary resources to ensure implementation of this pathway and the means to demonstrate the benefits of this. This resource is vital in driving the programme forward through continuous engagement with clinical teams from POACs and providing an opportunity to expand the benefits to other clinical areas within NHS Wales. A gap analysis is underway (January 2024) to determine current progress.

P833 | How do monitor quality indicators in transfusion medicine? A multicenter pilot studyM Bosch-Llobet¹, J M Garcia-Gala², G J Ferrer-Hidalgo³, V P Gonzalez Rodriguez⁴, L Guerra Dominguez⁵, A Laeref⁶, M Morales Sanz⁷, V Pons Escoll⁸, L Ramiro Infante⁹, N Rodriguez Torres¹⁰, P Solves Alcaina¹¹, M Vilariño Lopez¹², J Zubicarai Salegui¹³¹Banc de Sang i Teixits, Barcelona, ²Hospital Universitario Central de Asturias, Oviedo, ³Banc de Sang i Teixits, Hospital Sant Pau, Barcelona, ⁴Hospital Universitario Miguel Servet, Zaragoza, ⁵H. Universitario de Gran Canaria Dr. Negrín, Las Palmas de Gran Canaria, ⁶Centro de Transfusión, Tejidos y Células., Almería, ⁷Hospital Universitario de Guadalajara, Guadalajara, ⁸Banc de Sang i Teixits, Hospital Universitario Vall d'Hebron, Barcelona, ⁹Banc de Sang i Teixits, Hospital Joan XXIII, Tarragona, ¹⁰Hospital Universitario Virgen del Rocío, Sevilla, ¹¹Hospital Universitario Politécnico La Fé, Valencia, ¹²Hospital Clínico Universitario Santiago de Compostela, Santiago de Compostela, ¹³Hospital universitario Niño Jesús, Madrid, Spain

Background: Blood transfusion is a critical component of healthcare that requires efficient and safe management in the hospital setting. Quality indicators (QIs) in transfusion medicine have long been used at institutional or local level. However, there appears to have been a lack of widespread discussion about the importance of implementing, monitoring and comparing transfusion QI. Little is known about the actual application of QI in hospitals.

Aims: To assess the impact of monitoring specific transfusion QI in a diverse hospital context. Data on traceability, logistical supply, and response time and transfusion process were collected over a 9-month period. In addition, benchmarking was carried out to compare results between participating hospitals. Challenges related to indicators that rely on non-automated data, such as audits of transfusion use and adherence to guidelines, were identified.

Methods: Twelve self-selected public hospital transfusion services (HTS) agreed to implement a tailored QI. Data on traceability, blood supply, response times, haemovigilance and transfusion process were systematically collected during the 9-month pilot. A benchmarking process was used to facilitate comparison of results. Challenges were identified, particularly for indicators dependent on non-automated data, highlighting the difficulties in conducting audits related to transfusion use and adherence to guidelines.

Results: Despite progress, difficulties were noted with indicators that rely on non-automated data, particularly in conducting audits of transfusion use and adherence to clinical guidelines. Despite these

challenges, an overall improvement was observed across several indicators, highlighting the need to address data automation in specific areas. Response rates to QI related to blood supply and haemovigilance were 92% and 93% respectively. Traceability monitoring and QI related to the transfusion process were achieved in half of the HTSs (56% and 46%). Some differences in red blood cell (RBC) use per hospital discharge were found, not always related to complexity or hospital activity. None of the HTS could provide data on blood use in different medical and surgical settings, nor on the time of urgent transfusion (turnaround time). The use of ORh-D negative RBCs deserves special mention: one third of units (31.2%) were given to non-O Rh-D negative patients. In three hospitals, this percentage is almost half or more (46.3%, 44.5% and 64.9%). In addition, only one third of HTSs apply the Recommendations for the use of group O RBCs - 2019-AABB Association for Advancement of Blood & Biotherapies.

Summary / Conclusions: The implementation of the transfusion QI in 12 hospitals was successful, demonstrating the practical utility of the selected indicators in day-to-day blood bank management. Despite the challenges associated with non-automated data, the identification of areas for improvement and the application of benchmarking provide a sound basis for future system optimization. Emphasizing the importance of progressing towards full data automation in transfusion management is paramount to the continuous improvement of the field.

P834 | Patient-level key performance indicators and maximum surgical blood ordering schedule in vascular surgeries in a tertiary public hospital in Athens

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Background: An adequate supply of quality blood products with the minimal wastage of resources, ensuring patient safety is basic requirement in any hospital. Excessive blood ordering and absence of a specific schedule regarding the request of Red Blood Cell units (RBCs) are among the most common problems in hospitals, which lead to increased costs and waste of limited blood resources. Monitoring the blood transfusion process with patient-level Key Performance Indicators (KPIs) and having a Maximum Surgical Blood Order Schedule (MSBOS), as parts of Patient Blood Management (PBM) strategy are useful to minimize the use of blood products and improve patient's outcomes. According to studies conducted by the local Hospital

Transfusion Committee (HTC), 39 % of RBCs transfused during surgeries are used in vascular surgeries.

Aims: To evaluate the pattern of blood transfusion requests and utilization using indicators and to design an MSBOS for vascular surgeries.

Methods: In this retrospective, cross-sectional study, all vascular patients who underwent elective or emergency vascular surgery, at the Red Cross "Korgialenio-Benakio" General Hospital of Athens, from January 1st, 2022 to December 31st, 2022, were included. The data collected consisted of the number, sex, age of patients, number of surgeries, number of hospital admissions, Hb (g/dL) concentration at hospital admission, as well as the type of surgery, number of RBCs crossmatched and number of RBCs transfused. Using the appropriate data, the indicators: % of patients with anaemia at admission, Cross-matched to Transfusion Ratio (CTR), Transfusion probability (T%) and Transfusion Index (TI), were calculated. Additionally, MSBOS was calculated as one and a half times the TI for each surgical procedure.

Results: In the present study 182 patients (149 males and 33 females), who underwent a total of 218 elective and emergency surgeries in 203 hospital admissions, were included. The mean age of the 182 patients was 70 years (median:71, range:36-94). The mean Hb concentration at admission was 12 g/dL (median: 12.4 g/dL, range: 5.7-16.5 g/dL). According to the World Health Organization's definition of anaemia (Hb<13 g/dL for males and Hb<12 g/dL for females), 62 % of patients were found to be anaemic at admission. A total of 684 RBCs were cross-matched in 218 surgeries and 305 were transfused in 126 surgeries. The overall results of the CTR, T% and TI for all type of surgeries (amputations, abdominal aortic aneurysm repairs, bypass with graft, endarterectomies etc.) were 2.24 (684/305), 57.80% (126*100/218) and 1.40 (305/218), respectively. Indicatively, CTR, T%, TI and MSBOS for open abdominal aortic aneurysm repair were 2.07, 73.33, 1.9, 2.85 (rounded up to 3) and 3, 83.33, 1.17 and 1.75 (rounded up to 2) for bifemoral bypass.

Summary / Conclusions: It appears that pre-operative anaemia had a decisive influence on the number (684) of cross-matched RBCs but not on the number (305) of transfused RBCs intra-operatively. The overall CTR, T%, TI indicate significant blood usage. Accurate MSBOS protocols can help towards the proper requisition and utilization of blood and lead to appropriate use of blood stocks minimizing wastage of human and economic resources, and eventually, promote patient safety.

P835 | Intravenous Iron versus oral Iron for patient blood management in major orthopaedic surgery—an equivalence study

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Background: Patient blood management (PBM) is an evidence-based approach to minimize the use of allogenic blood components,

ensuring restrictive transfusion strategies. The preoperative identification of patients with anaemia or in risk of and their individualized treatment with iron supplementation is a strategy to ameliorate patient's physiological tolerance towards anaemia. PBM protocols mainly use IV iron replacement. While it is the quickest form to correct iron deficiency, it is a costly procedure. On the other hand, oral iron is an affordable, easily accessible and well tolerated alternative.

Aims: To assess equivalence of oral Iron (III) hydroxide polymaltose complex with intravenous (IV) Iron carboxymaltose in a PBM protocol in major orthopaedic surgery.

Methods: A statistical analysis using IBM SPSS® Statistics was conducted comparing the two treatments arms, with an outcome of reducing transfusion support needs. Iron deficiency was defined as a transferrin saturation <20% and a ferritin level <30ng/mL. Iron deficiency anaemia was defined as the former cut-offs with a haemoglobin level <13g/dL or <12g/dL in men and women, respectively. The decision between IV and oral iron was made considering the patient's comorbidities and the time available until surgery. Data was collected retrospectively from an informatic PBM database. Major orthopaedic surgeries considered were hip replacement, knee replacement, hip revision surgery, knee revision surgery and idiopathic scoliosis correction. All were elective procedures, completed during 2022 and 2023. Transfusions were considered for the whole inpatient admission period.

Results: A total of 96 patients treated preoperative with iron were included (table 1). Among these, 52 patients received IV Iron carboxymaltose and 44 patients received oral iron replacement as Iron (III) hydroxide polymaltose complex, with 100mg elemental iron per day. The IV group received the therapy with a median of 38.5 (IQR 50.3) days before surgery. The oral replacement group started therapy 85 (IQR 79) days prior to surgical treatment. Patients who received IV iron required a mean of 0.31 (± 0.76) red blood cell units and patients on oral iron replacement required a mean of 0.36 (± 0.69) red blood cell units. The two one-sided t test (TOST) analysis comparing the efficacy of oral iron and intravenous iron

revealed a combined p-value of 0.006, suggesting statistical evidence in favour of equivalence between oral and intravenous iron treatments for PBM.

Summary / Conclusions: We concluded that, given sufficient time, oral iron replacement provides equivalent results to IV iron replacement. In our centre, the direct costs of IV iron amount to 267€, while the oral iron formulation ranges from 9 to 18€. The total cost difference, however, is underestimated, as the IV iron is also accompanied by many indirect costs (days missed from work or school; travelling to the care centre; day-hospital admission). Therefore, oral iron replacement provides a much cheaper option for PBM, with equivalent efficacy.

P836 | Knowledge and practice among clinicians regarding patient blood management initiatives

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Background: Patient Blood Management (PBM) is a multimodal, multidisciplinary patient-centered strategy aimed at improving patient's own blood. Anemia and transfusion have been associated with increased morbidity and mortality in surgical patients, and the systematic application of PBM programs in the perioperative period has been consistently found to improve patients' clinical outcomes following surgery. Hospitals must strive towards the implementation of PBM that represents a new quality and safety standard.

Aims: This study aims to assess the knowledge, attitude and clinical practice of physicians who prescribe blood and its components to their patients regarding the PBM initiatives.

Methods: A survey-based cross-sectional study was conducted in 2021. The data was collected using a structured self-administered questionnaire distributed via online communication tools to the hospitals in Saudi Arabia. The population under study consisted of physicians practicing in the following specialties: Surgery, Gynecology & Obstetrics, ICU, and Anesthesia. The questionnaire consisted of physician's demographic and practice information, and the basic knowledge about PBM. The questionnaire was in the form of MCQs that consisted of five sections: Physician's demographic information, assessment of perioperative bleeding risk, monitoring of perioperative bleeding, management of perioperative bleeding and blood-conserving strategies. Analyses were performed using SPSS, software version 25. Ethical approval was obtained from the ethical committee at King Abdulaziz University Hospital, and a consent was taken from each participant before collecting the data.

Results: We received responses from 102 physicians. Most of them 43 (33.3%) were surgeons, and the rest were from anesthesia,

P835 - Table 1. Demographics, and type of surgery of patients submitted to elective major orthopaedic surgery treated with preoperative iron

	IV Iron	Oral Iron
Number of patients	52	44
Female/Male	43/9	39/5
Mean Age (Standard Deviation)	49.1 (± 27.2)	45.5 (± 28.9)
Iron Deficiency	17	31
Iron Deficiency Anaemia	35	13
Hip Replacement	13	11
Knee Replacement	15	12
Hip Revision	3	1
Knee Revision	2	1
Idiopathic Scoliosis	19	18

P836 - Table 1 shows the statistical significance for the knowledge about PBM and its related policies, protocols and guidelines.

Question	No N (%)	Yes N (%)	Don't Know N (%)	p-value
Did you hear about PBM initiatives?	63 (61.8%)	36 (35.3%)	3 (2.9%)	0.038788
Do you have a written protocol, guideline, or policy about the management of perioperative bleeding?	51 (50%)	22 (21.6%)	29 (28.4%)	0.003496
Do you have a written protocol, guideline, or policy about massive transfusion?	45 (44.1%)	29 (28.4%)	28 (27.5%)	0.190
Are you aware about management of perioperative bleeding?	47 (46.1%)	55 (53.9%)	0 (0%)	0.001
Did you attend a presentation/discussion about the management of perioperative bleeding?	71 (69.6%)	31 (30.4%)	0 (0%)	0.04
Do you believe that the current management of perioperative bleeding could be improved?	8 (7.8%)	75 (73.5%)	19 (18.6%)	0.317

intensive care, and obstetrics and gynecology divisions. The majority of these physicians were from governmental hospitals 75 (73.5%). Only 19 (18.6%) were consultant physicians.

Table 1: Statistical significance for the Knowledge about PBM.

Summary / Conclusions: The level of knowledge about transfusion practices is still inadequate. More training is necessary for prescribers of blood products. Learning from the working experiences, awareness, education and training on PBM are so important to set the evidence of health provider practices and to ensure medical and surgical outcomes by managing and preserving a patient's blood according to health care practice. These findings provide a baseline to develop joint action plans to further implement and strengthen PBM across hospitals in Saudi Arabia.

P837 | Management of anemia in patients with pre-end stage chronic kidney disease in a tertiary medical center in Taiwan

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Background: Reducing red blood cell (RBC) transfusions is a goal in managing anemia among patients with chronic kidney disease (CKD), although the usage still remains high in Taiwan. Understanding anemia management can help formulate strategies to reduce RBC transfusions among CKD patients, but it is undetermined, especially for non-dialysis patients.

Aims: To explore anemia management among patients with pre-end stage renal disease (pre-ESRD) in Taiwan and determine the risk factors associated with RBC transfusions.

Methods: We identified patients aged over 20 years with stages 3b, 4, or 5 CKD who were enrolled in the pre-ESRD care program at a tertiary medical center between 2007 and 2019 and retrieved their clinical data until December 2020. The treatments for anemia that we focused on included erythropoiesis-stimulating agents (ESAs), iron

agents, and RBC transfusions. In Taiwan, reimbursement for ESAs under the National Health Insurance in CKD patients is restricted to those in stage 5 CKD. The classification of CKD stages was determined based on the estimated glomerular filtration rate using the simplified Modification of Diet in Renal Disease equation. Anemia was defined as hemoglobin (Hb) <13 g/dL for males and <12 g/dL for females, categorized into five groups: Hb≥10, 9≤Hb<10, 8≤Hb<9, 7≤Hb<8, and Hb<7. We further defined the anemia episodes in CKD patients, which commenced upon the detection of anemia and terminated when either reaching normal Hb levels, worsening to lower Hb groups, or after 30 days from the initial onset of anemia, whichever came first. The anemia episodes occurring after amelioration and transitioning out of the pre-ESRD stage, after the initiation of dialysis, or following kidney transplantation were excluded. We used generalized estimating equations to determine the risk factors associated with RBC transfusions during anemia episodes.

Results: This study included 7113 pre-ESRD patients, comprising 2073 at Stage 3b, 2564 at stage 4, and 2476 at stage 5. Among them, 1314 (63.4%), 2186 (85.3%), and 2441 (98.6%) patients had anemia at stages 3b, 4, and 5 of CKD, respectively, when they were enrolled in the pre-ESRD program. We identified 85036 anemia episodes, and the treatments during these episodes which initiated with Hb≥10, 9≤Hb<10, 8≤Hb<9, 7≤Hb<8, and Hb<7 were as follows: 0.6%, 2.9%, 14.0%, 48.7%, and 76.8% received RBC transfusions; 14.1%, 31.0%, 39.1%, 42.1%, and 44.5% received ESAs; and 10.1%, 20.5%, 22.9%, 24.4%, and 25.2% received irons, respectively. We observed a slightly higher proportion of anemia episodes treated with RBC transfusions among patients with Stage 3b or 4 CKD than those with stage 5 CKD within the same Hb group. Multivariable analysis showed that older age, male gender, lower Hb levels, non-use of ESA/iron, a history of cancer, undergoing invasive procedures, and a prior history of RBC transfusion were associated with an increased risk of receiving RBC transfusions. In a subgroup analysis of anemia episodes with Hb≥9, patients with Stage 3b or 4 CKD had an increased probability of receiving RBC transfusions than those with stage 5 CKD (OR = 1.70, p = 0.0011).

Summary / Conclusions: A high proportion of pre-ESRD patients with anemia received RBC transfusions, and those in Stage 3b or 4 CKD

were more likely to receive RBC transfusions at a relatively liberal Hb threshold than those in stage 5 CKD. Further studies are needed to examine the impacts of RBC transfusion on these patients and minimize the usage through proactive patient blood management.

P838 | Appropriateness of platelet transfusions according to the transfusion indications at two tertiary hospitals

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Background: Appropriate platelet transfusion is important both in terms of patient blood management and the allocation of limited healthcare resources.

Aims: We evaluated the appropriateness of platelet transfusions at two tertiary hospitals.

Methods: At Chonnam National University Hospital (Hospital A) and Chonnam National University Hwasun Hospital (Hospital B), 1470 platelet transfusions (299 and 1171 cases at Hospitals A and B respectively) during a single month were retrospectively reviewed by reference to the Korean transfusion guideline (5th edition). The transfusion indications were categorized into four as follows: clinical judgments (regardless of platelet count), therapeutic purpose, prophylaxis for patients at risk for spontaneous bleeding, and prophylaxis before surgery or procedures. The chi-square or Fisher's exact test was used to compare the rate of transfusion indication and appropriateness between Hospital A and B.

Results: The most common indications were therapeutic transfusion to ensure hemostasis (54.8%) at Hospital A and to prevent spontaneous bleeding in patients with hematologic/oncologic diseases (65.8%) at Hospital B. In four categories of indications, all except for prophylaxis before surgery or procedures were significantly different between Hospital A and B ($p < 0.01$ or < 0.001). Overall, 87.3% and 76.3% of transfusions were appropriate at Hospitals A and B, respectively ($p < 0.001$). According to the various transfusion indications, the therapeutic transfusions at both hospitals were appropriate in more than 80% of cases; the appropriateness for preventive transfusions to address spontaneous bleeding were 80.7% and 69.3%, respectively ($p < 0.05$), and those before surgeries or invasive procedures were 72.0% and 66.2%. Of the 38 and 278 inappropriate transfusions at Hospitals A and B (as revealed by the pre-transfusion platelet counts), most cases exhibited platelet counts from 50 to $100 \times 10^9/L$ at Hospital A (23 cases) and from 20 to $50 \times 10^9/L$ at Hospital B (198 cases).

Summary / Conclusions: The two hospitals significantly differed in terms of transfusion indications, appropriateness, and instances of inappropriateness. The indications and appropriateness for platelet transfusion should be primarily reviewed on each hospital basis in real practice; this would be helpful to improve patient blood management.

P839 | Evaluation of perioperative anemia management in Japan—a multicenter retrospective observational study

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Background: Perioperative anemia correlates with long-term hospitalization and increased mortality rates. Patient blood management (PBM), an evidence-based, systematic approach for diagnosing and treating perioperative anemia appropriately, reduces the need for blood transfusions and improves patient outcomes. However, the significance of PBM remains insufficiently recognized in Japan.

Aims: To evaluate the current status of perioperative anemia management in Japan.

Methods: This study was a national, multicenter retrospective observational analysis. Patients were included if they underwent surgery for gastrointestinal or gynecological tumor resection or artificial joint replacement in 16 Japanese medical institutions between October and December 2019. Baseline characteristics and clinical and laboratory data of the patients were extracted from electronic medical records and analyzed.

Results: The study included 2248 surgical procedures (patient average age, 62 years; woman 67.7%), comprising 968 gastrointestinal tumor resections, 913 gynecologic tumor resections, and 367 artificial joint

P839 - Table 1. Type of surgery according to preoperative hemoglobin levels. Values are numbers (proportion).

	Hb<13g/dL	Hb<12g/dL	Hb<11g/dL
Total	985 (49.8%)	589 (29.8%)	335 (16.9%)
Gastrointestinal tumor resection	447 (52.0%)	307 (36.3%)	195 (23.1%)
Gynecologic tumor resection	379 (47.1%)	206 (25.6%)	105 (13.1%)
Artificial joint replacement	159 (48.2%)	76 (23.0%)	35 (10.6%)

Serum ferritin levels were measured in 142 patients with preoperative anemia; among them, 44 (31.0%) had serum ferritin $< 20\text{ng/mL}$, while 55 (38.7%) had levels ranging from 20 to 100ng/mL . Of the 100 preoperative anemic patients, 11 received low-dose iron intravenously and 89 received it orally. However, no significant difference was observed in the average hemoglobin level on the day of surgery between preoperative anemic patients with and without iron treatment. A total of 280 allogeneic and 219 autologous perioperative red blood cell (RBC) transfusion episodes were reported. Among these, 135 (48.2%) autologous RBC transfusions were administered to patients undergoing joint replacement surgery. Notably, despite intraoperative blood loss of $< 500\text{ mL}$, 28.1% of patients with preoperative anemia (Hb $< 11\text{g/dL}$) received RBC transfusions.

replacements. Across all surgical procedures, we found 985(49.8%), 589(29.8%), and 335(16.9%) patients with preoperative hemoglobin levels <13g/dL, <12g/dL, and <11g/dL, respectively (Table 1). In gastrointestinal tumor resection, we found 447(52.0%), 307(36.3%), and 195(23.1%) patients with preoperative hemoglobin levels <13g/dL, <12g/dL, and <11g/dL, respectively. In gynecological tumor resection, we found 379 (47.1%), 206 (25.6%), and 105 (13.1%) patients with preoperative hemoglobin levels <13g/dL, <12g/dL, and <11g/dL, respectively. In artificial joint replacement, we found 159 (48.2%), 76(23.0%), and 35 (10.6%) patients with preoperative hemoglobin levels <13g/dL, <12g/dL, and <11g/dL, respectively.

Summary / Conclusions: Our study reveals the current status of PBM in Japan. The findings indicate that low-dose iron treatment shows an insufficient effect on preoperative anemic patients. A more proactive approach to addressing preoperative anemia may be necessary to optimize the number of perioperative RBC transfusions.

P840 | Abstract withdrawn

P841 | Abstract withdrawn

P842 | Pre-operative patient blood management practices among patients undergoing elective major orthopedic surgery at National Hospital of Sri Lanka

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Background: Patient blood management (PBM) is a broader concept. The intension is to enhance patient outcome, encourage optimal use of blood and minimize health care cost. Focus is to address and modify the risk factors which increase the need of allogenic transfusions. Preoperative anaemia in surgical patients is common and associated with significant morbidity, mortality, increased blood transfusions and transfusion related adverse effects. National hospital of Sri Lanka (NHSL) being the first and largest in Sri Lanka does not have an established PBM protocol in practice yet. Implementation of PBM requires a change in culture, behaviour, structural adjustments and redirection of health care resources.

Aims: The study aimed to assess preoperative PBM practices among patients undergoing elective major orthopedic surgeries at NHSL. By identifying current practices, their limitations, and effects on patient outcome we intend to establish nationally accepted effective PBM practices in Sri Lanka.

Methods: We conducted a retrospective descriptive study in the orthopedic surgical unit, NHSL. All patients who underwent surgery during three months were recruited. Data analysis was done by SPSS statistical package version 26. Descriptive analysis of categorical and continuous variables were done. Categorical variables were reported as percentages and frequencies. They were compared with chi-square test and Pearson correlation. Continuous variables were expressed as means and standard deviations.

Results: Study population consists of 301 patients with 56.14% males. Age range was 12-103 years. Mean age of male (47.38) and female (55.12) patients showed a significance of $p < 0.001$. Pre-operative anaemia was identified in 65.12%. 5.98% were severely anaemic. Use of iron and vitamin supplementation were minimal during preoperative period. None received parenteral iron or rHuEpo. 19.38% received blood transfusions. Antiplatelet drugs were discontinued prior to surgery in all patients. None received platelet transfusions. Pre-operative anaemia in female and male patients were 53.6% and 46.4% respectively. Mean pre-operative Hb for female and male was 10.59 g/dL and 12.43 g/dL. Age of the patient and pre-operative Hb value showed negative correlation at a lower strong level. 24.58% of total population received transfusions. All severely anaemic patients received at least one transfusion. Mean number of units transfused is 1.78. Mean age for transfusion was 66.27 years. Mean age of transfused and non-transfused groups showed statistically significant difference. Gender and transfusion requirement showed a significant association with higher transfusion need in females ($p < 0.05$). No significant difference noticed between gender and number of days of hospital stay. Significant difference noticed between transfused and non-transfused groups in length of hospital stay.

Summary / Conclusions: Socio demographic data in orthopedic surgical patients varies depending on the selected cohort. Preoperative anaemia, severity of anaemia, increasing age, female gender increase the likelihood of blood transfusions. Transfusions increase length of hospital stay. Oral and/or parenteral iron, rHuEpo, vitamin B12, folic acid and blood transfusions were used to correct preoperative anaemia. Platelet/FFP transfusions in orthopedic surgery was found to be less common. Unavailability of local policy led to variations in patient management. Establishment of a nationally accepted PBM protocol will empower universal and optimum patient management.

P843 | Associations between third trimester hemoglobin level and incidence of post-partum hemorrhage

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Background: Maternal mortality was reported to be around 289,000 cases in 2020 with a slight reduction from 309,000 cases in 2016 when Sustainable Developmental Goals (SDGs) was first launched. Reduction of maternal mortality has been one of the main goals for SDGs with the target of reducing global Maternal Mortality Rate (MMR) to less than 70 deaths per 100,000 live births in 2030. According to the data reported in a World Health Organization (WHO) systematic analysis, haemorrhage was documented as the leading cause of maternal death worldwide (27.1%) with two third of the cases were classified as Postpartum Haemorrhage (PPH). Anaemia is defined as haemoglobin <11.0g/dl in pregnant population by WHO classification. It is a well-known complication of pregnancy and can be associated

with maternal and fetal morbidity. Anaemia has been recognized as a public issue in the developing countries and has been linked to an increase risk of PPH in some studies.

Aims: There are two objectives from this study. The first objective is to determine the association between 3rd trimester anaemia (Hb <11.0 g/dl.) and the risk of PPH and the second objective is to describe the transfusion outcome of patients with third trimester anaemia during delivery /postpartum.

Methods: This was a prospective cohort study design conducted from 15th December 2021 until 14th December 2022. Patients were recruited during antenatal check up at the outpatient clinic of Obstetrics & Gynaecology Clinic in Woman & Child Hospital Tengku Azizah (HTA), Kuala Lumpur, Malaysia. A total of 125 patients who have anaemia during the third trimester and another 125 patients who do not have anaemia during the third trimester were included in the study. However due to missing data and dropouts the final number recruited was 230 patients. The patients were followed up to see whether they developed PPH or not during delivery. The patients' delivery information and transfusion records will be retrieved from the Hospital Information System after the patients delivered.

Results: A total of 230 patients recruited for the study where 110 (47.8%) underwent Lower Section Caesarian Section (LSCS) and 120 (52.2%) underwent Spontaneous Vaginal Delivery (SVD). Out of this number 124 of the patients developed mild anaemia whereas 106 had hb > 11.0g/dl. Out of 124 patients who had anaemia 71 (57.3%) had mild anaemia whereas another 39 (31.5%) patients had moderate anaemia. No severe anaemia detected in our cohort of patients. Only four patients had PPH and all these four patients had moderate anaemia and all of them underwent LSCS.

Summary / Conclusions: The findings in our study were consistent with other studies where PPH was found to be associated with moderate and severe anaemia but not with mild anaemia. Unfortunately only few studies have addressed the causal-relationship between severe anaemia and PPH. Up until this date there was no exact value of Hb at which the potential uterine atony could be imminent and the link between low hb levels at delivery & risk of PPH is still currently under debate. The hypothesis behind this is that anaemia will decrease myometrium contractility and/or impair coagulation due to low Hb levels. Theoretically maternal anaemia greatly increases the risk of PPH simply because anaemia reduces the oxygen-carrying capacity of the blood hence anaemic women cannot tolerate the same volume of blood loss as healthy women.

P844 | Abstract withdrawn

P845 | B-thalassemia major—transfusion management challenges

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Background: β -thalassemia major remains a public health problem, particularly in high-prevalence areas

Aims: To reinforce transfusion safety in patients with β -thalassemia major from the moment of diagnosis through a specialized management protocol, in order to prevent immuno-hematological complications and avoid recourse to emergency transfusions

Methods: This study involved 113 patients with β -thalassemia major benefiting from a hypertransfusion program with 15 to 20 ml/kg of Red Blood Cell Concentrate every three to four weeks. Irregular Agglutinins Test was performed regularly, a Direct Antiglobulin Test in the event of signs of hemolysis, compatibility with an indirect antiglobulin test for alloimmunized patients and selection of Red Blood Cell Concentrate respecting the specificity of the identified allo antibodies in order to prevent post-transfusion immunological hemolysis. The aim was to maintain a pre-transfusion Hb level of 09-10 g/dl

Results: 45.13% of our patients had a standard Rhesus and Kell phenotype C+c+E-e+K-, Irregular Agglutinin Testing was positive in 16 patients (14.16%), 22 allo-antibodies were identified belonging essentially to the Rhesus and Kell systems, 05 patients were poly-immunized, 11.50% of patients had a transfusion rate higher than 15 ml/Kg every 03 weeks, due to the patient's unstable pre-transfusion status (allo-immunization, Splenomegaly...). a clear improvement in transfusion yield was observed for the majority of patients, with a satisfactory increase in Hb levels and a respected interval between two successive transfusions.

Summary / Conclusions: A well-adapted transfusion protocol can help prolong life expectancy and improve quality of life for patients with β -thalassemia major.

P846 | Frequency of anemia in the cardiovascular surgery department

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Background: Anemia represents a major global concern, arising from various factors and involving complex pathological mechanisms. The impact of anemia on hospitalized patients is considerable, increasing the risk of postoperative complications, morbidity, and the length of hospital stay.

Aims: The aim of this study was to analyze the frequency of anemia among patients hospitalized in the cardiovascular surgery department, and to evaluate the risk factors that could be associated with the development of anemia.

Methods: A retrospective study was conducted on patients admitted to the cardiovascular surgery department of Rabta Hospital over a three-month period, from January 2023 to March 2023. Demographic and clinical characteristics of patients were extracted from medical records, and hemograms were performed upon admission, post-operatively, and at discharge. These hemograms were performed in the hematology laboratory using the SYSMEX Automated Hematology Analyzer. The data collected were analyzed using SPSS software version 24.

Results: A total of 402 patients were included, among whom 49.3% ($n = 198$) were diagnosed with anemia, of which 7% ($n = 13$) acquired it during their hospital stay. Among anemic patients, 69.2% ($n = 137$) were male (sex ratio 2.2), with a mean age of 58 years. Following the analysis of data on mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC), it was observed that normocytic normochromic anemia accounted for 69.2% of cases, hypochromic microcytic anemia for 29.8%, and macrocytic anemia for 1%. According to hemoglobin values, patients were categorized into those with mild anemia (hemoglobin > 9 g/dL), accounting for 54% ($n = 106$) of cases, and those with moderate to severe anemia, comprising 46% ($n = 92$) of patients, with 38.1% ($n = 35$) of them receiving a blood transfusion. Among the risk factors implicated in the acquisition or exacerbation of anemia in hospitalized patients, extreme ages (0-93 years), average length of hospital stay of 6 days, and surgical interventions, of which 52% presented a severe hemorrhagic risk, were noted. Additionally, 7.7% of patients with severe anemia died.

Summary / Conclusions: This study reveals a significant incidence of anemia (49.3%) among patients hospitalized in cardiovascular surgery, with 46% presenting moderate to severe anemia, sometimes requiring transfusion. Risk factors such as extreme ages, duration of hospitalization, and surgical interventions with hemorrhagic risk must be considered in the management of these patients.

P847 | Bloodless surgeries—two similar approaches to two distinct Jehovah Witnesses patients before high hemorrhagic risk procedures

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Background: Jehovah Witnesses' (JW) refuse blood transfusion due to religious beliefs. This poses a challenging scenario in the event of high bleeding risk interventions, such as neurosurgery or orthopedic surgeries. On the other hand, anemia is widely prevalent in the general population, especially in the elderly. When all combined, doubts in patients' management arise.

Aims: We report two clinical cases of two elderly JW women who presented both baseline anemia and high-risk hemorrhagic surgeries.

Methods: We reviewed the clinical records of two JW patients referred to our institution in the patient blood management (PBM) setting.

Results: Case #1

Seventy-one-year-old female JW referred to our institution for symptomatic frontoparietal meningioma (46 × 56 × 46 mm) elective

treatment presenting mass effect and midline structures deviation. Patient had history of gastrectomy due to gastric adenocarcinoma, hemicolectomy due to colorectal cancer, paraneoplastic pulmonary embolism (under full dose apixaban), major depression, anemia of chronic disease and mixed deficiencies (baseline hemoglobin [Hb]: 11-12 g/dL) and frailty; G 4 P 4. She reported no personal or familial bleeding disorders. Patient was included in Patient Blood Management (PBM) program. Patient's workup revealed Hb 11.8 g/dL, reticulocytopenia, total iron deficit (Ganzoni's): 800 mg, normal eGFR. She underwent iron sucrose infusions (total: 1000 mg), folic acid PO and cyanocobalamin IM supplementation. Due to lack of response, she was switched to ferric carboxymaltose IV (total: 500 mg) and epoietin beta (rhEPO) 600 UI/kg sc and presented favorable response (Hb 13.3 g/dL). Before surgery patient stopped oral anticoagulation and underwent successful tumor chemoembolization at day -1 (D-1). Tumor was resected under tranexamic acid (TXA) 10 mg/kg IV (D0). Patient repeated twice rhEPO due to postoperative anemia (Hb 11.3 g/dL and active blood loss). Neurological deficits resolved. Discharge occurred at D5 (Hb 14.4 g/dL).

Case #2

Seventy-one-year-old female JW admitted at our institution after fall and post-traumatic spine fractures (D6-D7 levels). She was referred for open technique orthopedic surgery. She had history of obesity, dyslipidemia, major depression, and iron deficiency anemia (baseline Hb: 9 g/dL). Patient's workup revealed Hb 6.5 g/dL, total iron deficit (Ganzoni's): 1600 mg, normal eGFR. Due to anemia severeness, patient underwent ferric carboxymaltose (1000 mg) IV, iron sucrose IV (total: 600 mg), began rhEPO (600 UI/kg sc) and folic acid supplementation (5 mg p.o.). After two weeks she recovered to Hb 10.3 g/dL and underwent surgery under TXA 10 mg/kg IV. She lost approximately 200 mL of blood and maintained Hb 9 g/dL postoperative. After surgery 2 additional rhEPO doses were administered. Patient was discharged at D5.

Summary / Conclusions: In the face of life-threatening situations, the unavailability of blood transfusion poses practical problems. Since both patients had baseline anemia and were well adapted, their management was facilitated. Hemostatic agents and hematopoiesis stimulating agents are valid approaches when a patient cannot be transfused (Lawson, Br J Anaesth, 2015; Crowe, Curr Opin Hematol, 2019).

P848 | Transfusion in the emergency unit of a tertiary hospital—results of the development of a patient blood management program

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Background: Patient Blood Management (PBM) is a systematic evidence-based approach that aims to optimize patient management and transfusion to ensure effective and quality patient care. There is little literature on the usefulness of PBM in hospital emergency departments (ED).

P848 - Table 1 Characteristics of the patients

	Jan/22	Oct/23
RBC transfused / patients	216 / 110	181 / 106
Mean age (years)	72.29 (SD 18.37)	69.7 (20.24)
Sex (% women)	44.3	44.9
Mean Charlson comorbidity index	6.08 (SD 2.93)	6.39 (SD 3.87)
Diagnoses according to clinical history	anemia (74.8%), bleeding (21.4%) others (3.8%)	anemia (65.4%), bleeding (21.5%) others (13.1%)
Diagnosis of anemia in patient clinical records	48.1%	56.1%
Oral anticoagulant treatment	51.9%	70.4%
Patients had received iron (Fe) in last year	33.6%	44.1%
Patients transfused in previous 3 months	21.4%	27.1%
Mean Hb	6.92 (SD 1.4)	7.22 (SD 1.70)
Infusion times (60-90 minutes)	52.04%	40.35%
Diuretics (before / after)	71% (23.7% / 47.3%)	48% (15.9% / 32.1%)
AEs without diuretics vs diuretic	1.8% vs 13.2% $p = 0.01$	
Number of RBC Mean (% patients transfused 2 RBC in the episode)	1.96 (59.5%)	1.70 (56.60%)
Discharged at home vs hospitalized	40.5% vs 59.5%	43.4% / 56.6%

P848 - Table 2 Incidence of transfusion-associated adverse effects (AEs)

Adverse Effect	2022 N (%)	2023
Heart Failure	6 (4.58)	4 (3.77)
Hepatic decompensation	2 (1.52)	-
Fever	1 (0.76)	-
Dyspnea	1 (0.76)	-
Pneumonia	1 (0.76)	-

Aims: The objective of the study was to analyze the characteristics of patients who received red blood cell (RBC) concentrates in ED and to assess possible PBM strategies. And after implementing PBM measures, reanalyze and find differences in ED transfusional practice

Methods: Analytical, observational, cross-sectional study of patients transfused with RBC in the ED of a tertiary hospital. We included all patients who received RBC in the ED in January 2022. After an educational program on emergency physicians and nurses a new observational, cross-sectional study of patients transfused with RBC in October 2023 was performed. Data were analyzed using the statistical program SPSS.

Results: are shown in Table 1 and Table 2

Summary / Conclusions: The transfusion-associated AEs observed in 2022 in our study were higher than those reported in the literature. We proposed in the educational program transfusion one RBC and iron treatment over transfusion of 2 RBC, increase the time of infusion and individualize the patient risk of overload. The implementation of PBM strategies in ED led to an increased time of infusion and less % of AEs observed.

P849 | Abstract withdrawn

P850 | Abstract withdrawn

P851 | Management of a blood service through a pandemic using MSBOS in Central Institute of National Importance in India

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Background: The global impact of the COVID-19 pandemic has been profound, particularly in its effects on blood supplies. The unprecedented challenges posed by the pandemic prompted a comprehensive study to explore strategies for sustaining blood transfusion services during these critical times. This investigation is motivated by the imperative to address and fulfill the surging national demand for blood amidst the unique challenges posed by the pandemic.

Aims: (i) To assess the practice of blood utilization using various blood utilization indices and calculate the Maximal Surgical Blood Order Schedule for various categories of patients during COVID-19 period. (ii) To analyze the blood request forms in order to increase the efficiency of blood utilization and inventory management during COVID-19 period.

Methods: Data of Blood donation services were taken into consideration along with the issue details of all blood components were analysed from 1st April to 31st December 2020. The dates were decided as following the implementation of lockdown in country due to COVID 19. These data are analysed for utilization of blood

components, pattern of blood donation, reactive status of the donated blood in COVID 19 times.

Results: The crossmatch/transfusion ratio exhibited variations across different medical departments during the period from April to December 2020. The highest ratio was observed in the Gynecology department at 1.55, followed by the Ophthalmology department at 1.5, Trauma and Emergency at 1.46, and ENT at 1.35. Notably, a total of 382 out of 1886 blood donors (20.25%) were deferred during the COVID-19 pandemic. 136 (35.60%) were attributed to non-infectious factors such as tattoos, alcohol intoxication, history of vaccination, dental extraction, underweight, last blood donation within 3 months, underage, and lack of sleep. 130 donors (34.03%) were deferred due to anemia, while 64 donors (16.75%) faced deferral owing to various medical reasons, including a history of medications, increased blood pressure, active lesions, and fungal infections. A total of 41 donors tested positive for transfusion-transmissible infections (TTIs) out of 1504 donations (2.72%) during the same period.

Summary / Conclusions: The Medicine department demonstrated the highest number of crossmatches and issuances, underscoring its critical role in the blood supply chain. Interestingly, the Crossmatch/Transfusion ratio reached its peak in the Obstetrics and Gynecology (OBGY) department, highlighting the unique demands and challenges within this specialty. Notably, the COVID-19 pandemic did not significantly impede voluntary blood donations, as evidenced by 229 voluntary donations during April to December 2020, even without dedicated camps. In comparison, 261 voluntary donations occurred in the non-COVID period from April to December 2019, showcasing the resilience of our voluntary donor program despite the challenges of the pandemic and associated lockdowns. However, a reduction of 503 replacement donations was observed during the same span in COVID and non-COVID times, attributed to multifactorial reasons including lockdowns and the prioritization of COVID-positive cases in hospitals. This suggests that while voluntary donations remained robust, challenges persisted in replacement donations due to external factors associated with the pandemic.

P852 | Hypersensitivity reaction to ferric carboxymaltose in refractory iron deficiency anemia

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Background: Bleeding complications often cause iron deficiency anemia, the most common form of anemia. That may be treated by diagnosing and treating its etiological condition. Nevertheless, it's frequently necessary to prescribe iron supplements in order to improve symptoms and avoid transfusion.

Aims: To report a clinical case with challenging management of iron deficiency anemia.

Methods: 36-year-old female patient, with previous history of laparoscopic myomectomy, followed by gynecology due to anomalous uterine bleeding. As a consequence, she developed symptomatic iron

deficiency anemia (hemoglobin 10 g/dL, ferritin 4 ng/mL) and was referred to hematology consultation. The patient's study didn't reveal gynecological, gastrointestinal or thyroid abnormalities. Continuous oral contraceptive and oral iron supplementation was initiated. After 3 months, the patient still reported tiredness and lack of energy, although she had fewer uterine bleeding events. The analytic study showed no improvement as she maintained iron deficiency anemia. It was then proposed intravenous iron (IVI) therapy. Considering the patient's age, working status, the pandemic context, the hospital distance to the patient's home and the fewer doses of ferric carboxymaltose that would may be needed to replenish iron stores compared with other formulations, this was the preferred regimen.

Results: Shortly after the end of treatment, the patient developed a generalized rash and pruritus, without dyspnea or hemodynamic instability. Clemastine and hydrocortisone were immediately administered, with improvement of these symptoms. One month later, in the follow up appointment, the patient showed significant improvement both clinical and analytical, with hemoglobin 13.4 g/dL and ferritin 94 ng/mL.

Summary / Conclusions: Patient Blood Management (PBM) is a multidisciplinary approach that optimizes own red cell mass and patient-specific anemia reserve, minimizing blood loss, as well as reducing transfusion need and all the risks associated. For this purpose, IVI is a safe, well-tolerated, and effective strategy. Despite the low risk of hypersensitivity reactions, healthcare providers should be aware of these side effect when defining a therapeutic plan.

P853 | Haemoglobin and hematocrit levels before and after cardiac surgery in relation to administered erythrocyte transfusions

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Background: Support, as well as the availability of transfusion, plays a very important role in cardiac surgery. Of essential importance is the conditioning of the patient preoperatively, coordinate antiplatelet and anticoagulant therapy, compliance with the institutional MSBOS and an adequate strategy for the use of blood, due to the specificity of the course of the operation - the duration, the destruction of cellular elements of the blood when passes through the cardiopulmonary bypass.

Aims: To demonstrate the importance of adequate preparation of cardiac surgery patients as part of blood management at the Transfusion Department of the General Hospital.

Methods: A retrospective analysis of the medical records of patients undergoing cardiac surgery in 2023 at the Heart Center who received blood units was performed. We analyzed number of units according to hemoglobin level. The criteria for the inclusion of 92 patients were: elective myocardial revascularization operations, heart valve operations and combine, the outcome of the operation and applied transfusions of concentrated erythrocytes also. Patients with other types of

surgery, patients whose outcome was fatal, and patients who did not receive blood were excluded.

Results: Of the 220 operated patients at the Heart Center in 2023, 210 underwent coronary bypass surgery, valve surgery or complex surgery of both. Of 92 elective cardiac surgery patients, 44 men and 48 women, median age 66.5, 50% of patients received one unit of erythrocytes, 25% two, 10% three, and 15% more than three units erythrocytes. Preoperative hemoglobin and hematocrit values in 74 patients were within the reference range, borderline in 11 patients, and 7 patients had mild anemia. The target postoperative value of hemoglobin was 90 g/l. The largest number of transfusions was administered to patients undergoing valve surgery - 8, of which three were complex. Three patients with borderline preoperative hemoglobin values (2) and anemia (1) received more than three units of erythrocytes. The total number of erythrocyte units administered for 92 patients was 208.

Summary / Conclusions: The number of applied erythrocyte units according to the number of surgical procedures in 2023 in the Cardiac Surgery Center was 1 per operation, or 2.2 of transfused units for 92 blood recipient patients. Due to the necessity and importance of erythrocyte transfusion for tissue revascularization during cardiac surgery, the results indicate rational blood consumption. It is necessary to optimize hemoglobin values preoperatively in elective cardiosurgery.

P854 | Decreased fibrinogen levels in a patient with liver impairment—case study

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Background: Low fibrinogen is a strong risk factor for bleeding in a patients that needed to underwent surgical intervention. Any liver damage can be a sign of hypofibrinogenemia

Aims: Of this study is to show a correlation between low fibrinogen levels, and the bleeding tendency in a patient in the pre-operative period.

Methods: The haemostatic parameters, plasma fibrinogen levels and complete blood count were measured at the laboratory in the Department for haemostatic and thrombotic disorders at the Institute for Transfusion Medicine of Republic North Macedonia-Skopje.

Results: We present a 73 old patient, with chronic medical issues such as hyperlipidemia, high blood pressure, that needed to underwent surgical intervention for benign prostate hyperplasia. Routine pre-operative haemostatic analyses showed prolonged thrombin time (33 s), and correction studies showed (thrombin time 26 s). Later plasma fibrinogen levels showed decreased value (1.7 g/L). Later, substitution with 20 doses cryoprecipitate was conducted, and correction

of the plasma levels was noticed (2.6 g/L). There weren't noticed any bleeding signs during the whole pre and post-operative period.

Summary / Conclusions: preoperative low fibrinogen level may serve as a potential risk factor for postoperative bleeding. Therefore, diagnosis of any liver failure, and treatment is crucial in order to avoid any blood loss in those patients.

P855 | Sickle cell disease—the challenges of preventing immunohematological complications

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Background: Sickle cell disease is the most common genetic and hereditary disease in the world. Eastern Algeria is an endemic area and blood transfusion remains the major treatment for this disease.

Aims: To prevent anti-erythrocyte alloimmunization, which is the major obstacle to this therapy, to prevent its most serious consequence, post-transfusion hemolysis, and to improve transfusion yield in patients who are already immune.

Methods: A total of 65 sickle cell patients were included in this study, all of whom underwent ABO-Rh blood grouping, Rh-Kel phenotyping, an Irregular Agglutinins Test, and a Direct Antiglobulin Test in case of signs of hemolysis. Immunized patients or those with a history of post-transfusion hemolysis underwent extended phenotyping and RBCC selection based on Irregular Agglutinin Test result, patient's extended phenotype and compatibility with an Indirect Antiglobulin Test. The indication for transfusion was discussed on a case-by-case basis for patients at risk of severe post-transfusion hemolysis, and was only considered in cases of life-threatening emergency.

Results: 58.46% of our patients were homozygous sickle cell patients (SS), 11 sickle cell patients (16.92%) had a positive Irregular Agglutinin Test, 17 allo-antibodies could be identified, 04 patients were poly-immunized, and two with an anti-public allo-antibody, 06 patients benefited from an extended phenotype in order to select the most compatible RBCC. A database including the restricted and extended phenotype of donors enabled patients with the least frequent phenotypes to be managed in transfusion emergencies. By implementing this strategy, we were able to prevent the signs of hemolysis in these patients, with a good gain in terms of increased Hb levels, but also to unblock the transfusion deadlock situation for some of them.

Summary / Conclusions: Improved immuno-hematological characterization of the donor/recipient pair allows good management of complications linked to alloimmunization

Clinical transfusion—clinical / laboratory interface—TP initiatives

P856 | Abstract withdrawn

P857 | 2023 annual activities of the Korean Blood Safety Project—enhancing Transfusion Management Office activities

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Background: In accordance with the Blood Management Act, Korean medical institutions of a specified size are mandated to establish and operate a Blood Transfusion Management Office (TMO) with designated dedicated personnel, starting in 2021. The dearth of both domestic and international data on TMO installation and operation has posed practical challenges for medical institutions during the mandatory implementation. The Korean Blood Safety Project (KBSP), supported by Human Blood Safety Surveillance (HBSS), has been an annual initiative since 2012, dedicated to contributing to domestic blood and blood transfusion management.

Aims: This study aims to provide an overview of KBSP's activities in 2023, focusing on TMOs, to facilitate the establishment and operation of TMOs across Korea.

Methods: This initiative involved a range of activities such as face-to-face or online meetings, surveys, and satisfaction surveys conducted monthly or bimonthly through the TMO Network. Training sessions for Transfusion Practitioners (TPs) were provided, and counseling for inquiries was facilitated through the KBSP website's bulletin board. These activities were open to everyone including TPs, except for a face-to-face workshop for TPs.

Results: The TMO Network was established at the project's inception, hosting six meetings from May to November 2023, with a total attendance of 522 individuals. Meetings covered various TMO-related topics, including announcements, hospital status updates, the results of surveys, and Q&A sessions. Satisfaction survey results indicated high overall satisfaction, with an average score exceeding 4.4 out of 5 for content and over 4.5 for time and format, excluding the first meeting. Results from four surveys on specific topics were shared at the meetings. Additionally, 35 online Q&A counseling cases were addressed within 7 days. The face-to-face workshop for TPs attracted 185 participants, while a non-real-time online training, featuring six 30-minute videos developed in 2023, involved 1452 participants.

Summary / Conclusions: The KBSP's 2023 activities, encompassing the TMO Network, counseling projects, and training programs for TPs, were successful. They significantly contributed to the safe and efficient practice of blood transfusion, offering practical assistance for TMO operations in medical institutions across Korea.

P858 | Abstract withdrawn

P859 | Rhesus and Kell phenotyping implementation strategy in female pediatric patients at a public hospital in Ecuador—medical and economic impact in a resource constrained country

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Background: The Baca Ortiz Pediatric Hospital (HPBO) in Ecuador is renowned for its specialized care for the pediatric population aged 0 to 15 years. The Transfusion Medicine Service (TMS) began its operations in May 2011. Since 2013, the TMS has implemented a proactive strategy to ensure the availability of Rh and Kell phenotype-compatible blood for patients with hematological, oncological, renal diseases, whose medical condition anticipates prolonged transfusion requirements. In this context, the phenotyping of Rh and Kell antigens, in tube technique was replaced with column microagglutination ("card") method in August 2021 for all female patients, and is seen as an innovative and essential transfusion strategy. Its primary objective is to prevent sensitization to irregular antibodies in the short term and to prevent Hemolytic Disease of the Newborn (HDN) in the long term.

Aims: Actively promote Rh and Kell phenotyping among the pediatric female population attended at the HPBO, highlighting its importance in preventing HDN. In the Rh system the most relevant antigens were tested (D, C, c, E, e) as well as in the Kell system (K:1). Demonstrate the feasibility and effectiveness of these preventive practices in a resource-limited environment, providing a case study for replication in other hospital centers in the country.

Methods: A market analysis was conducted to identify the methodologies available in Ecuador, comparing tube techniques and column microagglutination techniques, considering their cost, efficacy, and feasibility of implementation. The column microagglutination card was chosen for its efficiency and adaptability. This methodology was integrated into the annual purchasing plan, with technical reviews and training to ensure proper implementation and standardization of the process. Initially, the investment in liquid reagents, costing approximately 5.50 USD per patient, raised questions about efficiency. The transition to phenotyping cards, at a cost of 9 USD per unit, was found fully justified considering their application in female patients, combined with superior sensitivity and specificity of the technique. Furthermore, the increase in the volume of tests performed underlines the effectiveness of this

transition, ensuring a valuable investment in diagnostic precision and patient safety.

Results: The implementation of Rh and Kell phenotyping using the card microagglutination technique at the Baca Ortiz Pediatric Hospital (HPBO) has shown significant results. Since its initiation in August 2021, there has been a decrease in the incidence of sensitization to irregular antibodies among the attending pediatric female population, contributing to the effective long-term prevention of HDN.

Summary / Conclusions: The systematic Rh and Kell phenotyping in female pediatric patients has established a new standard of care at the HPBO, enhancing the safety and quality of pediatric transfusion care. The results not only support the viability of this strategy in a resource-limited context but also highlight its potential for replication in other hospital centers in the country.

P860 | Transfusion medicine—indispensable part of a comprehensive and customized treatment protocol

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Background: In today's world there has been increasing emphasis on customization, even in the medical field. Each patient has their own set of co-morbidities depending on which their treatment protocols are formulated. Transfusion Medicine (TM) too has a major role to play in such customization, as many patients differ in their transfusion requirements, sampling techniques & compatibility testing protocols. An example of this became evident when we encountered a patient having a rare blood group phenotype. A multi-disciplinary approach had to be adopted towards this case, in which the role of TM department proved quite significant & indispensable.

Aims: To meet the transfusion requirements of a rare blood group phenotype patient, posted for a complex cardiac surgery in a resource limited setting.

Methods: Advanced Immuno-hematological Work-up. Autologous Blood Collections. Implementing Patient Blood Management Strategies. Cooperation, Coordination & Team Work.

Results: A 50-year-old Indian origin male was referred to our institute for atrial myxoma excision & CABG. We received samples for blood group (BG), compatibility testing & antibody screening (AS). BG & AS were performed by the column agglutination technique (CAT). The BG was reported as AB Rh (D) Positive, but his AS was pan-positive. The patient did not have any history of any sensitizing event. Multiple units cross-matched for the patient; ranging from AB Rh (D) Positive to O Rh (D) Negative were found incompatible at AHG phase by both the CAT & conventional tube technique. Family relative's blood samples were also incompatible with the patient. Suspecting the presence of an allo-antibody against a high prevalence antigen, the sample was referred to IBGRL-NHS, UK for resolution. They confirmed the

patients phenotype to be the rare Gerbich Phenotype (Ge: -2, -3, 4) with the development of clinically significant Anti-Ge2 allo-antibody. Procuring blood units from the rare blood donor registry was not feasible in our case. The initial patient Hemoglobin (Hb) levels were 14.1 gm/dl. It was decided to perform pre-operative autologous collections and meticulously implement patient blood management (PBM) strategies to help resolve this case. Initiating the first pillar of PBM, hematinics were started and his hemoglobin levels were built up. Autologous blood collection was performed based on his body mass index & targeted hematocrit levels. Three pre-operative autologous deposits along with volume replacement were performed in a planned manner, under strict medical supervision. Measures to minimize blood loss such as using a cell saver, deploying meticulous hemostasis and surgical techniques were also used.

Summary / Conclusions: The atrial myxoma excision & CABG could be performed successfully using his autologous blood units. The patient recovered well and was discharged on 10th post operative day at an Hb of 12.1 gm/dl. His third month follow-up was performed recently in which he was noted to be recovering well. Reference laboratory support for antibody identification; performing autologous blood donation & implementing PBM strategies played a major role in obtaining a successful outcome of this rare case. Customized and comprehensive care helped in its timely resolution. This highlights the significance of team work & co-ordination between not only the clinicians and TM department but also the various Blood Banks/Laboratories nationally & internationally.

P861 | Association between standard laboratory haematological test results and Rotational Thromboelastometry (ROTEM) parameters in patients with liver disease at Colombo North Teaching Hospital, Sri Lanka

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Background: The patients with liver disease are said to be at a state of "rebalanced haemostasis" due to the abnormalities in all haemostatic pathways. The standard laboratory haematological test results (SLHTs) often show that these patients are at an increased risk of bleeding whereas ROTEM parameters more closely show the situation in-vivo.

Aims: The main objective of this study was to determine the relationship between SLHT results and ROTEM parameters in patients with liver disease which has not been previously studied in this population.

Methods: In this retrospective patient record review study, the data from all the patients with liver disease at Colombo North Teaching Hospital, Sri Lanka during the study period were collected from the patient records, ROTEM requests and ROTEM data base. The strength and direction of association between SLHT results and ROTEM parameters were analyzed using Pearson's correlation

coefficient (r). These values were also compared for the whole study population and according to their bleeding status separately.

Results: Total of 257 patients' data were analyzed in this study. Out of those only 27.2% had bleeding whereas 72.8% did not have bleeding manifestations. It was found out that both SLHT results and ROTEM parameters are deranged in this study population, both in bleeding as well as non-bleeding patients. There was a statistically significant high degree positive correlations between platelet count and EXTEM A10 ($r = 0.568$, $p < 0.05$) and platelet count and EXTEM MCF ($r = 0.551$, $p < 0.05$), while prothrombin time (PT) and EXTEM CT values had a low degree positive correlation ($r = 0.254$, $p < 0.05$) which was also statistically significant. Both bleeding and non-bleeding patient groups had low degree positive correlations between PT and EXTEM CT. A better correlation between platelet count and EXTEM A10 was seen in patients with bleeding ($r = 0.649$, $p < 0.05$) when compared to non-bleeding patients ($r = 0.540$, $p < 0.05$). There were significant positive high degree correlations between platelet count and EXTEM MCF in both bleeding ($r = 0.630$, $p < 0.05$) and non-bleeding ($r = 0.523$, $p < 0.05$) patients.

Summary / Conclusions: Patients with liver disease show haemostatic abnormalities, which are evident in both standard laboratory haematological test (SLHT) results and ROTEM parameters. But most of these patients do not present with bleeding manifestations. According to our findings, platelet count can be used as a good predictor of clot strength even in the absence of ROTEM results in patients with liver disease. But EXTEM CT of ROTEM cannot be used in place of Prothrombin time (PT) in patients with liver disease and vice versa, as there is a poor correlation between the two parameters.

P862 | A case of post renal transplant erythrocytosis successfully treated with venesection

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Background: Post renal transplant erythrocytosis is defined as hematocrit (Hct) more than 51% which lasts over a period of one month in a person who has undergone renal transplantation. Typically, this condition develops within the first year following transplantation, though it can occur several years after. Spontaneous resolution can occur over a period of 1–4 years and treatment with angiotensin converting enzyme inhibitors (ACEi) or angiotensin receptor blockers (ARB) successfully reduces the Hct. Venesection has been used to treat patients with higher Hct (>0.57 – 0.60) but this is rarely required. Herewith we describe a case of post renal transplant erythrocytosis successfully treated with venesection.

Aims: Aim of this case report is to describe a case of post renal transplant erythrocytosis successfully treated with venesection.

Methods: Data for the case report was obtained prospectively from October 2023 to January 2024 using the clinical parameters and full blood count reports of the patient.

Results: A 37-year-old male with end stage renal disease preceding hypertension for 2 years underwent a live donor renal transplant in June 2023. Post renal transplant he was commenced on tacrolimus, mycophenolate mofetil, prednisolone and other supplements. Post transplant his hemoglobin (Hb) and hematocrit gradually increased from 12.2 g/dL (Hct 36.6%) to 18.8 g/dL (Hct 57%) by end of 3 months post transplantation. Thus, he was commenced on enalapril 2.5 mg nocte and deryphillin 150 mg bd. At the time of initiation of above medication, patient's serum creatinine was 1 mg/dL. Despite treatment patient's Hb gradually increased up to 19.5 g/dL with Hct remaining at 57% with rising serum creatinine of 1.2 mg/dL. Due to the possibility of medication related alterations in renal functions, enalapril was withheld and the patient was referred for venesection. Patient had symptoms of hyper viscosity at the time of admission. Two venesections were performed in two consecutive days with removal of 10% of blood volume at each occasion. He was reviewed after two days and the Hb and Hct was 17.2 g/dL and 51.4% respectively. Patient was discharged with a plan to review in one month with advice to admit immediately if hyper viscosity symptoms occur. One month post venesection patient presented with a Hb of 18.13 g/dL and Hct of 54.4% and patient underwent two consecutive venesections one day apart. Patient's hematological parameters were assessed 2 days after and it revealed a Hb of 14.8 g/dL with a Hct of 45%. Post procedure patient was discharged and was registered for regular follow-up.

Summary / Conclusions: Venesection can be recommended as a treatment modality for patients with post renal transplant erythrocytosis where medical treatment is contraindicated.

P863 | The clinical spectrum of ABO hemolytic disease of fetus and newborn in neonates born to group O mothers

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Background: Haemolytic disease of the fetus and newborn (HDFN) is the destruction of the red blood cells (RBCs) of a fetus and neonate by antibodies produced by the mother. The mother can be stimulated to form RBC antibodies naturally (ABO), by previous pregnancy, or transfusion (RBC alloimmunization). ABO incompatibility occurs in 15%–25% of pregnancies and produces a spectrum of haemolytic disease. ABO HDFN is currently the most common cause of neonatal jaundice attributed to maternal–infant blood incompatibility. Early diagnosis and adequate care is necessary to prevent complications.

Aims: To find the clinical spectrum of ABO HDFN.

Methods: Babies with blood group A, B, AB with positive DAT who were born to O blood group mother with antibody screening negative,

were included in this study. Titres of anti A and anti B were done in mothers. Onset and progression of jaundice were monitored subsequently by visual assessment and periodic serum bilirubin levels. Babies were managed either with Phototherapy, IVlg or exchange transfusions.

Results: Thirteen out of fifteen cases were treated with Phototherapy. Only two out 15 cases required IVlg. None of the cases required Exchange transfusion. Thereby, implying that most of the cases of ABO HDFN are self limiting and can be treated with phototherapy alone. The anti-A and anti-B titres were estimated. Titres above 64 were responsible for increased chances of jaundice and raised bilirubin levels.

Summary / Conclusions: Most of the ABO HDFN cases are mild to moderate which could be self limiting and require only phototherapy.

Cellular therapies—stem cell and tissue banking, including cord blood

P864 | Evaluation of apheresis quality and donor safety in a centralized model for peripheral blood stem cell collection and processing within Catalan donor center linked to Spanish Donor registry (REDMO)

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Background: Preserving donor's safety and guaranteeing donation quality are the most important aims for a donor registry. Centralizing activities would increase donation centres efficiency.

Aims: We reviewed collection activity, safety profile and quality data of a centralized model in Catalonia, where a single collection centre evaluates donors and performs peripheral blood stem cell collections (Hospital Sant Pau Blood and Tissue Bank) of the voluntary donors from Spanish Donor registry (REDMO), that are then transferred to the reference cell processing centre (at BST hub) for cellular count, packaging and delivery. Analysis was performed from 01/01/2015 to 31/12/2023.

Methods: Selected donors are contacted by our management team for verifying their willingness to continue and scheduling pre-collection visit. Blood tests, echocardiogram and x-ray are performed the same day as the medical visit (over three hours, trying to minimize donor's time investment). Within 72 h, results are available and donor's clearance can be released for REDMO's approval. Apheresis were performed with Cobe Spectra® (until 2017) and Spectra Optia® (since 2018), using ACD-A as anticoagulant.

Results: During the study period an increasing demand was observed attributed to improvements in time from work-up to delivery, HLA typing, younger donors and high quality collection products. Effective donors are still increasing. 355 apheresis were analysed. Adverse events (AE) were notified in 195 procedures (55%) and registered as mild AE notifications, except one severe AE (a donor developed splenomegaly with potential splenic fissure that resolved with analgesia (out-patient treatment)).

These notifications included 244 AE: paresthesias ($n = 122$), myalgia/arthralgia ($n = 86$), headache ($n = 25$), sickness ($n = 4$), hypertension ($n = 3$), bruising ($n = 3$) and transaminitis ($n = 1$). Peripheral venous access (18G vs 16 G Abbocath) was preferentially used for apheresis. 4 donors (1.5%) required a central venous access (subclavian vein). During the immediate follow-up (1-week after donation), non adverse effects have been reported. Donor and cellular product characteristics assessed are shown below:

P864 - Table 1.

Year	Total Apheresis Performed	Apheresis with Mild Adverse Events
2023	92	66
2022	57	28
2021	50	33
2020	47	18
2019	32	17
2018	39	15
2017	18	8
2016	14	6
2015	6	4
Total	355	195 (55%)

P864 - Table 2.

	Median	Min	Max
Donor Age	32	19	63
CD34/ml (E3)	99.3	11.8	338
CD34/kg (E6)	7.2	1.43	44.21
%MNC	85.7	46.3	99.1
CN/ml (E6)	163.2	65.6	673
%CD34 viability	99.9	84.8	100
%CD34 collection efficiency	50.6	30.1	206.5
CFU GM/kg (E5)	9.7	0.5	68.8
CFU CD34/CFU Total (% clone)	32.5	9.31	82.8

Products cell concentration and CD34+ cell viability (7AAD) after collection fulfil quality criteria (<200 E6 CN/ml. and $\geq 95\%$, respectively). Quality indicators median for % mononuclear cells (MNC), CD34/kg, collection efficiency and clonogenic efficiency (CFUs) were over our minimal reference values ($\geq 70\%$, $\geq 4E6$, $\geq 40\%$, $\geq 1E5$ and $\geq 10\%$, respectively).

Non mobilization nor graft failure has been notified in this series.

Summary / Conclusions: We present a model in which resources are centralized (comprising facility, equipment, team and processing unit). This has led to an achievement of expertise throughout the whole donation process, improving donor's profile and reassuring indeed donor's and patient's safety by minimizing deviations or incidences. It also has permitted its adaptation to the increasing demand for REDMO donors over the years, and constitutes a robust collection of effective donations and processing procedures of products from volunteer stem cell donors.

P865 | Abstract withdrawn

P866 | Abstract withdrawn

P867 | Differential Expression of miRNA associated with fetal haemoglobin expression among cord blood exosomes and maternal control

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Background: Cord blood contains an array of miRNA regulating gamma globulin expression. However, presence of these miRNA in exosomes derived from cord blood has not been reported yet. This study aims to evaluate the cord blood exosomes for miRNA regulating HBF expression.

Aims: This study primarily aims to explore and analyse the differential expression of miRNA regulating fetal haemoglobin expression in cord blood exosomes and exosomes from the corresponding mother sample.

Methods: A total of 20 Cord blood EDTA samples from full term live birth deliveries and corresponding mother EDTA blood samples were collected. The exosome was isolated from the plasma sample using the commercially available exosome isolation kit (Qiagen, Valencia, CA). The miRNA was extracted from the exosomes and using the miRNA easy mini kit (Qiagen, Valencia, CA) with the exosome using exosomes. The extracted miRNA was quantified and validated with the qRT-PCR using SYBER green miRNA qRT-PCR kit (Qiagen). The target miRNA expression level between two sample will be determined by $2\Delta Ct$, with $\Delta Ct = (\text{internal control Ct} - \text{target miRNA Ct})$ method. The result of the test was analysed and validated applying the bioinformatics tool.

Statistical Analysis: The results will be presented as fold change expression of miRNA from plasma sample. Descriptive statistics as median(interquartile range) will be used to represent other variables. The Wilcoxon rank sum test was used to analyse the differential expression of miRNA between the cord blood exosomes and maternal

exosomes. A value of $p < 0.05$ was accepted as the level of significance.

Results: The cord blood exosomes shown a significantly increased expression of miRNA 15a-5p, miRNA 381-3p, miRNA 210-3p, miRNA 326, and miRNA 103a-3p in cord blood exosomes versus mother sample exosomes with a ΔCt median (interquartile range), p -value of 2.57 (2.29-2.97) versus 3.57 (2.95-4.16), p -value (0.006); 8.91 (8.10-9.54) versus 12.16 (10.45-14.17), p -value (0.000), 4.56 (3.31-5.28) versus 7.53 (7.00-8.15), p -value (0.000), 7.00 (5.37-7.46), versus 8.86 (7.68-9.64), p -value (0.002), and 3.61 (2.98-4.00) versus 4.21 (3.50-5.06), p -value of (0.016), respectively. The differential miRNA expression was not different statistically in cord blood exosomes compared to mother sample exosomes for miRNA 486-3p, miRNA23a-3p, miRNA 27a-5p, miRNA 96-5p, miRNA 34a-5p, miRNA 23b-3p, miRNA let 7a-5p. The target gene were analysed with the bioinformatics tool and the miRNA showing significantly increased expression responsible for increased HBF level was associated with BCL11A, MYB, KLF-1, FOG-1, HIF1A, TAL1, and SOX6 gene.

Summary / Conclusions: The study find the various miRNA in cord blood exosomes responsible to increase the HBF level in cord blood. Further, the gene target analysed with the bioinformatics tool validated the result of the study. However, the sample size of this study is low and future study should be planned for confirmation of the presence of these miRNA in large sample and to exclude the racial differences.

P868 | Evaluating the effectiveness of stem cell recruiting policies using a dynamic registry simulation

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Background: When a stem cell registry's size is constrained by limits on recruitment effort, as it is in Canada, identifying the right person to recruit is a critical determinant of effectiveness. Since the distribution of HLA profiles varies by ethnicity, strategies must allocate recruiting effort between different ethnic groups to balance the genetic diversity of the registry while recruiting donors that will most likely match a person requiring a transplant.

Aims: In this study, we evaluate the impact of changes to donor recruitment effort within specific ethnic communities on the number of Canadian-to-Canadian matched made by the Canadian Blood Services Stem Cell Registry (CBSSCR) over a simulated ten-year time horizon.

Methods: Simulation methods are applied to create a cohort of donor recruits, over a ten-year time horizon, for the CBSSCR. Starting in 2021, 25,000 potential donors are recruited annually, of which about 16,000 become new recruits and are added to the registry. New registrants for the CBSSCR are recruited only between the ages of 18 and 35. Registrants age out at their 61st birthday. In a similar fashion, simulated patient cohorts of between

1144 and 1946 individuals are created over the time horizon. At the end of each simulated year, simulated patients are matched against the simulated registry. Experiments tested the impact of recruiting effort within ethnic groups on the number of matches expected. In the first set of runs, it is assumed that patient searches follow the historical patient search distribution for Canadian patients (85% Caucasian). Donor recruiting effort within ethnic groups is tested at two levels: R1 and R2. R1 has an ethnic distribution to the ethnic distribution of the Canadian population in 2021 (73% Caucasian) and R2 has an ethnic distribution within non-Caucasian groups twice that of the Canadian population in 2021 (63% Caucasian). In the second set of runs, the impact of a more diverse patient search population (73% Caucasian) is tested against the same two donor recruitment strategies.

Results: Statistically significant increases in the number of registrants of Black, Hispanic, and First Nations descent can be expected when non-Caucasian populations are recruited at rate R2. However, there are larger decreases in the number of registrants from the Asian-Pacific, Caucasian, and Other communities. Additionally, ethnic communities that have limited registrants in the CBSSCR in 2021 (Black, Hispanic) do not benefit from increased recruiting efforts. This result holds if patient searches match the historical ethnic distribution of patient searches (C1) or the ethnic distribution of the Canadian census (C2).

Summary / Conclusions: Preferentially recruiting from non-Caucasian populations (R2) will reduce the number of Canadian-to-Canadian matches, since increases in non-Caucasian populations will not fully counterbalance decreases to Caucasian matches. Nevertheless, since less than 30% of stem cell transplants for Canadians come from Canadian sources, most Canadian patients currently rely on international sources for transplant. Thus, given the size of the Canadian registry, international registries will remain an important source for matches, regardless of the recruiting strategies for the CBSSCR.

P869 | Analysis of cord blood units provided for transplantation in Korea over a 10-year period

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Background: The use of cord blood as a source of hematopoietic stem cells has been declining since haploidentical hematopoietic stem cell transplantation became available. In Korea, the Act on Cord Blood Management and Research came into effect in July 2011. Starting in

2021, cord blood with a total nucleated cell (TNC) count of 11×10^8 or more before processing is required to be stored. In addition, since 2011, government-assigned public cord blood banks have been operating to support the storage and use of high-quality cord blood.

Aims: To evaluate the characteristics of cord blood used for transplantation to help improve the effectiveness of cord blood storage and utilization.

Methods: Data on the annual storage volume of cord blood registered for transplantation were obtained from the Cord Blood Information Center of the National Institute of Organ and Tissue Blood Management of the Ministry of Health and Welfare. Information was received on cord blood supplied for transplantation over a 10-year period from 2013 to 2022. The information included TNC, CD34+ cells, age, weight, and disease of the cord blood recipients, the length of time the cord blood was frozen, and the number of cord blood transplants. The information provided was analyzed.

Results: As of December 31, 2022, 42,728 units of cord blood with a TNC count of 7×10^8 or more were stored for transplantation. During the 10-year period, 825 units of cord blood were supplied for transplantation, of which 720 (87.3%) were from nationally designated public cord blood banks. The mean TNC count was $14.7 \pm 4.5 \times 10^8$ and the mean CD34+ cell count was $5.7 \pm 3.9 \times 10^6$. Eighty-two units (9.9%) of cord blood stored for more than 10 years were used. There were 452 patients who received a transplant, with acute lymphoblastic leukemia accounting for 231 (51.1%). Patients aged 19 years or older accounted for 299 (66.2%). Most patients received two units of cord blood (349/452, 77.2%), and their average weight was 61 kg. Nine patients (2.0%) received additional units of cord blood after their initial cord blood transplant.

Summary / Conclusions: The use of donated cord blood funded by the government has been high, and it has become a good source of supply for patients who have difficulty obtaining hematopoietic stem cells from allogeneic donors or family members. Two-unit transplants and transplants to adults are common, and cord blood with a high TNC is utilized, requiring continuous storage of high-quality cord blood.

P870 | Feasibility study of constructing high-matching iPSC haplobank using the HLA data from cord blood bank

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Background: Human induced pluripotent stem cells (iPSCs) can differentiate into any types of cells and have the potential to apply regenerative medicine to the treatment of many diseases. In allotransplantation, reducing immune rejection is a key problem can be solved by two immune-compatible iPSC therapeutic pathways: the

use of genetic modifications to produce "universal donors" that reduce the expression of human leukocyte antigen (HLA) and other immune targets, or the development of haplobank containing specially selected iPSC lines to provide HLA matching products to large portions of the population.

Aims: To study the feasibility of constructing highly matched induced pluripotent stem cell haplobank using HLA data from umbilical cord blood bank.

Methods: 5421 cord blood samples (CBs) were randomly collected from the Zhejiang Cord Blood Bank, China. (This study was approved by the Ethical Review Committee of Zhejiang Blood Center). HLA-A, -B, -C, -DRB1, -DQB1 loci was genotyped using next generation sequencing method. The genotypes of the samples were assigned using the HLA TypeStream Visual Software version 2.0. The frequency of alleles, haplotype estimation and linkage disequilibrium analysis were performed with the Arlequin software 3.5.2.2. The matching probability was calculated with homozygous donors as assumed iPS donors and 5421 random donors as assumed patients. If the assumed patients' phenotype contained all the alleles present in the iPS cell's phenotype, this was considered a match.

Results: 52 HLA-A, 109 HLA-B, 52 HLA-C, 63 HLA-DRB1, 23 HLA-DQB1 alleles were identified from 5421 CBs. The top three frequent alleles of HLA-A, -B, -C, -DRB1, -DQB1 loci were A*11:01(25.83%), A*24:02(16.66%), A*02:01(10.87%); B*40:01(15.97%), B*46:01(12.02%), B*58:01(7.35%); C*07:02(20.05%), C*01:02(18.14%), C*03:04(10.43%); DRB1*09:01 (17.34%), DRB1*12:02 (9.70%), DRB1*15:01(9.65%); DQB1*03:01 (22.36%), DQB1*03:03 (18.31%), DQB1*06:01 (11.94%). A total of 3326 distinct HLA-A, -B, -C, -DRB1, -DQB1 haplotypes were estimated. Only 8 haplotypes with an haplotype frequencies (HFs) of more than 1.0% were as follows: HLA-A*02:07-B*46:01-C*01:02-DRB1*09:01-DQB1*03:03(3.95%), HLA-A*33:03-B*58:01-C*03:02-DRB1*03:01-DQB1*02:01(3.40%), HLA-A*30:01-B*13:02-C*06:02-DRB1*07:01-DQB1*02:02(2.95%), HLA-A*11:01-B*15:02-C*08:01-DRB1*12:02-DQB1*03:01(1.92%), HLA-A*33:03-B*58:01-C*03:02-DRB1*13:02-DQB1*06:09(1.32%), HLA-A*11:01-B*40:01-C*07:02-DRB1*08:03-DQB1*06:01(1.12%), HLA-A*11:01-B*40:01-C*07:02-DRB1*09:01-DQB1*03:03(1.08%) and HLA-A*02:07-B*46:01-C*01:02-DRB1*08:03-DQB1*06:01(1.02%). A total of 11 homozygous HLA-A, -B, -C, -DRB1, -DQB1 haplotypes that were observed among the stored CB units would cumulatively match 32.99% of the 5421 potential patients as zero HLA mismatch iPS donors.

Summary / Conclusions: High-matching iPS haplobank generated from CB units may be an economical and effective option in the allogeneic model of iPS therapy. Using high resolution typing data for at least HLA-A, B, C, DRB1 and DQB1 across different populations could provide starting material for iPS haplobank and have clinical consequence. In the expanded analysis, we expect a more sophisticated assessment and elucidation of the list of HLA-homozygous haplotypes covering almost the entire China population.

P871 | First year evaluation of the Ready to Ship (R2S) project in umbilical cord blood units from the Spanish registry

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Background: The R2S ("ready to ship") project was initiated by all five Cord Blood Banks (CBB) of Spain in 2023, in order to optimize the process of searching cord blood units (CBU), reduce the shipping time for transplant, improve the quality of CBU and increase the number of shipments. CBU R2S is a unit considered suitable to be sent and used for transplant immediately because all required information and quality control test are available. A working group of members from all CBB of Spain agreed on selection criteria for CBU to become R2S, based on cellularity and ethnic characteristics. 4920 UCB with high cellular quality were identified to be performed in 2 phases. Phase 1: 2412 CBU (TNC $\geq 180 \times 10^6$ and CD34 $\geq 60 \times 10^5$), Phase 2: 2271 CBU (TNC $\geq 150 \times 10^6$ and CD34 $\geq 60 \times 10^5$) and 237 CBU for ethnic minorities (TNC $\geq 120 \times 10^6$ and CD34 $\geq 40 \times 10^5$). Once every CBU have all requirements completed, these will be flagged as "ready to ship" in the Spanish registry (REDMO) and hopefully in WMDA.

Aims: The aim of the present work is to analyse the first year of activity of the "Ready to Ship" Spanish Project, describe the final use of R2S CBU and establish causes of units pre-selected as potential R2S but not finally selected as suitable.

Methods: The requirements to flag a CBU R2S:

- Records: Sex of the newborn. Processing method. Number and type of cryopreserved bags, type of cryoprotectant and additive solutions used. Microbiological cultures. Hemoglobinopathies screening. CCR5.
- Test prior to cryopreservation: Quantification of Total Nuclear Cells (CD45+) and CD34+. Cell viability (CD34+, CD45+).
- Test in segment or representative cryovial: Confirmatory HLA typing at least for loci A, B, C, DRB1, DQB1, DPB1. Cell numbers and viability (CD34+, CD45+) and clonogenic cultures (CFU).
- IDM: Infectious disease detection tests performed on mother and CBU.

The major reasons why the units were discarded and did not become R2S were:

- Quality control (QC): lack of reference sample (segment, cryovial), recovery or viability of CNT and/or CD34+ cells or sub-optimal clonogenic growth, HLA discrepancy, integrity and traceability of the bag.
- Storage issues.
- Positive infectious markers.
- Findings in family medical history.
- Lack of documentation.
- Used in transplant.
- Biobanking.
- Positive initial microbiological culture.

Results: The “R2S” project has been completed in 1088 CBU, 45.1% of the total units proposed for the first year. 12.6% (137 CBU) were not listed as “ready to ship” due to the following reasons: nine (0.8% of the total analyzed) shipped to transplant; seven (0.6%) released to biobank; 67 (6.2%) due to the lack of reference samples; 25 (2.3%) with suboptimal viability; six (0.6%) with absence or very low colony formation; five (0.5%) caused by suboptimal cellular recovery; seven (0.6%) CBU had positive infectious markers; six (0.6%) owing to lack of integrity or traceability of cord blood bags; and one (0.1%) with incomplete documentation.

Summary / Conclusions: 87.4% CBU of the initially proposed units have been identified as ready to ship despite of time of cryopreservation or differences in processing methods along years. The main causes of downgrade were the lack of a reference sample and the detection of suboptimal viability (< 70% in CD34 cells and < 45% in total nuclear cells) in quality controls. The speed of selection and shipment of these units should be evaluated, although preliminary data indicates greater utilization and faster shipment, compared to non-R2S units.

P872 | The impact of the cryopreserved period on the percentage of viable CD34+ cells among whole CD34+ cells of CB units

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Background: The CB banks have been checking the quality of the cryopreserved CB units through the pre-transplantation examination, including the total nucleated cells (TNC) and CD34+ cells, before releasing them for transplantation. The measurement of the TNC and CD34+ cells was conducted after

nonviable cells were excluded by incorporating the viability dye 7-aminoactinomycin D (7-AAD) in the flow cytometry procedure. To date, no assessment of the percentage change of viable CD34+ cells among CD34+ cells of cryopreserved CB units has been conducted.

Aims: We investigated the change in the percentage of viable CD34+ cells among whole CD34+ cells over the cryopreserved period. We also examined whether the percentage of viable CD34+ cells has been associated with the CD34+ cell count of CB units before cryopreservation.

Methods: The Seoul Metropolitan Government Public Cord Blood Bank (Allcord) was founded in May 2006, and the release of CB units for CB transplantation (CBT) started in July 2008. Among the CB units conducting the pre-transplantation examination until June 2023, 726 CB units are selected. Using the LMD files previously analyzed for the pre-transplantation examination by flow cytometry, we re-analyzed the percentages of viable cells among whole CD34+ cells. We conducted a correlation analysis between the cryopreservation period and the percentage of viable cells among whole CD34+ cells. After adjusting the cryopreservation period, we conducted the correlation analysis between the percentage of viable CD34+ cells and the CD34+ cell count before cryopreservation. One-way ANOVA and multiple comparison analysis were performed to investigate the differences in the viable CD34+ cells percentage according to the cryopreservation period group (up to 5 years, between 5 years to 10 years, over 10 years).

Results: The mean cryopreserved period of the 726 CB units was 2303 days (range, 376-5772). The percentage of viable CD34+ cells decreased according to the increase of the cryopreserved period ($r = -0.306$, $p < 0.001$). The percentage of viable CD34+ cells showed no correlation with the CD34+ cell count of CB units before cryopreservation when adjusted by the cryopreserved period ($r = -0.048$, $p = 0.242$). The overall percentage of viable cells among whole CD34+ cells was $91.2\% \pm 7.5\%$. The percentage of viable CD34+ cells was significantly different ($p < 0.001$) in the cryopreservation period groups: up to 5 years (298 units), $93.3\% \pm 7.2\%$; between 5 years to 10 years (317 units), $90.8\% \pm 6.2\%$; over 10 years (111 units), $87.0\% \pm 9.5\%$.

Summary / Conclusions: These results suggest no significant correlation between the viability of CD34+ cells and the absolute count of CD34+ cells before cryopreservation. The overall percentage of the viable cells among whole CD34+ cells was higher than that of the viable cells among whole TNC ($60.5\% \pm 12.6\%$, data not shown). Commonly, hematopoietic cells (HPC), namely CD34+ cells, are more resistant to cryopreservation damage than nucleated cells due to the difference in membrane lipid content and protein. However, the viable cell proportion of CD34+ cells of the cryopreserved CB units has been decreased according to the duration of cryopreservation. Therefore, the minimal number of CD34+ cells of CB units suitable for cryopreservation needs to be determined, considering the relatively low absolute count of CD34+ cells and the proportion of viable CD34+ cells.

P873 | Umbilical cord blood platelet gel for cutaneous lesions in a patient with severe graft-versus-host disease

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Background: Umbilical cord blood platelet concentrates (UCB-PC) has been shown to be a rich source of growth factors applicable in regenerative medicine (Samarkanova D, et al., Front Immunol. 2020). Its indication has been reported for the treatment of ocular surface disorders, skin wounds, and orthopaedics. The appearance of skin ulcers resulting from Graft-versus-Host Disease (GvHD) can become a complex challenge to solve with conventional treatments and cures. Faced with these difficulties, UCB-PC may be a good option in cutaneous healing and regeneration. UCB-PC derived gel (CBPG) is currently a treatment available under compassionate use via the authorization of the competent Spanish Agency (AEMPS), under product in investigation number PII:15-034.

Aims: To describe a clinical case where three lower limb skin lesions were treated with CBPG.

Methods: CBPG is a standardized drug containing a concentration of $800\text{--}1200 \times 10^9$ platelets/L in 10 ± 5 mL. Products were dispensed from the Cell Therapy Service of the Blood and Tissue Bank (BST) in batches frozen at -80°C . Additionally, vials of calcium gluconate were supplied to perform platelet activation for the gel formation on the day of treatment. A treatment schedule was agreed upon with the Clinical Unit of the patient. Treatment was performed once a week and for as long as required. On the treatment day, the gel was activated at the local BST and the Clinical Unit nurses applied it to the target lesion. In total, 29 gels were used to treat 3 wounds localized on the patient's legs.

Results: A 19-year-old male diagnosed with Acute Lymphoblastic Leukemia at the age of 15 years underwent allogeneic hematopoietic stem cell transplantation from an identical sibling in January 2020. He received a CD34+ boost due to poor graft function and afterward developed a severe chronic GvHD with cutaneous and osteoarticular involvement, which was refractory to conventional systemic and local treatments. Application of CBPG was initiated on a lesion of the outer face of the left lower limb. After 5 applications, the wound size was reduced from 15 to 3 mm. Treatment was continued onto 2 more lesions located on the anterior tibia of the left leg and the outer face of the right lower limb. In the first case, a total of 15 applications were performed, reducing the wound size from 48 to 3 mm in 14 weeks. In the second case, 9 applications were performed, resulting in a decrease from 38 to 6 mm in 5 weeks. No adverse effects were reported, related to treatment.

Summary / Conclusions: Topical administration of CBPG proves to be an effective treatment for complex ulcers in patients developing severe chronic cutaneous GvHD.

P874 | Delayed cord clamping is compatible with public cord blood banking—lets do the best of both worlds

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Background: Delayed umbilical cord clamping is the current gold standard for attending labour. There is controversy whether this practice affects the proportion of cord blood units (CBU) suitable for public cord blood banking (CBB).

Aims: This study was designed to assess the impact of delayed cord clamping practice on the number of cord blood units suitable for therapeutic uses.

Methods: To minimize variability, activity from the four most-active collection centers within the Programa Concordia, Banc de Sang i Teixits CBB in Spain was included. Data on CBU samples obtained after in-utero collections from mothers following normal vaginal deliveries during the period July 2018 to December 2021 were analyzed. Collection bag's weight (as a surrogate of volume) and total nucleated cell count were analysed according to three defined clamping times: (i) 30s, (ii) 60s and (iii) ≥ 120 s. To assess utilization rate, CBU were stratified on units suitable for stem cell transplantation (≥ 110 g and $\geq 1500 \times 10^6$ nucleated cells per unit) and for other clinical applications (≥ 100 g failing to meet TNC threshold).

Results: CBU distribution according to clamping times was 131 (18%), 548 (76%), and 40 (5%) at 30s, 60s and ≥ 120 s, respectively. Median weight gradually decreased according to time, with a significant difference between 30s and 60s ($p=0.036$) clamping. Consequently, CBU meeting minimal bag's weight criteria (100g) was significantly lower at 60s compared to 30s ($p=0.002$). However, this was not reflected on total nucleated cells available, resulting in non-statistical differences on CBU eligible for banking between those times. Actually, major predictor of the collection success was newborn's birth-weight.

Summary / Conclusions: In spite of having less volume collected, nucleated cell count is maintained when comparing 30s and 60s cord clamping resulting in similar numbers of CBU eligible for banking. Therefore, delayed cord clamping practice is compatible with public cord blood banking needs.

P875 | A population-based study of the prevalence of celiac disease among healthy donors of the hematopoietic stem cell registry

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Background: Celiac disease is a chronic, autoimmune disease in which the proximal part of the small intestine is affected. People who regularly consume products from cereals (wheat, rye, barley and oats) are genetically predisposed to this disease, which determines the prevalence rate of 1% and a strong variation by country and region. An important genetic factor in the development of celiac disease is the presence of HLA class II genes HLA-DQA1 and HLA-DQB1 and a combination of alleles in various variants that determine the risk of developing the disease. Thus, the detection of a combination of genes in the combination of HLA-DQ2.5 suggests a very high, and HLA-DQ2.2, on the contrary, a low predisposition to celiac disease. Given the historically developed diet of the population of Kazakhstan, which is dominated by gluten-containing foods, it became necessary to study genetic markers that carry a high risk of developing celiac disease and to make an objective assessment of the spread of these genes.

Aims: To study the distribution features of HLA haplotypes (HLA-DQ2, HLA-DQ8) associated with celiac disease in donors of the hematopoietic stem cell registry (HSC).

Methods: The studies were conducted on the basis of NGS typing data of 2755 potential donors of the HSC register for 2019-2022 in Scientific and Production Center of Transfusiology. HLA typing was performed at high resolution by the method of NGS HLA-DQB1 and DQA1 alleles. Sample preparation of samples was carried out using the Hologate HLA 24/7 kit (Omixon). High-performance sequencing was carried out on the MiSeq (Illumina) platform. The primary and secondary data analysis was carried out using the MiSeq Reporter Software (Illumina) and Twin (Omixon) software. The study considered eight main alleles as HLA markers: HLA DQ2. 5-cis (DQA 0501 / DQB 0201), HLA DQ2.5-trans (DQA 0505 / DQB 0301 + DQA 0201/DQB 0202), HLA DQ2.2 (DQA 0201/DQB 0202) and DQ8 (DQA 0301 /DQB 0302).

Results: The study of the prevalence of allele variants HLA-DQ2, HLA-DQ8 in potential donors of the HSH Registry showed that the proportion of carriers of the studied haplotypes was 5.4% (n = 148). The analysis of genetic markers revealed the predominance of combinations of DQ2.5-cis alleles (n = 102), of which heterozygous DQ2.5 (n = 13) and homozygous DQ2.5 (n = 18), DQ8 was detected in 45 donors, while heterozygous DQ8 (n = 7) and homozygous DQ8 (n = 18), the DQ2.2 alleles was detected in 1 donor (heterozygous DQ2.2 (n = 1) and homozygous DQ2.2 (n = 0)).

Summary / Conclusions: The study of HLA-haplotypes of celiac disease among adults in Kazakhstan showed a wide distribution of genetic markers (HLA-DQ2 and HLA-DQ8), which carry a high risk of developing this disease - 5.4%. At the same time, a combination of DQ2.5 alleles prevailed (n = 102). These studies are the beginning of more extensive research in this area.

Cellular Therapies—collection, processing, storage and release

P876 | Generating NKG2D-CAR-transduced natural killer (NK) cells for treating osteosarcoma

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Background: Osteosarcoma (OS) is the most common primary malignant bone tumor, with an incidence worldwide of approximately one to three cases annually per million. It typically affects children and young adults with a second smaller peak of incidence in those individuals over the age of 50. The current main treatment is based on the combination of surgery and neoadjuvant chemotherapy leading to a long-term survival rate of 70%. However, these strategies are inefficient for the metastatic or relapsed OS patients, in who the 5-year survival rate is less than 20%. As a result, in the last decades, alternative novel therapeutical approaches are being developed, including **chimeric antigen receptors (CARs)-based immunotherapy**, in which immune effector cells such as T cells are genetically engineered to express CARs in their surface membrane for improving the recognition and killing of tumor cells. **CAR-transduced T (CAR-T) cells** have revolutionized the cancer therapy, however toxicities associated to this autologous therapy such as cytokine release syndrome and immune effector cell-associated neurologic syndrome limit its wide-spread. Efforts have been made to find a more appropriate cell type for being genetically engineered with CARs. Recently, **natural killer (NK) cells** have stood out among other immune cell types as a potential allogenic alternative due to their cytolytic capacity and their safety profile. NKG2D receptor (NKG2DR) is one of the most important activating receptor expressed in NK cells and OS cells express ligands for NKG2DR. Therefore, the use of genetically engineered **natural killer (NK) cells** expressing **NKG2D-CAR** is proposed as a potential allogenic alternative to treat OS.

Aims: The aim of this study is the production of NKG2D-CAR-NK cells in order to target and kill more effectively tumor cells specifically OS cells

Methods: Study the expression of NKG2D-ligands (NKG2DL) in different cell lines and primary cells of OS by flow cytometry. Isolation and expansion of NK cells from umbilical cord blood and adult peripheral blood samples. Optimization of the transduction protocol using different enhancers. Analysis of the level of transduction of NK cells

with NKG2D-CAR by flow cytometry. Study the functionality of NKG2D-CAR-NK cells against recombinant NKG2DL and/or target cells expressing NKG2DL.

Results: Target cell lines expressed different levels of NKG2DL. The use of polybrene promotes a high level of transduction but decreases drastically the number of cells, while other enhancers increase the fold expansion of NK cells upon transduction with NKG2D-CAR. The level of expression of NKG2D in NKG2D-CAR-NK cells is higher than in MOCK-NK cells in both, AB-NK cells and CB-NK cells. NKG2D-CAR-NK cells present a higher degranulation activity than MOCK-NK cells when they are exposed to recombinant NKG2DL. The increased expression of NKG2DR on the surface of effector cells is correlated with an increased cytotoxic capacity.

Summary / Conclusions: Different enhancers can be used to increase the level of transduction while increasing the fold expansion of the cells. NK cells from both CB and AB samples can be successfully transduced with NKG2D-CAR. There is an increase in the level of degranulation of NKs transduced with NKG2D-CAR compared to MOCK-NK cells when faced with NKG2DL. When NKG2D-CAR-NK cells are exposed to Jurkat cells, the specific lysis of the target cells is higher than in MOCK-NK cells. NKs transduced with NKG2D-CAR could be good candidates for the treatment of solid tumors such as OS.

P877 | Abstract withdrawn

P878 | To wash or not to wash cryopreserved unrelated peripheral blood stem cells for transplantation?

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Background: Previous studies have shown that cryopreservation guarantee the potency of the allogeneic peripheral blood stem cells (PBSC). However, different policies exist regarding washing allogeneic cryopreserved products and a few data is available about the impact of washing on clinical outcomes in allogeneic setting.

Aims: To investigate the primary clinical outcomes, such as myeloid engraftment, transfusion requirements, hemolysis and adverse reactions at infusion of patients, acute graft versus host disease (GVHD), progression-relapse free survival (PFS) and overall survival (OS), who underwent unrelated donor (UD) hematopoietic stem cell transplantation (HSCT) with washed cryopreserved PBSC and compare them with those of patients who had undergone UD HSCT with no washed cryopreserved PBSC

P878 - Table 1. Baseline characteristics of patients according to wash or unwashed UD cryopreserved PBSC

Characteristic	Unwashed (n = 14)	Washed (n = 8)	p-value
Age at transplantation (years)	53 (39-63)	57 (52-63)	0.356
Sex (female), n (%)	8 (57)	3 (38)	0.659
Disease, n (%)			0.516
Acute leukemia	8 (57)	3 (38)	
Mielodysplastic/ Mieloproliferative neoplasm	3 (21)	3 (38)	
Lymphoma	3 (21)	1 (13)	
Myeloma Multiple	0 (0)	1 (13)	
10/10 HLA compatibility, n (%)	13 (93)	3 (38)	0.011
Myeloablative conditioning, n (%)	4 (31)	1 (13)	0.606
GVHD PTCy prophylaxis, n (%)	11 (79)	4 (50)	0.343
Cellularity cryopreserved			
WBC × 10 ⁸ /kg	5.8 (4.5-7.9)	5.9 (5.1-8.6)	0.473
Neutrophil × 10 ⁸ /kg	1.2 (0.9-1.6)	1.0 (0.7-1.6)	0.891
CD34 ⁺ × 10 ⁶ /kg	6.0 (6.0-6.6)	6.3 (5.7-6.5)	1.000
Cryovial			
CD34 ⁺ /7-AAD negative (%)	91 (81-95)	81 (57-91)	0.232
Granulomacrophage-Colony forming units/kg	6.2 (5.4-7.5)	6.0 (5.6-12.1)	0.633

Results are reported as median (IQR) unless otherwise stated.

No graft failure, hemolysis or adverse reaction at infusion were observed in both groups. More platelet transfusion and days until platelet engraftment were observed in patients who received washed products (Table 2).

P878 - Table 2. Univariate analysis according to wash or unwashed products

Variable	Unwashed (n = 14)	Washed (n = 8)	p-value
Days to neutrophil recovery	19 (16-21)	21 (17-24)	0.730
Days to platelet recovery	28 (13-57)	46 (35-56)	0.071
Number of platelet transfusion requirement at 100 days	9 (4-17)	18 (7-41)	0.029
Number of RBC transfusion requirement at 100 days	8 (5-12)	12 (6-23)	0.180
Acute GVHD at 100 days, rate	29	38	0.780
PFS at 100 days, rate	93	87	0.960
OS at 100 days, rate	93	100	0.180

Results are reported as median (95% CI) unless otherwise stated.

Methods: From July 2020 to October 2023 two cohorts of UD HSCT were analyzed according to the use of washing or not washing cryopreserved unrelated PBSC (donor isohemagglutinine titles above or below 256, respectively). Patient-, disease-, transplantation, and graft-related factors were compared between both groups using a

comparison test (Fisher and Wilcoxon). Time to primary outcomes were calculated using survival analyses (log rank).

Results: No differences regarding patient-, and graft-related characteristics were observed between two cohorts but HLA match (Table 1).

Summary / Conclusions: In summary, washing cryopreserved PBSC in allogeneic setting was safe for recipients. However, a higher platelet transfusion requirements at 100 days after transplantation and a trend of delay platelet engraftment was observed in wash cohort.

P879 | Abstract withdrawn

P880 | Improving CAR-NK cells performance against target cells - comparative study of transduction enhancers in game

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Background: B-cell hematologic cancers such as leukemia and lymphoma are common forms of pediatric and adult cancers worldwide. Acute Lymphoblastic Leukemia (ALL) is the most common cancer among children with a prevalence of 20%-25% of all cases. The survival rate for these patients at 5 years is 79.2%, but it is still an incurable disease in many patients. Chimeric Antigen Receptor (CAR) T cells therapy has arisen as a new alternative to conventional therapies in order to treat advanced refractory cancers. However, this therapy has some undesirable side effects such as cytokine release syndrome (CRS) and neurotoxicity. Also, despite the good results in clinic, some patients are refractory or relapsed after CART cell treatment. Moreover, another potential problem with this treatment could be the need of using allogeneic T cells when is not possible to perform an apheresis to the patient, as allogeneic T cells carry a risk of graft versus-host disease (GVHD). NK cells could be a good alternative for CAR based therapy, as they do not cause this kind of side effects, and exhibit a potent graft versus leukemia (GVL) effect without causing GVHD. However, NK cells are known to be resistant to be infected with any viral particle, which leads to low transduction ratios. This could translate into less specific effector-target recognition and poorer CAR-NK performance. In this study, we compare different transduction enhancers to determine which one offers a more favorable environment to the NK cells, in terms of not only transduction, but viability and performance as well.

Aims: Determine the best transduction scenario for NK cells in order to improve the viability, transduction efficiency and cytotoxic performance of the cells.

Methods: (1). CD19-CAR transduction of NK cells with different enhancers (2). Check of viability and transduction in NK cells by flow cytometry (3). Cell cycle and apoptosis analysis (4). Functionality assays against CD19 presenting target cells.

Results: Polybrene provides a great transduction efficacy; nonetheless, NK cells viability gets much compromised, resulting in the lack of expansion of the cells. The other enhancers result in great transduction efficacy, while maintaining the viability and functions of NK cells. Vectofusin is an optimal option to translate CAR-NK cells to the clinic, as it allows a great transduction efficiency and GMP versions are available

Summary / Conclusions: Although polybrene has been extensively used for transduction, it is not the best option for the integrity of the NK cells. Vectofusin presents a great option for the translation of the CAR-NK therapy from the bench to the clinic, as it offers an affordable GMP version while performing remarkably. Apart from using different enhancers, the conditions of the transduction protocol of NK cells are crucial for an optimal infection outcome.

P881 | Abstract withdrawn

P882 | Possibility of Hemopoietic Progenitor Cells (HPC) as an indicator for CD34-positive cell counts in allogeneic peripheral blood stem cell harvest from G-CSF mobilized donor

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Background: Peripheral blood stem cell harvest (PBSCH) is a safe and convenient method of collecting stem cells, but to ensure a successful stem cell transplant, the required number of cells must be reliably collected. In general, the number of peripheral blood stem cells is measured by counting CD34(+) cells using flow cytometric analysis. Currently, HPC (hematopoietic progenitor cells) count using Sysmex XN series hematologic analyzer is reported to have a good correlation with CD34(+) cells. Therefore, we have been investigating its clinical application.

Aims: We examined if HPC could be an indicator of mobilized CD34(+) cells in PBSCH.

Methods: We prospectively analyzed CD34(+) cell counts and HPC counts using the same sample in 57 patients who underwent autologous PBSCH (APBSCH) and in 8 related allogeneic PBSC donors. Stem cells were mobilized with G-CSF + plerixafor in all patients and with only G-CSF in related donors. HPC counts were measured using a

Sysmex XN-1000 analyzer and CD34(+) cell counts were analyzed by flow cytometry in accordance with the ISHAGE protocol. We analyzed both peripheral blood (PB) and apheresis products. PB samples were collected on the day before the PBSCH, the morning of the apheresis, one hour after the beginning of the apheresis, and at the end of the apheresis. Apheresis samples were collected from the bag one hour after the beginning and at the end of the apheresis. This prospective analysis was approved by the ethics committee of Keio University School of Medicine.

Results: In the APBSCH group, the median age was 60 (range: 38 - 70) years old. The underlying diseases were malignant lymphoma ($n = 29$), plasma cell dysplasia ($n = 27$) and acute promyelocytic leukemia ($n = 1$). In the related donor group, the median age was 42 (range: 25 - 53) years old. In total, median processed blood volume was 5.57 (range: 3.07 - 10.9) L, median run time was 180 (range: 110 - 292) min and median CD34(+) cells per patient weight was 3.40 (range: 0.32 - 13.47) $\times 10^6$ /kg. Median collection efficiency was 52.8 (range: 19.8 - 95.3) %. In the APBSCH group, the correlations between HPC counts and CD34(+) cell counts in PB and apheresis products were weak to moderate in both patients with malignant lymphoma ($r = 0.56$ in PB, $r = 0.37$ in products) and plasma cell neoplasms ($r = 0.59$ in PB, $r = 0.25$ in products), while the correlations were much stronger both in PB ($r = 0.92$) and products ($r = 0.98$) in the related donor group. To assess the consistency of HPC and CD34(+) cell counts in the samples, HPC counts of FACS-sorted CD34(+) and (-) fractionated cells in apheresis products from 13 patients were re-analyzed using the XN series. HPC were detected not only in CD34(+) fraction, but also in CD34(-) fraction. The median HPC percentage is 86.1 (range: 34.4-95.0) % in CD34(+) fraction and 8.05 (range: 3.45-16.87) % in CD34(-) fraction.

Summary / Conclusions: In related donors, there was a strong correlation between HPC counts and CD34(+) cell counts while HPC counts differ significantly from CD34(+) cell counts in APBSCH patients mobilized with G-CSF + plerixafor in some cases. HPC could be a useful surrogate marker of the CD34(+) cell counts in donors who mobilized with G-CSF without plerixafor. Sorting fraction analysis showed that HPC exists in CD34(-) fraction as well as CD34(+) fraction. It may be one of the possible explanations for the discrepancy between the result of the two methods.

P883 | Role of NK cells induced from PBMCs in killing leukemia cells based on an in vitro culture

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Background: Natural killer (NK) cells have the ability to identify and kill target cells without MHC restriction, and are widely used in tumor immunotherapy. Therapeutic NK cells can be obtained from various sources, including umbilical cord blood (UCB), peripheral blood mononuclear cells (PBMCs), induced pluripotent stem cells (iPSC), and NK cell lines. Among these, PBMCs have been recognized as a promising

source for cell immunotherapy due to their convenience for obtaining and isolation. However, the main practical problem is that the quantity of NK cells isolated from PBMC are not adequate for clinical treatment in vitro.

Aims: In the present study, we induced and expanded NK cells derived from PBMCs in vitro, then we activated and detected the cytotoxic effect of NK cells on Leukemia cell line (K562) to evaluate its feasibility in the treatment of leukemia.

Methods: 2 ml of heparin anticoagulated whole blood was collected from a female healthy individual with her informed consent. This study was approved by the regional ethics committee in Blood center of Zhejiang Province. PBMCs was separated by ficoll density gradient separation solution. The isolated PBMCs were calculated and adjusted to 3×10^6 , then were cultured in NK cell culture medium with 10% her serum. The density was adjusted to $0.8-1 \times 10^6$ every two days with new medium. Specific activators were added to induce cells developing towards NK cells. After culturing for 14 days, the obtained cells were collected and the surface markers of CD3 and CD56 were detected by flow cytometer. The cytotoxic effect of cultured NK cells on K562 cells was detected by LDH release assay.

Results: The culture showed that the isolated PBMCs maintained a high activity rate of over 90% during the whole growth and expansion process. With the increase of culture days, the cells were observed to grow in clusters and proliferate rapidly. After 14 days of culture, all cells showed CD3 negative, and 80% cells showed CD56+CD3-, which meant that most PBMCs were induced to NK cells. The cytotoxicity of NK cells against K562 cells was 70.99%, 35.42%, 24.05%, and 20.52% at different effector-target ratios (10:1, 5:1, 2.5:1, and 1.25:1) indicating that the cytotoxicity increased as the effector-target ratio increased. These results indicate that PBMCs induced amplification in vitro can yield functional NK cells with high purity and sufficient quantity.

Summary / Conclusions: The induction and expansion of NK cells from healthy individuals PBMCs in vitro is expected to be a promising source of cellular immunotherapy for leukemia.

P884 | Abstract withdrawn

P885 | Is total cell recovery a surrogate marker for the quality of cryopreserved peripheral blood apheresis products?

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Background: The CD34+ cell dose infused in autologous hematopoietic stem cell transplant (HSCT) is a predictor of hematopoietic engraftment success. A CD34+ cell dose $>2 \times 10^6$ /kg of patient bodyweight correlates with rapid engraftment, while low doses seem to especially retard platelet engraftment. Several factors, such as

centrifugation, storage conditions and the thawing process can cause cell loss during processing of apheresis products (AP). Therefore, quantifying viable CD34+ cells post-thawing may have clinical relevance, especially in AP with borderline content. However, this is often not feasible in clinical practice due to pre-analytical and logistical reasons. The paucity of evidence in this regard highlights the need for more available variables, such as the recovery of total nucleated cells (TNC), which can indirectly report about the total cellular loss in the AP.

Aims: To assess the impact of low total TNC recovery (TNCr) in thawed AP on leukoplatelet engraftment (LPE).

Methods: Observational and retrospective study on AP of patients undergoing an autologous HSCT during 2023 at an academic hospital. TNCr was defined as (TNC post thaw/TNC pre thaw) x100. The results are expressed as median (range). LPE was determined according to EBMT recommendations. Differences in the LPE rates were compared (Mann-Whitney U test) between the groups of patients who received a product with a TNCr over or under 70%, as this was our customary acceptance rate.

Results: Thirty-five patients were included, 22 (71%) men. The median age was 55 years (30-70) and weight 77 kg (57-118). Eighty-four percent were plasma cell diseases, 12.5% diffuse large B cell lymphomas; and 6.3% were Hodgkin lymphomas. G-CSF alone was used as mobilization agent in 93.5% of patients, and chemomobilization in 6.5%. Preemptive plerixafor was used in 37.5% of patients due to poor mobilization. The median cell count of the collected products was 5.6×10^8 TNC/kg, $3.3 \text{ CD34} \times 10^6$ /kg and 5.1×10^8 mononuclear cells (MNC)/kg. The thawed products had a median cell count of 2.1×10^3 TNCs/kg (0.7-3.3), 0.5×10^3 MNC/kg (0.2-1) and 1.4×10^3 neutrophils/kg (0.1-2.3). The median TNCr was 92.2% (32.8% - 164%). Only 9.7% of the products had a TNCr < 70%. The leukocyte engraftment (100 and 500 neutrophils/ μ l) was achieved in a median of 12 (1-19) and 14 days (10-21), respectively. Platelet engraftment (>20,000 and >50,000 platelets/ μ l) was reached in 15 (12-29) and 18 days (13-31), respectively. No significant differences were observed in neutrophil engraftment time in those with a TNCr of < 70%. Nevertheless, this group presented a significant delayed early platelet engraftment (>20,000 platelets/ μ l) of 25 vs 14 days (p: 0.03).

Summary / Conclusions: Although we found a significant delayed on early platelet engraftment in patients with TNCr < 70%, this was not reproduced in late platelet engraftment (>50,000/ μ L) and, given the small sample size, we consider it as an incidental finding. TNCr seems to inform poorly about graft cell loss and seems not to be a predictor of significant delays in LPE. Two reasons may explain this findings: first, higher concentration of TNC in the AP has been shown to adversely influence the CD34 viability. Second, TNC loss happens mainly at the expense of granulocytes, which have lower osmotic tolerance than CD34 cells. Given that several studies show a significant loss of CD34 cells after thawing, their measure may be important in some settings, especially in the absence of other straightforward informative variables.

P886 | Relation between mobilization protocol in stem cell apheresis and product purity

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Background: An autologous stem cell transplant is part of the standard of care regimen for newly diagnosed multiple myeloma (MM) patients and can be a part of therapy for lymphoma patients. All 289 apheresis performed in the Blood Bank of Hospital Mútua de Terrassa has been recorded between the years 2016 and 2023. There are 172 male patients (80 MM (46%), 1 POEMS (0.6%), and 91 lymphomas (53.4%) with a median age of 59 (age range 22-72), and 117 female patients (67 MM (57%) and 50 lymphomas (43%) with a median age of 59 (age range 24-73)). Blood groups are distributed as follows: 119 O (41%), 136 A (47%), 25 B (9%), and 9 AB (3%).

Aims: A descriptive study of hematopoietic stem cell (HSC) apheresis was conducted to assess if mobilization using granulocyte colony-stimulating factor (G-CSF) grants a purer final product than mobilization with both chemotherapy and G-CSF, regardless of diagnosis. All patients who used Plerixafor were excluded.

Methods: Apheresis was performed using a SPECTRA OPTIA[®] instrument in all patients. Vascular access was either via peripheral intravenous or a central venous catheter. The anticoagulant citrate dextrose solution, solution A, was used as an anticoagulant, and up to 6 blood volumes were targeted for collection. All patients were 22- to 73-year-old adults eligible for an autologous stem cell transplant. For evaluation, patients were divided into two groups. Those mobilized using G-CSF: 199 patients (68%), 147 (73%) with a MM diagnosis, 52 (27%) with a lymphoma diagnosis and 1 with a POEMS diagnosis; and those mobilized using G-CSF after a chemotherapy cycle: 90 patients (31%) all with a lymphoma diagnosis. The main parameters for evaluation were product purity (% MNC), defining adequate purity as >70%, and the range of HSCs mobilization, defining adequate mobilization as >10,000 CD34+ cells/ μ L.

Results: In the chemotherapy + G-CSF group, 74 were mobilized without Plerixafor: 68 (92%) had an adequate mobilization, 27 (40%) of those had >70% MNC; 6 (8%) had an inadequate mobilization and of those 2 (33%) had >70% MNC. The mean product purity was 62% in adequate mobilizations and 66% in inadequate mobilizations. And

P886 - Table 1.

Chemotherapy + G-CSF (74)	>70% MNC	<70% MNC
Adequate mobilization (68, 92%)	27 (40%)	41 (60%)
Inadequate mobilization (6, 8%)	2 (33%)	4 (66%)
G-CSF (160)	>70% MNC	<70% MNC
Adequate mobilization (150, 93%)	126 (84%)	24 (16%)
Inadequate mobilization (10, 7%)	8 (80%)	2 (20%)

in the G-CSF group, 160 were mobilized without Plerixafor: 150 (93%) had an adequate mobilization and 126 (84%) of those had >70% MNC; 10 (7%) had an inadequate mobilization and of those 8 (80%) had >70% MNC. The mean product purity was 83% in adequate mobilizations and 89% in inadequate mobilizations.

Summary / Conclusions: In conclusion, in our experience, both mobilization protocols are adequate for successful HSCs as there are no differences observed in the adequate range of mobilization (93% in G-CSF and 92% in G-CSF + chemotherapy). However, regardless of adequate or inadequate mobilization, we found more products with adequate purity are obtained from patients using only G-CSF as a mobilization agent compared to the other protocol [84% in G-CSF, 40% in G-CSF + chemotherapy in adequate mobilizations; 80% in G-CSF, 33% in G-CSF + chemotherapy in inadequate mobilizations].

P887 | Extracorporeal photopheresis in treatment of graft versus host disease using new online system

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Background: Hematopoietic stem cell transplantation (HSCT) is a potentially curative treatment of hematologic diseases, Graft versus host disease (GVHD) is still a limiting factor for the outcome of these patients. Corticosteroids remain first-line therapy for both acute and chronic GVHD, but it is associated with significant toxicity and an increasing number of patients develop a steroid-refractory disease. Extra Corporeal Photopheresis (ECP) is a second-line treatment, particularly in steroid-refractory form of GVHD with a response rate of 80% in acute and chronic GVHD patients.

Aims: The aim of this paper is to report the authors' experience in the treatment of GVHD post HSCT using a new online method recently made available in Italy.

Methods: In our apheresis unit we adopted the Fresenius-Kabi Amicus Blue online FCE system which is composed of the AMICUS cell separator and the PHELIX photoactivation device, we used the Correct Connect circuit which allows to perform both single and double needle procedures. Collection Efficiency (CE) was calculated using the CE2 equation that requires the total volume processed, pre procedure blood mononuclear cells (MNC) count and MNC count in apheresis product.

Results: As reported in table 1, from August to December 2023, 84 EP procedures were performed in six patients with GVHD arising after HSC transplant from a familial (1) or unrelated (5) donor. Each patient received from 5 to 25 EP procedures; mean processed volume was 2043 ± 85.6 mL, mean time for procedure was 103 ± 9.7 min, mean collection efficiency for mononuclear cells was 63.6 ± 7.8%. FCE procedures have been generally well tolerated, we recorded 4 (4.7%) unwanted events: in one case we observed a febrile reaction and in three cases we had problems in managing venous access.

Summary / Conclusions: ECP exerts multiple effects on the immune system leading to the production of anti-inflammatory cytokines and reduced production of proinflammatory cytokines. In our experience the Fresenius-Kabi Amicus Blue online FCE system was fast (mean time for apheresis was 103 ± 9.7 min), safe (only 4.7% of observed side effects), highly automated, and productive. Collection efficiency (CE2) for MNC was satisfactory (from 52.9% to 75.1%). So, in our opinion, the Fresenius Kabi Amicus Blue System for online ECP appears to have the operational characteristics making it particularly suitable for ECP treatments in GVHD patients.

P887 - Table I: Results obtained evaluating 84 online ECP procedure performed in six patients with GVHD

Patient, Gender, Age	Main Clinical Localizzazione	Performed ECP Procedures	Mean Time / Mean Volume	Collection Efficiency (CE2)
BC, Male, 62	Skin	19	991 min / 1999 mL	75%
CA, Female, 65	Skin + Eye	8	97 min. / 1995 mL	58%
DOF, Female, 61	Eye + Joints	5	101 min. / 1951 mL	64%
DF, Male, 58	Gut	16	114 min. / 2167 mL	68%
PP, Male, 29	Skin, + Gut + Membranes + Lung	25	115 min. / 116 mL	53%
RS, Male, 38	Skin + Liver	11	103 min. / 2015 mL	62%

P888 | First step to success in car T-cell therapy: Increased collection efficiency of CD3+ cells with a new cell separator software version 6.1

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Background: Collection of autologous CD3+ cells using an apheresis device is the first step for manufacturing chimeric antigen receptor (CAR) T-cells. A new software version 6.1 for collecting cells is available for the Amicus Separator (Fresenius Kabi, Germany). The software includes a new optional method for transferring the collected mononuclear cells (MNCs) from the centrifuge pack to the final storage container, trying to improve MNC collection efficiency.

Aims: To compare the collection efficiency (CE1) of CD3+ cells using software version 6.0 and 6.1 on Amicus.

Methods: We retrospectively reviewed all MNC performed with Amicus for CAR T-cell manufacturing at our institution from January 2020 to December 2023. Only donors with pre-collection, post-collection and product collection CD3+ cell count were included. The collections were divided in two groups according to the date 30th of May 2023 when the new software v 6.1 was implemented. The basal characteristics of both groups were studied. Qualitative data were reported as number (frequencies) and compared with Chi Square two-tailed test. Comparisons between two groups for quantitative data, including CE1 efficiency formula (figure 1) were performed with U-Mann Whitney two-tailed test. A p value of less than 0.05 was considered significant.

Results: A total of 195 autologous MNC collections were performed in 195 donors, 151 (77.43%) with software 6.0 and 44 (22.57%) with software 6.1. Table 1 shows baseline characteristics of both groups and no statistically significant differences between groups were observed. The median CE1 of CD3+ cells was 54.21 % (interquartile range: 22.75 % - 70.58%) with Amicus software 6.0 and 60.56% (interquartile range: 36.29% - 74.61%) with Amicus software 6.1, and such difference was statistically significant (p = 0.022).

Summary / Conclusions: The new Amicus software 6.1 provides a more efficient collection of CD3+ cells.

P888 - Table 1. Baseline characteristics of patients, according to Amicus software version.

		Amicus version 6.0	Amicus version 6.1
Sex*	Male n (%)	83 (55)	30 (68.2)
	Female n (%)	68 (45)	14 (31.8)
Age*	Median	58.00	58.00
	p25	44.50	49.00
	p75	66.00	66.00
	Mean	53.70	55.42
	Standard deviation	15.73	15.07
Diagnosis*	Acute lymphoid leukemia n (%)	48 (32.4)	8 (18.1)
	Non-Hodgkin lymphoma n (%)	44 (29.7)	17 (38.6)
	Chronic lymphocytic leukemia n (%)	22 (14.9)	4 (9.1)
	Multiple myeloma n (%)	32 (21.6)	13 (29.6)
	Mantle cell lymphoma n (%)	2 (1.4)	2 (4.5)
Vascular	Peripheral veins n (%)	148 (98)	41 (93.2)
Access*	Central line n (%)	3 (2)	3 (6.8)

* For all baseline characteristics studied, p-value was > 0.05.

P888 - Table 2. Collection Efficiency (CE1) of CD3+ cells according to Amicus software version.

CE1(%)	Amicus version 6.0	Amicus version 6.1	p value
Mean	54.21%	60.56%	0.022
Mediana	43.56	48.81	
Standard deviation	46.77	38.91	
Interquartile range	22.75 - 70.58	36.29 - 74.61	

P888 - Figure 1. Collection efficiency (CE1) formula.

$$CE1 (\%) = \frac{\text{Product cell count} \times \text{Product volume}}{\frac{\text{Precollection CD3 count} + \text{Postcollection CD3 count}}{2} \times \text{Whole blood proceeded}} \times 100$$

P889 | PBSC mobilization and collection in healthy pediatric sibling donors - a single center experience from South India

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Background: Peripheral blood stem cell transplantation (PBSCT) is a curative treatment for malignant and non-malignant diseases in children. Stem cell donors are often HLA-matched siblings belonging to the paediatric age group bring various challenges during stem cell harvest including lower extracorporeal blood volumes, difficult peripheral access and monitoring when compared with adult donors.

Aims: To review the PBSC mobilization and collection in healthy paediatric sibling donors in Paediatric PBSCT

Methods: PBSC collection data were retrospectively analyzed for all allogeneic paediatric PBSCT with pediatric sibling donors at Dr. Rela Institute & Medical Centre, Chennai, India between June 2019 to Sep 2023. PBSC collection were performed with Spectra Optia (Terumo Bct) and ACD-A anticoagulant using Continuous mononuclear cell collection (CMNC) protocol via a central line /peripheral line after G-CSF ± Plerixafor mobilization. Data collected includes donor weight, pre-procedural peripheral blood (PB) CD34+ cell counts, total blood volume (TBV) and whole blood processed volumes (WBPV), post collection processing, total CD34+ collection results, blood prime information, calcium prophylaxis, and adverse events.

Results: During study period, 121 pediatric allogeneic PBSCT were performed with matched related (MRD 78%) and unrelated donors (MUD 19%). Indications for PBSCT were Thalassemia ($n = 13$), hematological malignancies ($n = 5$), Immunodeficiency syndromes ($n = 5$), Others ($n = 3$). In MRD group, 29% were paediatric age sibling donors (brother 17, sisters 11; Full match 22, haplomatch 6), with mean age 9.5 years (range 3 -17 years), mean weight 35 Kg (12.5-80 Kg) with 11 donors requiring custom prime of extracorporeal circuit with red cells. Average Donor Recipient weight ratio (DRWR) was 1.9 (range 0.33-6.6) with DRWR <1 in 6. Pre-collection CD34 count was 165.9 cell / μ L (Range 13-673 cells / μ L). Inj Plerixafor was given in 9/28 donors mean dose 7.2 (range 3-13) mg with average Pre-CD34 count 255 cells/ μ L (53-673). 3 out 29 procedures were done using peripheral lines. During PBSC collection, WBPV, and TBV ratio was 5660 ml (1992 -15,333 ml) and 2.3 (1-3.8) volumes respectively. Donors were monitored and given calcium prophylaxis as continuous slow infusion. Mean duration of procedure was 220 minutes (range 99-326 min). The mean harvested product volume and CD34 cells dose per procedure was 166 ml (45-317 ml) and 16.2 (range 1.4 - 53.2) $\times 10^6$ cells /recipient body weight (kg). The targeted cell dose of 6 $\times 10^6$ CD34/recipient kg was met for all patients in one collection in 25 Patients and required 2 procedures in 3 patients. Out of total, 4 were minor ABO incompatible and required plasma depletion while 5 were major incompatible (3 ABO and 2 Rh) and red cell contamination were kept low using high collection preference settings and colour scale monitoring in Optia Spectra. All apheresis procedures were well tolerated.

Summary / Conclusions: PBSC collection from Paediatric sibling donors is a safe procedure and can be successfully completed with lab monitoring, altering collection parameters, and with a good coordination between transfusion specialists, Paediatric haemato-oncology and laboratory team.

P890 | Efficacy of autologous lymphapheresis for production and validation of the first CD19 chimeric antigen receptor T cell therapies in Mexico

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Background: Anti-CD-19 chimeric antigen receptor (CAR) T-cell therapy has revolutionized the treatment of neoplastic diseases. To guarantee the quality of the product, proper collection is essential; which is performed by apheresis of unstimulated peripheral blood. Collection efficiency (EC) >40%, absolute lymphocyte count >500/ μ L and >150 CD3+/ μ L cells prior to apheresis, has been reported to increase the possibility of reaching the target of >1 $\times 10^9$ T cells/product, sufficient to initiate the manufacture of CAR T lymphocytes. Similarly, a higher CD4+/CD8+ ratio prior to apheresis has been associated with better antitumor activity and greater treatment success.

P890 - Table 1. Comparison of pre-apheresis counts and final product

Variable	Median (Range) Pre-harvest	Median (Range) Harvest
Leukocytes k/ μ L	5.71 (4.9-6.0)	94.0 (77.0-117.5)
CMN %	43.30 (40.9-47.9)	97.1 (96.6-98.6)
CD45+ $\times 10^6$ cell/mL	4.10 (3.0-5.3)	76.6 (64.0-98.2)
CD3+ $\times 10^6$ cell/mL	1.00 (0.7-1.4)	40.9 (32.4-65.9)
CD4+/CD8+%	21.87 (19.3-32.2)	47.8 (44.8-59.4)

MNC: Mononuclear Cells.

P890 - Table 2. Correlation of results obtained against the expected objective

Variable	Median (range)	Aim
PRODUCT VOLUME	114.4 (58-163)	50 - 280 ml
CE2% (CD3)	68.9 (57.7-92.3)	>40%
CD3+ $\times 10^9$ Cell/Total Volume	6.1 (2.4-7.5)	>1.0 $\times 10^9$
Viability %	99.7 (89.2-99.8)	>80
Aerobic, anaerobic, fungal culture	No development	No development

Aims: The aims was to demonstrate the efficacy of autologous lymphapheresis in the production and validation of the first anti-CD 19 CAR T cell therapies for application in Mexico.

Methods: A prospective study was conducted in the hematology service of the "Dr. José Eleuterio González" University Hospital in Monterrey, Nuevo León, Mexico, during August to October 2023. Lymphapheresis was performed in healthy volunteer donors, previously a CD3+ count was performed with the MACS Quant10 flow cytometer, apheresis was performed using Spectra OPTIA cell separators; Aerobic, anaerobic and fungal culture tests were performed on the lymphocyte harvest, as well as blood count, flow cytometry for viability and lymphocyte quantification. Finally, the data obtained were collected and correlated using the parametric spearman test. The manufacture of CAR T cells in the CliniMACS Prodigy.

The following formula was used for the EC calculation:

$$CE2 = (T \text{ lymphocytes in apheresis product}) / (\text{Peripheral blood T lymphocytes/L} \times (\text{blood volume processed in liters}) \times 100$$

Results: A total of 5 donors were included, 4 male, with a median age of 33 years (range 26-50), the median blood volume processed was 1.5 (1-2) equivalent At 7822 mL of total blood volume processed (5051-9128 mL), procedures were performed through peripheral access with medians of time and velocity of 137 min (106-215) and 55 ml/min (38-60) respectively. A positive correlation was found between the CD3+ count pre-apheresis and harvest ($p = 0.65$), and a very high positive correlation between the percentage of CD4+/CD8+ cells before apheresis and harvest ($p = 0.93$, sig. = 0.022, $R^2 = 0.867$). The absolute lymphocyte count prior to apheresis was 2000/ μ L (1890-2630/ μ L). A higher percentage concentration of CD4+ vs CD8+ was observed pre-aférseis and at harvest, being 13.77% vs 8.31%, and 32% vs 19.69% respectively. No adverse reactions were reported during the procedures. Comparisons of pre-apheresis counts and final product are shown in Tables 1 and 2

Summary / Conclusions: Products derived from autologous lymphapheresis were shown to be optimal for successful production and validation of the first anti-CD19 CAR T lymphocytes in Mexico.

P891 | Impact of pre-transplant hematological parameters on peripheral blood stem cell product—insights from a quaternary care center in South India

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Background: Peripheral blood stem cell transplant (PBSC) collection by apheresis has become the preferred approach of collection in both Allogeneic and Autologous Hematopoietic stem cell transplants as it has fewer side effects and benefit of short time to engraft and faster immune reconstitution in the recipient. A stem cell dose of 4.0–5.0 $\times 10^6$ /kg of recipient is desired for a successful engraftment. To achieve the required dose, peripheral blood CD34 cell count is

essential along with blood counts to assess the volume to be processed in apheresis.

Aims: To analyze the prehematological variables in the PBSC donor that correlates better with the product CD34 yield for efficient collection.

Methods: This is a prospective study done from April 2021 to December 2023. PBSC collection was done by Apheresis using Spectra Optia[®] Apheresis system(Terumo Bct,Lakewood) by the transfusion medicine team after careful assessment of the stem cell donor. PBSC collection was done on Day4 or Day5 of G-CSF administration. Data was entered manually in Excel and analysis was carried out on allogenic transplants with respect to the donor and patient characteristics, procedure details, correlation between pre transplant hematological variables like Peripheral blood (PB) white blood cell Count (WBC),PB-CD34 count, PB-Mono nuclear Cell (MNC) Count, and the product variables (harvested CD34, WBC, MNC) was assessed using SPSS version 20.

Results: A total of 81 patients underwent 85 allogenic PBSC transplants in which Matched Related Donor(MRD) were 38 (46.9%), and Haploidentical (HID) were 43 (53%) during the study period. Mean PB-CD34 cells was 168.91 ± 120 (34-673) cells/ μ L and whole blood processed was 6350 ± 2586 (1140-15037) ml. Mean CD34 harvest processed per litre in all the procedures was 4.7×10^6 /Kg /L ± 4.4 (0.01 to 24.9). In our study, Harvested CD34 count in the product showed positive significant correlation with PB-CD34 count (PCC = 0.701; p-0.000) than PB WBC (PCC = 0.416, p-0.000) & no significant correlation with PB MNC(PCC = 0.096, p-0.386).

Summary / Conclusions: PB-CD34 count helps to achieve adequate collection avoiding over collection which is proved in our study. Our analysis also confirms a good correlation between PB-CD34 cells and the harvested CD34 cells in the product and is a good guide for the efficient collection and improved donor safety in apheresis.

P892 | Infectious complications in patients undergoing extracorporeal photopheresis—experience in a center

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Background: Extracorporeal photopheresis (ECP) is an immunomodulatory therapy indicated in the treatment of cutaneous T-lymphoma, acute graft-versus-recipient disease (aGVHD) and chronic graft-versus-recipient disease (cGVHD), solid organ transplant rejection, and autoimmune disorders resistant to conventional treatment. One of its advantages is that it does not cause immunosuppression or drug interactions. Although it has been shown to be effective, safe and well tolerated, its application in severely immunosuppressed patients.

Aims: To describe the incidence of infectious complications in patients with ECP in our center.

Methods: We retrospectively reviewed the medical records of patients who underwent ECP for any diagnosis in our center. Analysis period: 2014-2022.

P892 - Table 1.

	SS/MF	GVHD
Number of patients	9	31
Central catheter	3 (33.33%)	28 (90.32%)
Catheter infection	3 (100%)	6 (21.42%)

The mean number of procedures performed in SS/MF with catheter until the infection was documented was 5.66, while SS/MF who performed peripheral procedures performed a mean of 16.66 procedures and did not present any infectious complications. The mean number of procedures performed on catheter-bearing patients with GVHD in which catheter infection was documented was 26.

Results: Number of patients included in the study: 41. Mean age 45.17 years (5-77). Sex: male 27 (65.85%). The indication for ECP was: S. Sezary/Mycosis fungoides (SS/MF) 9 patients (21.95%), GVHD 31 (75.60%), acute GVHD 9 (21.95%), chronic GVHD 22 (53.65%), Others 1 (2.43%). Most of the patients required a central venous catheter: 32 patients (78.04%). The mean number of procedures performed was 23.44 (2-51). Three patients underwent more than one cycle of procedures. The response to the ECP was: complete 9 (21.95%), partial 16 (39.02%), non-response 16 (39.02%). Five patients died (12.19%), 2 due to acute GVHD and 3 due to intercurrent infection. Infections were documented in 12 patients: central catheter infection in 9 patients (21.95%) a total of 12 episodes (three patients had two episodes) In addition, 3 infectious conditions not associated with catheter infection were diagnosed: 2 pneumonias and 1 sepsis. The analysis of patients with SS/MF compared to those with GVHD is described in table 1.

Summary / Conclusions: Patients undergoing ECP frequently require a central catheter (78.04%). The incidence of catheter infections in the analyzed population was 21.95%. If we analyze different diagnoses, in the subgroup of patients with SS/MF only 33.33% required a catheter but all of them presented catheter infection with a smaller number of procedures performed until the infection was documented. Possibly skin involvement in this group of patients is a predisponent condition to colonization by skin germs.

P893 | Experience in CAR-T apheresis and its application in pediatrics

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Background: On January 20, 2016, the Sant Joan de Déu University Pediatric Hospital (Esplugues de Llobregat, Barcelona), begins the Chimeric antigen receptor (CAR) T-cell therapy (CAR-T) therapy as a clinical trial and is subsequently approved by the American FDA and AEMPS in its indication as treatment.

Aims: The period to be analyzed was from January 20, 2016 to March 10, 2023. The objective is to perform a retrospective analysis of the experience in lymphocyte collection for CAR-T therapy in pediatrics.

Methods: The diagnoses of the patients who underwent CAR-T therapy at our center were mostly Acute Lymphatic Leukemia type B (B-ALL) (99%) and Non-Hodgkin Lymphoma type B (1%). The patients previously underwent a pre-apheresis study that incorporates: hepatitis B and C serology, HIV, HTLV1/2, Treponema pallidum, Chagas and malaria (if applicable), ABO and Rh group, screening for irregular antibodies. Apheresis procedures were performed with the cell separator. The age of the patients undergoing this therapy was between 2 and 27 years, with the average being 9.71 years. Patients weighing less than 25kg underwent prior priming of the collection machine with irradiated red blood cells, patients weighing more were assessed based on their analytical parameters (hematocrit and hemoglobin) and its general condition. An average of 3.36 volumes were processed to reach the number of cells requested by the corresponding type of trial or treatment.

Results: The number of patients undergoing CAR-T therapy was 100: 59 men and 41 women and 105 aphereses were performed, mean age 9.71 years (range 2-27). 99% of the treated patients were diagnosed with B-ALL and 1% with type B NHL. All apheresis procedures were performed through a central catheter. The average weight of the patients was 35.68kg, range (14-130kg), of which 53 required priming of the machine with irradiated red blood cells. The average number of blood volumes processed was 3.36. An average of 7435.54 ml were processed to obtain the requested number of cells.

Summary / Conclusions: Most procedures have been performed without adverse effects. 5 procedures have been repeated due to low cell count in the first apheresis. In 4 cases, apheresis could not be performed due to malfunction of the apheresis catheter and in one of them due to catheter infection. It is concluded as a safe procedure, to continue with its application in pediatric patients and in the future in new pathologies.

P894 | Abstract withdrawn

P895 | Abstract withdrawn

P896 | Abstract withdrawn

P897 | The current efficacy and safety of hematopoietic stem cell collection by apheresis for transplantation—a prospective one-year study

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Background: Hematopoietic-cell transplantation (HCT) is widely used in the management of inherited and acquired disorders such as hematopoietic malignancies, solid tumors and, in recent years for autoimmune and metabolic disorders. Currently, the main source of hematopoietic stem cells (CD34+) for transplantation is peripheral blood, after an appropriated mobilization regimen, using apheresis. Thanks to the improvements in the mobilization and collection processes, the number of apheresis

P897 - Table 1.

	Healthy Donors	Patients
N	36	57
Age*	46.5 (15 – 74)	57 (18 – 69)
Gender (women; %)	16; 44.4	28; 49.1
Diseases	-	MGM: 34 NHL: 11 HL: 7 Autoimmune: 4 Breast cancer: 1
Total blood volume (mL)*	4.667 (3.451 – 7.305)	4.531 (2.737 – 6.468)
GCS-F µg/Kg*	11.1 (8.8 – 22.2)	21.2 (3.5 – 24)
Chemotherapy (%)	-	33%
Plerixafor (%)	16.6	47.3
Peripheral venous access for collection	36 (100%)	52 (91.2%)
Number of collections		
1	36 (100%)	53 (93%)
2	0	4 (7%)
Collection duration (min)*	214 (55 – 335)	252 (92.0 – 314)
Volume processed (mL)*	14,000 (2,462 – 35,000)	18,000 (5053 – 31,029)
Total blood volumes processed*	2.7 (0.4 – 6.6)	4.0 (1.0 – 8.8)
Maximum ACD-A rate* ⁺	1.4 (0.6 – 2.4)	1.6 (0.8 – 2.4)
Volume of ACD-A used (ml)*	550.5 (107 – 1355)	722 (202 – 1172)
Volume solution Ca-Mg infused (mL)*	224 (39 – 442)	271 (68 – 442)
Adverse events (any):	5 (13.8%)	6 (10%)
Headache	2 (5.5%)	0
Nausea	1 (2.8%)	3 (5%)
Vomiting	0	1 (1.7%)
Dizziness	0	1 (1.7%)
Tingling	2 (5.5%)	1 (1.7%)

* Median (range).

MMG: malignant monoclonal gammopathies.

NHL: Non-Hodgkin lymphomas.

HL: Hodgkin lymphoma.

⁺ ACD-A rate in ml ACD-A/L/min.

collections to achieve the desired CD34⁺ goal and the adverse events has been reduced.

Aims: To prospectively analyze the efficacy and safety of CD34⁺ mobilization and collection by apheresis at our institution during one year.

Methods: Patients were mobilized using G-CSF with or without chemotherapy. Healthy donors were mobilized with G-CSF. In both cases, plerixafor was administered pre-emptively in case of poor mobilization. Apheresis collections were performed using, whenever possible, a peripheral vein access (with ultrasound guided vein canalization) in a Spectra Optia separator with the 12.0 version software. Double citrate concentration ACD-A (citrate concentration 42.4 mg/mL), in

order to reduce the final positive balance, was used at a ratio of 1/24 to 1/30. Prophylactic calcium and magnesium (1 mol of calcium per 10 mol of citrate) were administered intravenously. Complete blood count (CBC), ions, venous blood acid-base balance, and CD34⁺ count were measure before and after collection. A CBC and CD34⁺ count were performed from the collection bag. Adverse events were prospectively followed and recorded. Collections were stopped when the required amount of CD34⁺ was reached.

Results: From January 1st to December 31st, 2023, 97 apheresis collections were performed to 93 patients. The results are summarized in Table 1

Summary / Conclusions: Our current strategies for mobilizing CD34⁺ to peripheral blood and prophylactic measures taken to prevent metabolic changes provoked by citrate infusion during the apheresis allowed us to collect the desired goal of CD34⁺ for HCT in an effective and safe manner for healthy donors and patients, mostly using peripheral accesses in a single collection, even when processing of large volumes of blood is required.

P898 | Bed-side versus on-site thawing of cryopreserved hematopoietic stem cells

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Background: Thawing of hematopoietic stem cells cryopreserved in liquid nitrogen can be administered either patient's bed-side (BS) or on-site (OS, in the Laboratory of Blood Centre). Since prolonged exposure of HSCs to the cryoprotectant agent DMSO negatively impacts cell viability, several centers perform laboratory thawing using automated washing to reduce adverse reactions and cell damage. We evaluated the feasibility of performing thawing in the laboratory without automated washing, maintaining cell product viability, procedural safety and cost effectiveness.

Aims: We aimed to compare two hematopoietic stem cell (HSC) thawing protocols: BS and OS. Our primary endpoint was to evaluate differences in engraftment time, while a secondary endpoint assessed post-infusion infection risk.

Methods: Phase 1: we evaluated and validated in 3 consecutive batch/runs the CD34⁺ viability% (V%) at baseline (T0) and at T +8 min (T8), twice the Laboratory to bedside transfer time. Aliquots taken at T0 and T8 were labeled with MoAb CD45/CD34/7AAD (ISHAGE met.). Measurement was performed by FACS Cantoll (BD). Phase 2: we evaluated 52 patients affected by Multiple Myeloma, who underwent an Autologous Transplant at Campus Biomedico Polyclinic, in Rome. In 26 patients Autologous Transplants were administrated between 2012 and 2018; using thawing BS with water bath at 37°C. In the other 26 patients, Autologous Transplants were administrated between 2021 and 2023, using thawing OS, with water bath at 37°C. Post-thawing CD34⁺ cells count was 5 × 10⁶/Kg in BS

and $4 \times 10^6/\text{Kg}$ in OS. In our study we measured, for each patient, time to Neutrophils engraftment (considered as Neutrophils count higher than $500/\text{mm}^3$ for at least three consecutive days) and to Platelets engraftment (considered as Platelets count higher than $20,000/\text{mm}^3$ for at least three consecutive days). We also registered cases of CMV and Bacterial Infections in both groups.

Results: Phase 1: the viability of the samples at T8 (83.4%) differed by -1% compared with T0 (83.9%). There was no statistically significant difference between V%T0 and V%T8 ($p < 0.7$), with a V%T8/V%T0 Ratio of 0.99 and a correlation index of 0.91. Phase 2: the two thawing settings showed no significant statistical differences in days to Neutrophils engraftment (BS: 10.2 days; OS: 10.4 days) and days to Platelets engraftment (BS: 13.2 days; OS: 13.6 days). CMV infections occurred in 2 patients of BS; there was no CMV infection in patients of OS. Bacterial infections occurred in 3 patients of BS and in 2 patients of OS.

Summary / Conclusions: Our analysis at validated timepoints revealed no detrimental effect of DMSO on the viability of infused CD34+ cells, nor were any adverse reactions observed. We also demonstrated that OS is comparable to BS with regard to engraftment and risk of infections during the immuno-deficiency phase which comes before the hematologic recovery. OS thawing ensures product quality, eliminates the transportation of the cryogenic container between departments, enhancing procedural safety and minimizing qualified personnel redeployment.

Cellular therapies—clinical applications

P899 | Gene editing-based therapy to restore functional hematopoietic lineage from APDS2 patient-derived hiPSCs

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Background: APDS2 syndrome is an inborn error of immunity caused by mutations in *PIK3R1* gene. Such genetic alterations affect to

immune cells, resulting in an immunologic dysfunction. APDS2 patients have a high susceptibility to virus and bacterial infections, autoimmune diseases and even cancer. To date, the only available treatment against APDS2 disease consists in hematopoietic stem cells transplantation, bringing poor efficacy and many adverse effects.

Aims: For this reason, our aim is to develop novel gene and cell strategies to uncover effective specific-therapies directed to APDS2 patients.

Methods: For this purpose, we nucleofect sgRNA-CRISPR/Cas9 complex to correct the *PIK3R1* mutation in the genome of an APDS2-derived iPSC line generated in our laboratory. Then, we select corrected clones by restriction enzyme digestion and Sanger sequencing, and assess off-targets. Next, we characterize corrected clones by the detection of pluripotency and trilineage differentiation markers. Finally, we differentiate the cells towards hematopoietic stem cells, T- and NK-cells and corroborate their functionality.

Results: Findings show that our nucleofected sgRNA-CRISPR/Cas9 complex presents an edition efficiency of a 60% without off-target effects or chromosome aberrations. The corrected clones maintain their identity as pluripotent stem cells and are able to differentiate towards the three germ layers more efficiently compared to the APDS2-mutated cells. Furthermore, our clones are able to be driven towards functional CD34+ hematopoietic progenitors more effectively compared to the APDS2-mutated cells. Finally, we generate fully functional T and NK lymphocytes derived from APDS2-edited clones, which restored normal immune cell activity and cytokines secretion compared to immune cells obtained from APDS2-mutated cells.

Summary / Conclusions: Taking into account such findings, our work represents an innovative therapy that could improve current treatments, providing an exceptional and powerful tool to study APDS2 syndrome as well as personalized gene and cell therapy. Moreover, our results could be extrapolated to the research of similar pathologies, facilitating the development of advanced targeted gene and cell therapies.

P901 | CD34+ cell dose—predictors and effect on patient outcome in HSCT—a retrospective cohort from a tertiary care health centre in India

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Background: Peripheral blood (PB) hematopoietic stem cell (HSC) is preferred source for hematopoietic stem cell transplant (HSCT). Predictors for HSC dose quantified by CD34+ cell count is debated in the past and its effect on patient outcomes is under evaluation. We attempt to combine the two and connect the HSCT chain.

Aims: Primary objective is to correlate pre-procedure PB hematological parameters to the product CD34⁺ cell count and then establish a minimum threshold of this parameter to obtain adequate CD34⁺ cell dose. Secondary objective is to analyze the effect of CD34⁺ cell dose on patient outcomes after autologous and allogeneic HSCT

Methods: A retrospective cohort analysis was conducted at a tertiary care health centre in India. All HSCT from January 2016 to December 2022 were included in the study excluding repeat salvage procedures. Donor details, HSC product details and patients' details were collected from the Hospital Information Management system (HIMS, Akhil Diagnostics Pvt Ltd). Multiple regression analysis was done for pre-procedure mononuclear cell count (MNC) i.e. absolute lymphocyte count (ALC) and absolute monocyte count (AMC), total leucocyte counts (TLC), and CD34⁺ cell count with the HSC product. Significant parameters were further analysed for minimum threshold to obtain an adequate dose of $2 \times 10^6/\text{kg}$ using ROC analysis. Patient outcomes were evaluated by categorising CD34⁺ cell dose into three groups: Group I < $5 \times 10^6/\text{kg}$, Group II $5-7 \times 10^6/\text{kg}$ and Group III > $7 \times 10^6/\text{kg}$. Categories were evaluated for overall survival (OS), relapse free survival (RFS), engraftment, primary graft failure (PGF) and non-relapse related mortality (NRM). Survival analysis was done considering non-informative censoring and adjusting for confounding factors in multivariate analysis to a fixed follow up date.

Results: Total 78 HSCT procedures (allogeneic: autologous = 50:28) were analysed for predictors of adequate dose and 63 patients (allogeneic: autologous = 43:20) were analysed for patient outcomes. Male:female ratio was 36:27. PB ALC ($r = 0.35$, $p = 0.07$) and AMC ($r = 0.09$, $p = 0.34$) had mild positive correlation while TLC ($r = -0.15$, $p = 0.47$) had negative correlation with product CD34⁺ cell count, though not significant. Significant correlation was observed with PB CD34⁺ cell count and product yield ($r = 0.64$, $p = 0.002$). ROC curves of pre CD34⁺ cells predicted dose adequacy with 25 cell/ul in PB (area under curve 82%) with 92% sensitivity and 75% specificity. Six year OS and RFS was 61.9% and 46% respectively. Incidence of relapse (RI), PGF, NRM, engraftment and GvHD was 17.4%, 11.1%, 14.2%, 85.7% and 18.6% respectively. CD34⁺ cell dose showed no association with OS in both allogeneic ($p = 0.37$) and autologous ($p = 0.63$) HSCT. Dose categories II (HR: 0.003, 95% CI: 0.00-0.659, $p = 0.035$) and III (HR: 0.001, 95% CI: 0.000-2.7, $p = 0.08$) had less risk of RI as compared to Category I. Dose did not affect engraftment in autologous ($p = 0.94$) or allogeneic ($p = 0.86$) HSCT. Category II (HR: 0.923, 95% CI: 0.15-5.5, $p = 0.93$) and III (HR: 0.91, 95% CI: 0.15-5.5, $p = 0.910$) had less risk of PGF as compared to Category I though not significant.

Summary / Conclusions: Study highlights the predictive strength of PB CD34⁺ cell count for an adequate CD34⁺ cell dose. MNC count may also serve as a rough guide to predict the dose when flow cytometry is unavailable. Dose effect on patient outcomes associate higher dose to less RI although overall survival remains unaffected. This study may be limited by informative censoring which requires competing risk regression analysis.

P902 | NK cells proliferation from blood donors and killing activity against tumor cells

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Background: Natural killer (NK) cells are important innate immune cells that monitor and rapidly eliminate senescent, infected and aberrant cells in the body. NK cells can be obtained from peripheral blood, cord blood, or induced pluripotent stem cells. PBMCs from blood donors contain large amounts of NK cells, but are discarded in the blood component. The isolation of NK cells from the buffy coat of blood donors can turn waste into treasure. Clinical treatment requires a large number of active NK cells, which can be met by in vitro expansion and activation.

Aims: NK cells were isolated from the buffy coat of blood donors, and the method of increasing NK cells by serum-free culture in vitro was studied. The amplified NK cells were used to kill various tumor cells and their killing function was studied.

Methods: NK cells in buffy coat of blood donors were separated by microbeads and cultured in four groups. Group A: control; Group B: Anti-CD16; Group C: Anti-CD137; Group D: Anti-CD16+Anti-CD137. On the 5th, 7th, 9th, 11th, 13th and 15th day of culture, the number of cells in each culture group was counted, and the expression of NK cell surface molecules was detected by flow cytometry. NK cells obtained by the optimal amplification scheme were used as effector cells to carry out killing tests on tumor cells according to different target effect ratio. The level of interferon- γ (IFN- γ) in the culture supernatant was detected by ELISA, and the killing activity of NK cells against tumor cells was analyzed by flow cytometry.

Results: After comparative analysis, the amplification efficiency of NK cells in group D was the highest. At 15 days, the amplification ratio exceeded 700 times, and the amplification efficiency in group D was significantly higher than that in other groups ($P < 0.05$). Therefore, the culture regimen of group D was selected as the regimen of serum-free expansion of NK cells in vitro. The killing test showed that NK cells expanded at different target ratios had a very strong killing ability against a variety of tumor cells. In addition, with the increase of efficiency target ratio, the killing of NK cells was stronger, and the release of IFN- γ was also higher.

Summary / Conclusions: Pre-coated with Anti-CD16+Anti-CD137 in group D could rapidly activate the proliferation of NK cells, and the amplified NK cells showed killing activity against a variety of tumor cells in vitro.

P903 | Dendritic Cell (DC) immunotherapy increases monocyte count in peripheral Blood mononuclear cells and attenuates CD80 expression in monocyte derived dendritic cells culture of breast cancer patients

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Background: Cell immunotherapy has opened new avenues in cancer therapy, with dendritic cell (DC)-based strategies holding promise. Autologous DC pulsed with germline cancer antigens might actively enhance the immune response against breast cancer. Furthermore, DC culture maturation status is essential in determining DC capacity to prime T-cells and induce anti-tumor activity. However, the effect of DC immunotherapy on immune cells, such as monocyte and DC culture maturation, has yet to be fully elucidated.

Aims: This research explores the effect of DC immunotherapy on the immune cells of peripheral blood mononuclear cells (PBMC) and the DC culture maturation of breast cancer patients.

Methods: Seven Breast Cancer (BC) patients were recruited to the study. PBMC was taken from 110 cc of peripheral whole blood, and monocytes were isolated. Monocytes were then treated for five days with Granulocyte Macrophage Stimulating Factor (GM-CSF) and Interleukin-4 (IL-4) to induce Monocyte Derived Dendritic Cells (MoDCs). MoDCs were then pulsed for two days with germline cancer antigens (GCA). MoDCs were transferred to subjects subcutaneously. One month after MoDCs' transfer, PBMC was taken from subjects. Monocytes, lymphocytes, and granulocytes were counted using flow cytometry. Monocytes' count and viability were measured using an automatic cell counter. Isolated monocytes were then differentiated to MoDC using GM-CSF and IL-4 and stimulated by GCA. MoDC is defined as HLA-DR+ CD11c+ CD209+ cells. Mature DCs were identified using anti-CD80 and anti-CD86 antibodies and were measured using flow cytometry. DC culture maturation status is determined by the proportion of DC culture with CD80 and CD86 of more than 75%.

Results: There is an increase in monocyte proportion (32.69 % \pm 12.97% to 40.39% \pm 15.69%; $p = 0.4$) and the total number of monocytes in PBMC of BC patients after DC immunotherapy (32,005 \pm 10.94 to 69,269 \pm 66.59; $p = 0.19$). Granulocyte proportion is also slightly increased (5.73% \pm 4.94% to 7.66% \pm 11.20%; $p = 0.69$), meanwhile lymphocyte is decreased (60.90% \pm 12.37% to 51.27% \pm 21.00%; $p = 0.20$). CD80 is markedly decreased after DC immunotherapy (84.3% \pm 11.64% to 69.2% \pm 16.14%; $p = 0.02$). Meanwhile, CD86 remained similar (95.9% \pm 3.40% to 94.6% \pm 3.45%; $p = 0.58$). The proportion of DC culture with CD80 of more than 75% decreased (71% to 29%). The proportion of DC culture with CD86 of more than 75% remained unchanged (100% to 100%). This research highlights the relationship between DC immunotherapy and monocyte count. Transfer of autologous DC pulsed with germline cancer antigen increases the monocyte count of BC patients. On the other hand, a marked decrease of CD80 suggests lower maturity of MoDC culture after DC immunotherapy. Low CD80 DC can induce tolerance that might hinder anti-

tumor activity. The shift toward tolerogenic phenotype is perhaps due to repeat stimulation with a similar antigen or lack of adjuvant. Therefore, the underlying mechanism, how this affects anti-tumor activity, and the relationship to clinical outcome should be studied.

Summary / Conclusions: DC immunotherapy increases the monocyte count of PBMC and decreases MoDC culture maturation in BC patients. Further study should be done to explore the effect on clinical outcomes.

P904 | SMAD4 regulates the expression of LCK affecting the proliferation of CAR-T cells

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Background: In vitro and clinical studies have confirmed that chimeric antigen receptor (CAR) T-cell therapy is an efficient treatment strategy for hematological tumors. However, CAR-T cell therapy applied to treat solid tumors or autoimmune diseases is still not ideal. One reason is that the proliferation of CAR-T cells was hindered and cannot play its killing function well. However, the regulatory mechanism of CAR-T cells proliferation is not fully understood till now. LCK regulates T cell activation and proliferation by involving in TCR signal transduction. The transcription factor SMAD4 also plays an important roles in T cell immune response. However, there was no much evidence show the regulation between SMAD4 and LCK in regulating the proliferation of CAR-T cells.

Aims: To explore the effects and mechanism of SMAD4 and LCK on the proliferation of CAR-T cells.

Methods: The basal activation of Jurkat-CAR cells were measured by flowcytometry, and the basal activation of LCK was measured by western blot with anti-phospho LCY(Y505) antibody. Lenti-virus infection was used to over-express or knock-down target genes as indicated. The mRNA and protein expression level were detected through qPCR and western blot, respectively. CCK-8 assay was used to measure the proliferation of CAR-T cells. To further explore the regulation between SMAD4 and LCK the luciferase assay was performed.

Results: The flowcytometry and western blot results showed that the recombinant expression of CAR in Jurkat cells significantly increased the basal activation level of CAR-T cells and LCK kinase, indicating that CAR-T cells are not exactly same as T cells. Over-express or knock-down the expression of LCK significantly promotes or inhibit the proliferation of CAR-T cells, respectively. Over-expression of SMAD4 promotes the proliferation of CAR-T cells, on the contrary, knock-down the expression of SMAD4 inhibited the proliferation of CAR-T cells. Knock-down the expression of LCK has no much effects on the expression of SMAD4. However, knock-down the expression of SMAD4 in CAR-T cells, the expression of LCK was significantly inhibited, indicating that SMAD4 is an upstream regulator of LCK regulating the expression of LCK kinase. The luciferase assay results showed that the transcription factor SMAD4 interacted with the promoter of LCK involving in regulating the expression of LCK kinase.

Summary / Conclusions: Taken together, our results showed that the transcription factor SMAD4 affected the proliferation of CAR-T cells by regulating the expression of LCK kinase.

P905 | Abstract withdrawn

P906 | Outcomes after ABO-incompatibility peripheral blood stem cell transplant a center experience

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Background: In ABO-incompatible (ABO-i) peripheral blood stem cell transplantation (PBSCT), incompatibility is determined by the donor/recipient blood group and corresponding hemagglutinins, whose titers (HT) can influence potential complications: hemolytic anemia (HA),

passenger lymphocyte syndrome (PLS), pure red cell aplasia (PRCA), acute graft-versus-host disease (aGVHD), and delayed engraftment.

Aims: To establish the prevalence of the minor and major ABO-i PBSCT consequences, as well as its differences between high and low HT groups.

Methods: Data was collected retrospectively identifying patients aged ≥ 18 undergoing minor and major ABO-i PBSCT from related donors, at our center between 2018 and 2023 (using RStudio version 4.3.1 for the analyses). HA definition included: decreased hemoglobin levels, unconjugated bilirubin elevation, increased LDH, or schistocyte presence. PLS was defined as HA appearing 7 to 14 days after PBSCT. Only grade III-IV aGVHD cases were included. Engraftment was defined as 3 consecutive days with absolute neutrophil count (ANC) $\geq 0.5 \times 10^9/L$, 3 consecutive days with platelet count $\geq 20 \times 10^9/L$, and the day of the last red blood cells (RBC) transfusion. Product quality controls were defined as: $\geq 4 \times 10^6$ CD34⁺/Kg; $\geq 70\%$ of Mononuclear cells (MNC); $\geq 1 \times 10^5$ granulocyte-macrophage forming units per/Kg, and $\geq 10\%$ of Eclone. Plasma reduction (SEPAX; smart-wash[®]) was performed for minor incompatibility with HT ≥ 256 . In major incompatibility, if RBC volume was ≥ 1 ml/kg (recipient weight), prod-

P906 - Table 1.

	Minor incompatibility		
HT	>256 (n = 4)	<256 (n = 18)	p-value ²
Patient's age	57 (52-62)	46 (41-57)	0.17
Patient's sex (male/female)	3/1	8/10	0.59
Donor's sex (male/female)	4/0	10/8	0.25
CD34 ⁺ /Kg	6.83 (6.01-7.12)	5.99 (5.05-6.06)	0.26
%MNC	92 (87-95)	83 (80-92)	0.3
Hemolysis	1 (25%)	6 (33%)	>0.99
aGVHD	1 (25%)	1 (5.6%)	0.34
ANC engraftment day	24 (21-25)	20 (18-21)	0.05
Platelet engraftment day	25 (22-34)	17 (13-26)	0.17
RBC engraftment day	54 (48-64)	27 (22-56)	0.19

P906 - Table 2.

	Major incompatibility (n = 24)		
HT	>32 (n = 15)	<32 (n = 9)	p-value ²
Patient's age	57 (43, 59)	56 (41, 64)	0.86
Patient's sex (male/female)	10/5	3/6	0.21
Donor's sex (male/female)	7/8	8/1	0.08
CD34 ⁺ /Kg	5.99 (5.03- 6.06)	6.01 (5.02-6.35)	0.56
%MNC	86 (81-93)	76 (76-88)	0.19
Hemolysis	3 (20%)	4 (44%)	0.36
aGVHD	0	0	0
ANC engraftment day	21 (19-24)	21 (18-24)	0.83
Platelet engraftment day	23 (16-24)	20 (12-26)	0.63
RBC engraftment day	53 (36-164)	69 (48-101)	0.75

uct was released recommending intensive hydration, and slow infusion.

Results: For minor and major ABO-i groups, means of CD34⁺/Kg and engraftment days are described in Tables 1 and 2. No statistically significant differences were found between patient's groups, according to age, patient/donor sex, CD34⁺/Kg or %MNC. In vitro clonogenic potential was optimal in all products. Neither were they found for HA, aGVHD, nor engraftments. No graft failure was observed in any group. PRCA was diagnosed in 4 (16%) patients from major ABO-i cohort, achieving engraftments at 6-12 months (1 of them belonging to the low HT group). In the minor ABO-i group, 2 cases (9%) of LPS were diagnosed (days +10 and +14)

Summary / Conclusions: Non-statistically significant differences were observed regarding HA, aGVHD, and engraftment in minor or major ABO-i PBSCT. Borderline results were observed in ANC between high and low HT groups in minor ABO-i cohort. This support that our procedures aimed at mitigating risks in ABO-i PBSCT have shown efficacy. Compared to the literature, our population exhibited lower rates of passenger LPS (9% vs. 15%) and PRCA (16% vs. 29%).

P907 | Application of hematopoietic stem cell transplantation in alleviation of symptoms in patients with adrenoleukodystrophy

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Background: Transplantation of hematopoietic stem cells is used not only in the treatment of hematological malignancies but also in the treatment or alleviation of symptoms of severe aplastic anemia, congenital anemia, hemoglobin abnormalities, congenital severe immune disorders and some metabolic diseases. Adrenoleukodystrophy (Siemerling-Creutzfeldt disease) is a genetically determined disease affecting men, with an X-linked inheritance type. A mutation in the ABCD1 gene is responsible for the symptoms of the disease. Affected males may manifest disease in the adrenal glands, testes, and/or central nervous system. Transplantation of hematopoietic stem cells, used

P907 - Table 1.

	Patient 1: DOB: 2017.07.12	Patient 2: DOB: 2020.05.14	Patient 3: DOB: 2020.05.14
HLA-A*	26:01:01G 30:01:01G	01:01:01G 25:01:01G	01:01:01G 25:01:01G
HLA-B*	13:02:01G 27:05:02G	08:01:01G 18:01:01G	08:01:01G 18:01:01G
HLA-C*	01:02:01G 06:02:01G	07:01:01G 12:03:01G	07:01:01G 12:03:01G
HLA-DRB1*	01:01:01G 07:01:01G	03:01:01G 16:01:01G	03:01:01G 16:01:01G
HLA-DQB1*	02:01:01G 05:01:01G	02:01:01G 05:02:01G	02:01:01G 05:02:01G

since the 1980s, helps alleviate the symptoms of the disease. The human leukocyte antigen (HLA) complex plays a crucial role in matching for hematopoietic stem cell transplantation (HSCT) because allele-level HLA matching between donors and recipients reduces the likelihood of rejection and graft-versus-host disease. The HLA genes encoding cell-surface proteins are the most polymorphic in the human genome. Therefore, the changes in their sequence affect the specificity of antigen presentation and histocompatibility in transplantation.

Aims: The aim of our study was to determine HLA antigens in three patients (male siblings) with adrenoleukodystrophy for HSCT.

Methods: DNA samples were isolated from peripheral blood mononuclear cells using NucleoSpin DX Blood kit (Macherey-Nagel). HLA typing for five of the HLA genes (A, B, C, DRB1, DQB1) was performed using sequence-based typing (SBT) method. All procedures on genotyping of the samples were performed according to the HLA assay manual (SeCore) using the Applied Biosystems 3500 Genetic Analyzer. The typing kits were targeted on the most hypervariable regions of the MHC class I and class II genes. The raw sequencing data were assembled and analyzed using uTYPE Software for SeCore.

Results: High resolution HLA typing was performed for three siblings suffering from adrenoleukodystrophy. The results are presented in the table below.

Summary / Conclusions: The global database found many donors potentially compatible with patients at the 10/10 allelic level, who come mainly from the Polish and German registry. In addition to HLA compatibility, the donors were compatible with the patients' blood type and CMV status. Symptoms of adrenoleukodystrophy appeared in patient no. 1, but were not observed in the twins. Boys with early, presymptomatic disease and sufficiently HLA-matched allografts face favorable estimates of long-term functional survival following HSCT.

P908 | A single center five-year experience blood transfusion in the peri-transplantation period of autologous peripheral hematopoietic stem cell transplantation

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Background: Individuals undergoing hematopoietic stem cell transplant may need numerous blood transfusions, a situation that could be complicated by incidents of adverse transfusion reactions (ATR).

Aims: This study aimed to examine the utilization of blood components and the occurrence of ATR during the peri-transplantation phase of autologous peripheral blood stem cell transplant (APBSCT) at Hospital Universiti Sains Malaysia (USM).

Methods: A retrospective analysis was carried out on all lymphoproliferative disorders (LPD) adult patients who underwent (APBSCT) and received blood transfusions over a five-year period from January 2019 to December 2023 in Hospital USM. Secondary

data, encompassing demographic details, patient diagnoses, the types of blood products administered, and adverse transfusion reactions, were obtained from the MyTransfusi Laboratory Information System.

Results: APBSCT was conducted on 55 LPD adults encompassing 32 men and 24 women included Hodgkin lymphoma diffuse large B-cell lymphoma, and multiple myeloma, constituting 42%, 25%, and 24% of cases, respectively. A total of 415 units of blood components were administered comprised 47 units (11%) of crossmatched compatible packed red blood cells, 354 units (86%) of platelet concentrates, and 14 units (3.37%) of platelet apheresis, specifically attributed to hemoglobin levels less than 8 g/dL with platelet counts less than $10 \times 10^9/L$. Remarkably, only 5 cases (1.2%) of ATR were recorded, all of which were mild allergic reactions linked to platelet transfusions.

Summary / Conclusions: The audit revealed that PRC and platelets are frequently administered blood components in these patients. This observation aligns with existing literature, indicating that anemia and thrombocytopenia are the predominant hematological alterations during transplantation. The occurrence of ATR is lower than that reported in other studies, and it could be attributed to factors such as specific interventions or precautions during transfusions, or the absence of a hemovigilance system.

P909 | Analysis of the early and transient cytopenias in the post-CAR-T period—a single center experience

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Background: CAR-T cell therapy emerged as a promising treatment, but hematotoxicity is an adverse reaction associated. Its comprehension is still a challenge. CAR-HEMATOTOX (HT) score has arisen as a tool to help predict the risk of hematotoxicity. Knowing the risk of complications can be crucial in patients' management.

Aims: Determine the influence of HT score on clinical outcomes (early and transient cytopenias post-CART) in patients receiving CD19 CAR-T therapy with axicabtagene ciloleucel (axi-cel) in our center.

Methods: Retrospective study of patients who have received axi-cel in Canarias between 2020 and 2023. 33 patients underwent autologous lymphoapheresis. Table 1 summarizes baseline patient features. Median prior therapies: 2.8. 27/33 (81.8%) infused (5 deceased pre-infusion (4 progression / 1 COVID), 1 pending). 21/27 needed bridging therapy. All ECOG ≤ 2 , received lymphodepletion (fludarabine/cyclophosphamide). HT score calculated pre-lymphodepletion. Patients classified high vs. low risk. 9 excluded due to loss of follow-up. Early cytopenias (severe neutropenia/thrombocytopenia) at days

P909 - Table 1

Patient's center:

Hptal Dr Negrín: 6
Others: 27

Median age (range)

52 (27 – 74)

Gender (M/F)

(%): (19/14) (57% / 43%)

Diagnosis

Diffuse large B-cell lymph: 22
Transformed follicular lymph: 3
Primary mediastinal B-cell lymph: 1
High-grade B-cell lymph: 6
Follicular lymph* (*compassive use): 1

ECOG status pre-infusion

0: 14

1: 11

2: 2

>2: 0

Hematotox score pre- lymphodepletion

Low HT score (0-1): 22

High HT score (> 0 = 2): 5

P909 - Table 2 shows patients classified into HT score and cytopenia's degree on days +30 and +60 post-CAR-T.

		Low HT	High HT	All
Neutrophils (+30)	<500 u/mcL	1 (10%)	1 (12.5%)	2 (11.1%)
	>500 u/mcL	9 (90%)	7 (87.5%)	16 (88.9%)
Platelets (+30)	<20.000 u/mcL	1 (10%)	2 (25%)	2 (11.1%)
	>20.000 u/mcL	9 (90%)	6 (75%)	16 (88.9%)
Neutrophils (+60)	<500 u/mcL	2 (20%)	2 (25%)	4 (22.2%)
	>500 u/mcL	8 (80%)	6 (75%)	14 (77.8%)
Platelets (+60)	<20.000 u/mcL	1 (10%)	2 (25%)	3 (16.7%)
	>20.000 u/mcL	9 (90%)	6 (75%)	15 (83.3%)

No statistical correlation between HT score and neutrophils' and platelets' recovery on days +30 and +60 was found.

+30 and +60 post-infusion. χ^2 Test analyzed HT score correlation with post-CART severe cytopenia probability.

Results: Before infusion, 15/27 (55.6%) had refractory disease, 7/27 (25.9%) partial response (PR), and 5/27 (18.5%) complete remission (CR). No infusion-related reactions occurred. All experienced cytokine-releasing syndrome (15: grade 1, 11: grade 2, 1: grade 3), resolved with tocilizumab (19) and/or steroids (7). Neurotoxicity was observed in 15/27 (55.6%) (8: grade 1, 3: grade 2, 4: grade 3), all returning to baseline neurological status with dexamethasone or methylprednisolone. Average hospital stay was 23 days (range 17-35). At +30, disease re-evaluation showed: 18/27 CR, 6/27 progression, 3/27 PR. Post-CART, 10/27 patients died (8/10: progression; 2/10: other causes). 17/27 patients survived (1-36 months): 12/17 CR; 1/17 PR; 4/17 relapse. Only

2 patients completed 18-month follow-up for CART's 2nd payment submission (based on Spanish Ministry of Health's financial criteria).

Summary / Conclusions: Due to the limited number of patients in our sample subject to analysis, no statistical correlation between HT score and risk of cytopenia post-CART has been observed. Thus, prospective studies with more patients and longer follow-up need to be performed.

P910 | Efficacy of local autologous platelet-rich plasma prepared by single spin and double spin method in the treatment of chronic non healing ulcers

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Background: Chronic ulcers present a significant hurdle to health and quality of life, necessitating effective solutions. Autologous platelet-rich plasma (PRP) therapy offers a promising avenue, being both straightforward and minimally invasive. This method harnesses the body's own growth factors, found in concentrated form within the patient's blood, to stimulate tissue regeneration. However, the efficacy of PRP depends on various factors, such as donor platelet count, activation techniques, and centrifugation methods. Such variability underscores the importance of standardized procedures to maximize therapeutic outcomes and address the pressing challenge of chronic ulcer healing, ultimately improving patients' well-being. This study was to assess the efficacy of

autologous PRP prepared by single spin method and double spin method in the management of non-healing chronic ulcers.

Aims: To evaluate the clinical outcome of nonhealing chronic ulcers treated with platelet-rich plasma prepared by single spin method and double spin method.

Methods: This prospective randomized controlled trial was conducted between December 2020 and March 2022. Patients, aged 18-85, with ulcers causing from conditions like diabetes, pressure, or leprosy, were randomly assigned to two groups: one receiving PRP prepared via the single spin method (SSM), and the other via the double spin method (DSM). Patient's on immunosuppressive and anti-coagulant medications, with low platelet counts ($<1.5 \times 10^3/\mu\text{L}$), bleeding disorders, or severe cardiovascular conditions with aspirin therapy were excluded. Detailed patient histories were recorded, and 100 mL of blood was drawn into 14 mL CPDA-containing bags. In the SSM group, blood underwent centrifugation at 3400 rpm for 10 min. For the DSM group, the supernatant underwent a subsequent centrifugation at 1800 rpm for 9 min. Fibrin gel was then prepared by mixing the patient's serum (1:5) with the PRP and 10% calcium gluconate (1:2). Assessment tools, including surface area (in cm^2), ulcer volume (in mL), percentage of healed area, and healing rate (cm^2/week), were utilized on days 1, 5, 15, 20, and 30 post-interventions to evaluate therapeutic effectiveness. The results were compared by Wilcoxon rank sum test.

Results: - Out of 31 ulcers cases, 13 underwent the single spin method (SSM), and 18 the double spin method (DSM). Predominant age groups were 41-50 and 51-60 years. Table 1 details surface area reduction and ulcer volume in both groups.

Summary / Conclusions: Our study underscores PRP's efficacy in treating non-healing chronic ulcers. Notably, PRP prepared via the single spin method exhibited superior effectiveness compared to the double spin method. Nevertheless, further validation through long-term investigations with larger sample sizes is imperative for consolidating these findings.

P910 - Table 1.

Groups	Double spin (n = 18)	Single spin (n = 13)	p-value
Surface area (cm sq)			
First sitting (Day 1) (Mean \pm SD)	16.27 (11.94)	9.87 (7.73)	0.049
Second sitting (Day 5) (Mean \pm SD)	16.13 (11.82)	9.66 (7.57)	0.043
Third sitting (Day 15) (Mean \pm SD)	15.23 (11.45)	8.36 (6.56)	0.025
Fourth sitting (Day 20) (Mean \pm SD)	15.23 (11.45)	8.32 (6.52)	0.022
Final sitting (Day 30) (Mean \pm SD)	14.76 (11.54)	7.65 (6.23)	0.020
Volume (mL)			
First sitting (Day 1) (Mean \pm SD)	29.10 (34.15)	14.72 (17.39)	0.047
Second sitting (Day 5) (Mean \pm SD)	28.41 (34.22)	13.88 (16.49)	0.035
Third sitting (Day 15) (Mean \pm SD)	25.79 (32.11)	10.42 (12.72)	0.017
Fourth sitting (Day 20) (Mean \pm SD)	25.79 (32.11)	10.15 (12.65)	0.012
Final sitting (Day 30) (Mean \pm SD)	24.38 (32.35)	8.78 (11.07)	0.005

Clinical immunogenetics—HLA in transfusion medicine

P911 | Donor increase without specific HLA antigens for patients with severe platelet immune refractoriness after rituximab treatment

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Background: Immune refractoriness platelet (IRP) is a challenge for transfusion therapy, especially in patients requiring hematopoietic stem cell transplantation (HSCT). IRP is caused by the destruction of transfused platelets from donors with antigen-specific antibodies (AAD) for antibodies against platelet antigens: HLA or HPA previously formed by the patient. Therefore, platelet transfusion in patients with IRP should be performed with platelets from donors without antigens specific (DWAS) for the antibodies that the patient

presents, which is obtained from a registry of platelet donors typed for HLA or HPA antigens. However, when a patient with IRP is highly alloimmunized it is difficult to find DWAS, and it is necessary to look for alternatives that allow effective transfusion therapy, one of them being the use of the monoclonal antibody Rituximab. Patients with aplastic anemia and myelodysplastic syndrome with IRP, who received 3 weekly infusions of Rituximab (375 mg/m²) and had improvements in the response to post-treatment platelet transfusion, have been reported, being an alternative that can be considered in severe IRP.

Aims: To report the management with drugs and transfusions in a patient with thrombocytopenia and severe immune platelet refractoriness.

Methods: Case report: 33-year-old patient with acute lymphocytic leukemia, candidate for HSCT, with IRP and high requirement of platelet transfusions. In the first HLA antibody study performed, 48 different specificities of HLA class I antibodies were identified. The search for DWAS for the 48 antibody specificities was performed in the database of HLA-typed platelet donors of the Clínica Santa María Blood Bank. Of the 300 platelet donors belonging to the database only 1 did not have the specific antigens for the 48 antibodies identified. The patient was administered 4 doses of 375 mg/m² of Rituximab post chemotherapy and prior to HSCT on days 1, 8, 15 and 22. A second antibody study was performed 10 days after the last dose. Of the 48 specificities found in the first study, 18 were not detected; of the 30 specificities that were detected in the first and second study, 28 decreased their mean fluorescence intensity (MFI), in 10 of which the MFI was < 1000. The number of DWAS found increased from 1 to 9. In addition, considering donors antigen specific (DAS) for antibodies with an MFI < 1000, the list increased to 20 platelet donors, substantially increasing the possibility of transfusing platelets from DWAS or with DAS for antibodies with low MFI.

Results: Of the 48 specificities found in the first study, 18 were not detected in the second study; of the 30 specificities that were detected in the first and second study, 28 decreased their MFI (mean fluorescence intensity), in 10 of which the MFI was < 1000. The number of DSAEs found increased from 1 to 9. In addition, considering DAEs for antibodies with an MFI < 1000, the list increased to 20 platelet donors, substantially increasing the possibility of transfusing platelets from DWAS or with DAS for antibodies with low MFI.

Summary / Conclusions: The administration of Rituximab in a patient with severe IPR increased the number of DWAS, essential for effective transfusion therapy.

P912 | Analysis the association between HLA Class II allelic polymorphisms with the possible risk of developing of RBC alloantibodies in Taiwanese

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Background: HLA class II molecules have been known to play a key role in red blood cell (RBC) alloimmunization. Previous studies have reported association between RBC alloantibody formation and HLA-DRB1 alleles among different populations, such as HLA-DRB1*15 in Caucasian group. We hypothesized that HLA class II restriction could play a role in the overrepresentation of anti-“Mi^a” and anti-E alloantibodies in Taiwanese population.

Aims: The distribution of HLA class II allele frequencies varies among different ethnic populations. In this retrospective study, we aimed to investigate the association between RBC alloantibodies and HLA class II phenotypes among Taiwanese population.

Methods: From blood bank database of National Taiwan University Hospital (NTUH), 7471 individuals with RBC alloantibodies were included. We conducted a retrospective case-control study to compare the HLA class II allele frequencies among 323 RBC alloimmunized patients with those among 7648 unrelated individuals with routinely performed HLA typing in NTUH using Pearson's Chi-square test. The odds ratio (OR) with 95% confidence interval (CI) and the *p* value were calculated. Corrected *p* value, *pc* value, less than 0.05 was considered to be statistically significant.

Results: Anti-“Mi^a” and anti-E antibodies were the predominant RBC alloantibodies in our study population, both occurring in 47.54% and 33.38% of alloimmunized individuals. Then, 200 (61.92%) of the 323 immunized individuals had single antibody specificity, and the other 123 (38.08%) of whom had multiple specificities. Anti-“Mi^a” was found in 49% of single responders, and anti-c, -E was presented by 45.5% of multiple responders. The allele frequencies of HLA-DRB1*04, HLA-DRB1*09 and HLA-DQB1*03:02 in immunized group were significantly higher than those in control group, giving odds ratios of 1.49, 1.38 and 1.79, respectively. Significant positive associations were found on HLA-DRB1*04 and HLA-DQB1*03:02 among single responders (vs control). No significant difference was observed between these two subgroups (single vs multiple responders). HLA-DRB1*04 (24.4% vs 14.9%, OR = 1.84, *Pc* = 0.001) and HLA-DQB1*03:02 (14.2% vs 7.7%, OR = 1.98, *Pc* = 0.003) were positively associated with the presence of anti-“Mi^a” antibody. Both HLA-DRB1*09 (22.2% vs 14.4%, OR = 1.70, *p* < 0.001) and HLA-DQB1*03:03 (22.6% vs 14.8%, OR = 1.68, *p* < 0.001) alleles showed a significant increased frequency among E-immunized patients.

Summary / Conclusions: In conclusion, both HLA-DRB1*09 and HLA-DQB1*03:03 alleles showed a significant increased frequency among E-immunized patients. HLA-DRB1*04 and HLA-DQB1*03:02 were positively associated with the presence of anti-“Mi^a” antibody. These results support that HLA restriction plays an important role in RBC alloimmunization and could be helpful for evaluating the risk of RBC alloimmunization of transfusion patients and incorporated into prophylactic red cell antigen matching strategy based on HLA genotypes of patients in the future.

P913 | Study on 5' untranslated region of the HLA-A gene contributes to the variation in HLA-A mRNA expression levels

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Background: The human leukocyte antigen (HLA) system plays an important role in both the adaptive and innate immune responses and displays extensive polymorphism. The mRNA expression of alleles in different HLA-A locus is different, which has important biological effects on the occurrence and development of certain diseases. The variation of HLA-A mRNA expression level is closely related to the presence of specific polymorphisms in the 5' untranslated region (5'UTR).

Aims: In this study, we have examined the expression levels of HLA-A allele in the Zhejiang Han population using a real time polymerase chain reaction assay, and the 5'UTR of HLA-A gene was sequenced by polymerase chain reaction sequence-based typing (PCR-SBT) method.

Methods: 45 HLA-A homozygous individuals were included from the voluntary blood donors in Blood Center of Zhejiang with informed consent. The 1700bp length of 5'UTR was amplified and sequenced using primers 5'UTR-F (AATTGAGCCCCCTCCAGTT) and 5'UTR-R (AGGMGGGCCCTTGCTT). HLA-A and the housekeeping gene B2M were amplified with SYBR[®] Green qPCR method using cDNA. The HLA-A expression level was normalized to B2M and calculated using the $2^{-\Delta\Delta C_t}$ method.

Results: The genotypes of the 45 HLA-A homozygous samples are HLA-A*02 ($n = 15$), HLA-A*11 ($n = 18$), HLA-A*24 ($n = 9$), HLA-A*33 ($n = 3$). The median expression of HLA-A*02 was higher (0.36; range 0.12-1.04) than the HLA-A*11 (0.13; range 0.02-0.43; Mann Whitney, $p < 0.0001$, two-tailed). The median expression of HLA-A*24 and HLA-A*33 were 0.32 and 0.24, respectively. The 5'UTR of HLA-A showed single nucleotide polymorphisms (SNPs) between different alleles, which may induce change in potential binding sites for regulatory elements and affect the expression of a reporter gene. The -1261A (rs9260080) in the HLA-A*11 allele led to the disappearance of the binding sites for forkhead box protein 1 (FOXP1) and created a new binding motif for ETS-related gene (ERG). Besides, the sequence around position -992 (rs9260084) is predicted to contain a binding site for GATA1 in the HLA-A*02 allele (high expression) by AnimalTFDB. The HLA-A*11 allele (low expression) presented a SNP (G>A) responsible for the loss of this binding site and the creation of a new binding motif for progesterone receptor (PGR).

Summary / Conclusions: 5'UTR contributes to the differential expression of specific HLA-A alleles by altering transcription factor binding sites which needs further research.

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P914 | Abstract withdrawn

P915 | The characteristic of 11 HLA loci in Zhejiang Han population

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Background: Human Leucocyte Antigen (HLA) molecules express on the surface of various cells, associated with the outcome of a wide range of diseases. The Zhejiang Han community comprises approximately 60 million individuals, constituting a substantial 99.15% of the province's total population. Studying the diverse HLA-A, -B, -C, -DRB1, -DRB3/4/5, -DQA1, -DQB1, -DPA1, and -DPB1 alleles within this population will contribute valuable insights into an expanded HLA diversity within the Chinese Han population.

Aims: In this study, we conducted HLA genotyping for 813 voluntary umbilical cord blood donors from the Zhejiang Han population, targeting 11 HLA loci using the next-generation sequencing method. HLA genotype was assigned using the HLA TypeStream Visual Software version 2.0 (TSV 2.0). The frequencies of HLA alleles, as well as the composition and frequencies of HLA haplotypes, were assessed using the Arlequin 3.5.2.2. The Chinese HLA common and well-documented (CWD) principle was used to assign the G group alleles or the ambiguous HLA allele combination.

Methods: All specimens were genotyped for HLA-A, -B, -C, -DRB1, -DRB3/4/5, -DQA1, -DQB1, -DPA1, and -DPB1 loci respectively using the AllTypeTM NGS kit (One Lambda Inc, Canoga Park, CA, USA) according to manufacturer's instruction.

Results: Our analysis of the alleles and haplotypes revealed a high degree of polymorphism within these loci. A total of 289 unique HLA alleles were identified, with the HLA-B locus exhibiting the most significant diversity, while HLA-DRB4 displayed the lowest variation. Due to the inherent limitations of the sequencing method, some unresolvable alleles in the specific loci, such as HLA-DRB1, -DPA1, and -DPB1, were assigned as G group designation. In our comprehensive analysis across all 11 HLA loci, a total of 1204 haplotypes were estimated. The distribution of these alleles were similar to those of the Chinese Southern Han population while highly different from the Caucasian population.

Summary / Conclusions: our study represents the first comprehensive report on the characteristics of 11 HLA loci, encompassing alleles and haplotypes of HLA-A, -B, -C, -DRB1, -DRB345, -DQA1, -DQB1, -DPA1 and -DPB1 at two-field resolution level within the Zhejiang province of China. Our data highlights the diversity distribution of HLA alleles across different ethnicities while revealing a close resemblance among East Asian populations.

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Clinical immunogenetics— histocompatibility in stem cell transplantation

P916 | Establishment of full-length sequencing method for HLA loci using nanopore technology

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Background: Human leukocyte antigen (HLA) matching tests of patients and donors are critical for hematopoietic stem cell transplantation (HSCT). However, the HLA typing ambiguities are increasing in laboratory due to the short read lengths of the next-generation sequencing (NGS) platforms and increasing number of HLA alleles. Nowadays, nanopore sequencing technology, a technology of single-molecule, real-time and long-read length sequencing, was more and more used in the characterization of HLA as the increasing accuracy of nanopore sequencing these years.

Aims: To establish full-length sequencing method for HLA loci based on nanopore technology, which can resolve the ambiguity of HLA typing generated by NGS and PCR-SBT method and obtain more accurate HLA typing results.

Methods: We designed the gene specific primers for the HLA-A, -B, -C, -DRB1, -DRB3/4/5, -DQA1, -DQB1, -DPA1 and -DPB1 loci separately according to the IPD-IMGT/HLA gene database. And all the HLA loci were separately amplified from the 5' -UTR to the 3' -UTR, except for the HLA-DRB5 locus from the intron 1 to the 3' -UTR. The purified PCR production of 11 HLA loci were pooled equimolar in one, and ligated to Oxford Nanopore Technology (ONT) native barcodes, then pooled equimolar again. After ligated to ONT Adaptor Mix, the production was loaded onto an Spot-ON Flow Cell on the MinION MK1C for sequencing. The resulting data was converted to FASTQ files and assigned using NGSengine-Turbo software (GENDX). In this study, a total of 50 samples with HLA typing ambiguous generated by NGS were collected during 2022–2023, and were identified in our laboratory using the new full-length sequencing method.

Results: The full-length sequencing method for HLA loci based on ONT were established successfully. According to analysis of the NGSengine-Turbo software, the average reading depth of the HLA-DRB1 was above 300, and the other HLA loci were above 600. Among the collected 50 samples, there were 51 different common ambiguities with the NGS platform, including 2, 5, 2, 5, 1, 1, 1, 3, 7, 1 and 23 kinds of common ambiguities in HLA-A, -B, -C, -DRB1, -DRB3, -DRB4, -DRB5, -DQA1, -DQB1, -DPA1 and -DPB1 loci respectively. The HLA typing results in two-field high resolution generated by the new method in the 50 samples were 100% conference to those by NGS, and the HLA typing results in three-field high resolution were clearly and unique, except for the HLA-DRB5 locus. That is, we identified 50 different common ambiguities based on the new method, and

only one allele ambiguities HLA-DRB5*01:02/01:126 was not resolved because the exon 1 of HLA-DRB5 was not sequenced in the study.

Summary / Conclusions: We established a full-length sequencing method for HLA loci based on ONT, and offers a promising solution to resolve the ambiguities generated by NGS and PCR-SBT methods in HLA typing.

P917 | Analysis of chimeracy after transplantation of allogenic hemopoietic stem cells

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Background: Transplantation of allogeneic hematopoietic stem cells is one of the effective methods of treating patients with various onco-hematological diseases. Chimerism monitoring allows one to assess not only graft engraftment, but also potentially predict the risk of developing primary/secondary graft failure, the likelihood of disease relapse and graft-versus-host disease.

Aims: Analysis of the influence of the compatibility degree between the donor and recipient according to the HLA phenotype on the result of graft engraftment in patients with AML (acute myeloid leukemia) and ALL (acute lymphoblastic leukemia).

Methods: 254 donor-recipient pairs were studied, of which 142 pairs were diagnosed with AML and 112 pairs were diagnosed with ALL. Couples with 100% HLA phenotype compatibility were included in group A ($n = 75$), with 50% compatibility in group B ($n = 67$) for AML and in group A ($n = 60$), in group B ($n = 52$) for ALL. All recipients and donors were typed by HLA-A, B, C, DRB1, DQB1 at the allele level. To determine chimerism, peripheral blood samples were collected on specific days. Chimerism determination was performed using the quantitative fluorescence polymerase chain reaction method with the commercial AmpFLSTR™ Identifiler™ Plus kit (Thermo Fisher Scientific). Separation and detection of PCR products were performed on a 3500XL Genetic Analyzer (Thermo Fisher Scientific). ChimerMarker 3.1.0 software (Softgenetics) was used to interpret the results.

Results: The results of donor chimerism with AML nosology in group A are significantly better compared to group B. Thus, on the 60th day, the average result of chimerism was 93% in group A and 86% in group B, on the 100th day—95% and 91%, respectively, on the 520th day—97% and 95%, respectively. At the same time, on the 20th day the result of chimerism in group A looked lower in comparison with group B—85% and 89%, respectively, on the 240th day 83% and 89%, respectively. The results of donor chimerism among patients with ALL nosology are as follows: the best results for group A were the 60th (96% in group A and 89% in group B) and 300th days (99% and 79%, respectively). The worst results were on days 100 (91%/95%), days 520 (84%/96%), and days 1080 (93%/99%). Among patients

diagnosed with AML, the proportion of patients with mixed chimerism was 4.6% in group A and 22.3% in group B; complete chimerism was achieved in group A in 85% and in group B in 73.1% of patients. The proportion of patients who did not achieve engraftment was 2.6% in group A and 4.4% in group B. Among patients diagnosed with ALL, mixed chimerism was detected in group A in 23.3% of patients and in group B in 15.6%. Complete chimerism was achieved in group A in 75% of patients and in group B in 82%. Patients who did not achieve engraftment in group A were 1.6% and group B were 3.9% of the total number of patients in each group.

Summary / Conclusions: The results obtained show that the engraftment effect in patients with full HLA gene compatibility is better in patients diagnosed with AML. To fully understand the impact of HLA antigen compatibility percentage on graft engraftment, additional studies must be conducted taking into account the age, gender of the donor and obstetric history of the recipient, as well as the antibody status of the recipient.

Clinical immunogenetics— histocompatibility in organ transplantation

**P918 | Pre-existing HLA-DP donor-specific antibodies
associated with poor renal transplant outcomes**

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Background: Anti-DP antibodies have been considered less significant for renal transplantation. Limited literature exists on the role of anti-DP antibodies in kidney transplants.

Aims: Both pre-formed and de novo anti-HLA-DP antibodies appear to be common, especially in historic kidney transplants. This case is based on a probable case of anti-HLA-DP antibody-mediated renal transplant rejection.

Methods: A 31-year-old male with end-stage renal disease secondary to hypertensive nephropathy received a first kidney transplant from a live donor (Father) in 2008. They share the same blood group. Recipient and donor Human Leucocyte Antigen (HLA) typing was carried out at the low-resolution level. HLA mismatch for the A, B, and DR loci was 0-1-0, respectively. HLA-DP typing is not routinely

performed in kidney transplant recipients, and any DP mismatch between the donor and the recipient was unknown at the time of transplantation. Complement-dependent cytotoxicity crossmatch (CDC-XM) was compatible for both T and B cells. At 12 years post-kidney transplant, the grafted kidney function deteriorated. HLA Panel Reactive Antibody (PRA) level was done by Luminex assay and showed no significant DSA except Positive Class II (20%) with the specificity of DP (1312-527 MFI). Anti MHC class I polypeptide-related sequence-A (MIC-A) was negative. The patient was placed on dialysis due to rising serum creatinine. Renal biopsy revealed T cell-mediated chronic rejection. In the second transplantation HLA workup, the HLA mismatch for the A, B, DR loci was 0-0-1 respectively. HLA PRA level was tested by Luminex assay and was negative for class I and 17% for class II with the specificity of DP (584-220 MFI). MIC-A was negative. Subsequent HLA PRA was negative for both class I & II with anti DP2-1178 MFI. Pre-transplant CDC-XM was Compatible. As the patient had a historical peak level of anti-HLA DP antibodies, it was suggested to do prospective donor's DP typing. However, the patient received the second kidney transplant from that prospective donor of the same blood group before the DP typing. Post kidney transplant Day 7, Graft doppler was suggestive of early rejection and Acute Kidney Injury (AKI). An allograft kidney biopsy revealed borderline T-cell-mediated rejection with features suggestive of active antibody-mediated rejection (AMR). Later C4d was positive in vessel walls. The patient was managed with 5 cycles of TPE and IVIG 5 doses.

Results: Analysis of the Single Antigen Bead (SAB) reactions together with the HLA molecular typing results revealed that the patient had IgG-DSAs against DPA1*01:03-DPB1*02:01. MFI of 10916 in a historic booking screen sample and 1178 in a pre-2nd KT sample. Post 2nd KT rejection sample had an extremely low reactive antibody with MFI of 201.

Summary / Conclusions: Extremely low Mean Fluorescence Intensity (MFI) of anti-HLA DP antibody could be due to possible graft absorption-sponge effects, immunosuppression/intra laboratory variability of MFI. Anti-HLA-DP antibodies have strong potential to cause active rejection & chronic rejection and are refractory to classical desensitization attempts. Preformed anti-HLA-DP DSA are as deleterious as other DSA.

P918 - Table 1.

Locus	HLA-A*	HLA-B*	HLA-DRB1*	HLA-DPA1-DPB1*
Candidate Typing	24	7.40	14.15	DPA1*01-DPB1*01, DPA1*01-DPB1*03
Immuniser Typing	24	7.15	14.15	DPA1*01-DPB1*02, DPA1*01-DPB1*03
Donor Typing	11.31	40.51	10.15	DPA1*01-DPB1*02, DPA1*01-DPB1*04

P919 | Case report—acute antibody-mediated rejection of transplanted kidney due to anti-HLA-Cw 10 antibody in Sri Lanka

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Background: Kidney transplantation (KT) is often complicated by the presence of donor-specific antibodies (DSA) targeting human leukocyte antigens (HLA). DSA against HLA-A, HLA-B, HLA-DR, and HLA-DQ are well-documented contributors to post-transplant rejection but the role of HLA-Cw antigens remains less explored. HLA-C antigens have received less attention due to lower cell surface expression compared to other HLA loci. Recent evidence suggests that anti-HLA-Cw antibodies can induce an immune response, albeit at reduced levels when compared to HLA-A and HLA-B. But Sri Lankan kidney transplant programs, like many others worldwide, tend to disregard the significance of HLA-Cw in acute antibody-mediated rejection (aABMR) due to low prevalence and cost effectiveness.

Aims: This case report presents a rare occurrence of aABMR caused by anti-HLA-Cw antibodies in a kidney transplant recipient in Sri Lanka.

Methods: A 50-year-old male with focal segmental glomerulosclerosis and end-stage renal disease was scheduled for live donor KT. Patient was O Rh D positive, while the prospective donor (elder sister) was O Rh D negative. HLA work up was performed according to the local transplant protocols and donor was accepted. Pretransplant immunosuppressants, induction therapy and hemodialysis were done prior to KT. Since post-transplant day one, serum creatinine levels were elevated and urine output was reduced, raising suspicion of aABMR. Renal biopsy confirmed active antibody-mediated rejection, with positive C4d immunostaining. Samples were sent to the HLA reference laboratory for Panel Reactive Antibody (PRA) testing. Therapeutic Plasma Exchange (TPE) was initiated and total of 5 TPE cycles were completed, along with 6 doses of intravenous immunoglobulin.

Results: In pretransplant workup, HLA typing of donor and recipient mismatch was 0:0:1 respectively in HLA A, B and DR loci. No DSA detected against donor in booking PRA screening. HLA Class I single antigen reactive specificity was 8% with possible anti HLA-Cw10 and several other HLA antibodies (9149-1267 MFI). The complement-dependent microlymphocytotoxicity assay (CDC XM) indicated compatible T cell and B cell crossmatch. Extended donor typing was recommended for further histocompatibility confirmation. The donor and recipient had a mismatch in the HLA-C locus (HLA -C01,02 in patient and HLA-C01, 03 in donor). Most possible allele code for HLA-C03 was HLA-C*03:02. Post-KT PRA and single antigen bead assays revealed the presence of Anti-HLA-Cw10 antibody (1988 MFI), which corresponds to a split antigen of HLA-C03. Anti-MHC class I polypeptide-related sequence-A (Anti-MIC-A) and non-HLA antibody screening for Angiotensin II Type 1 Receptor antibodies (Anti-AT1R) were negative.

Summary / Conclusions: In this case, the patient's sole sensitization event resulted from a blood transfusion. Although no donor-specific antibodies (DSAs) were found in the HLA Class II, anti-HLA-Cw10 antibodies were detected, albeit with low mean fluorescence intensity (MFI) values. After excluding other potential causes (AT1R and MIC-A), the most likely reason for aABMR is attributed to the Anti-HLA-Cw10. These findings underscore the importance of considering HLA-C antigens more broadly in transplantation management. Understanding the impact of HLA-Cw antibodies in aABMR is crucial for optimizing transplant outcomes and refining global transplant allocation strategies.

P920 | Virtual cross match for HLA antibodies—role in deceased donor renal transplant testing and organ allocation

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Background: It is important to strengthen deceased donor organ transplant program to help the patients who do not have living kidney donor available. But there are lot of challenges in the program & one of the main challenge is correct xm & organ allocation. Luminex anti-HLA antibody detection assay is more sensitive & specific than Complement Dependent Cytotoxicity (CDC) or flow cytometric xm. This technique is solid-phase assay in which purified HLA molecules (single HLA type or from donors cells) are attached to beads. These molecules will bind to anti-HLA antibodies in the patient's serum. Advantages of the Luminex methodology are the speed, high through put, less subjectivity, more sensitivity and specificity of results. In India there is no standard testing protocol followed by all transplant centers. We have a well structured deceased donor transplant program and nearly 100 donations take place every year. Organ used are kidney, liver, heart and sometimes lungs. Karnataka State Registry & transplant coordination committee "Jeeva-Sathakathe/JSK" selects the patients for cross match & allocate organs mainly on the basis of CDC. For patients who are sensitized Single Antigen Bead assay was done and Virtual cross match is performed when donor is available.

Aims: To evaluate the results of virtual cross match for Deceased Donor renal transplant cases & correlate with graft dysfunction.

Methods: Patients who had history of sensitization events, Single Antigen Bead assay (Life Codes, Immucor, Stamford, CT 06902, United States) is done & the antibodies assigned. Whenever donor is available Luminex SSO HLA typing (Life Codes, Immucor, Stamford, CT 06902, United States) of done on the donors and virtual cross is performed as per the request of transplant center along with CDC xm. All the results are sent to JSK and organs are allocated on the basis on XM results as well as waiting time.

Results: In the years 2022-23 total 172 deceased donations were accepted. At HLA Lab we received total 562 potential kidney recipient samples (Male 421 & Female 217) for xm. We received request for virtual xm for 33 patients who were sensitized & their Single Antigen test

was done. It was done using Life code Single Antigen Bead Assay and unacceptable antigens were assigned. After HLA typing by SSOP (Loci A, B DR & DQ) virtual cross match was done. We found 23 virtual cross positive with patients have donor specific antibodies (MFI range: 1152-12666). Out of 23, we found only 2 patients had CDC cross match positive & remaining 21 negative. Total nine patients had virtual cross match negative & CDC was also found negative. Six patients underwent transplant & the post-transplant outcome was satisfactory.

Summary / Conclusions: Detection of anti-HLA antibodies prior to kidney transplantation is an evolving science. The CDC xm is considered classical & gold standard test for detection of pre transplant HLA antibodies. This test has low sensitivity and it is time & labor intensive method and especially challenging for deceased donor transplant cases. Therefore virtual xm is performed at many centers. In present study we found virtual xm very useful for organ allocation in highly sensitized patients.

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