



Immunohematology Case Studies 2017 - 5

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Clinical History



47 year male donor, who donated in voluntary blood donation camp. Repeat donor who had donated 4 times in the past with last donation 1 year ago.

Sample of the donor received in immunohematology lab for blood group discrepancy – Preliminary testing by technical person by tube method reported as RBCs grouping as O positive and serum grouping as A

Anti- A	Anti- B	Anti- D	Anti-AB	A cell	B cell	O cell	Auto control
0	0	4+	0	0	4+	0	0

Serologic History



Blood grouping both CELL and SERUM done by tube method as per departmental SOP.

ABO/Rh:

Anti- A	Anti- B	Anti- D	Anti-AB	A cell	B cell	O cell	Auto control
Weak+ (delayed)	0	4+	Weak + (delayed)	0	4+	0	0

As cell and serum grouping results don't match it is a blood group discrepancy

Serologic History (continued)



Extended blood grouping done by tube method

Anti-AB	Anti-A1	Anti-H
Weak+ (delayed)	0	4+

Serum grouping	Immediate Spin (IS)	Room Temperature (RT)	Anti Human Globulin (AHG)
A cell	0	0	0
B cell	4+	4+	4+
O cell	0	0	0
Auto control	0	0	0

Preliminary Blood group: A_{weak} RhD positive

Current Sample Presentation Data



DAT: Negative by gel technology (Bio-Rad)

Antibody Screen Method: Bio-Rad 3 cell panel

Antibody Screen Results: Negative

SCREEN I	SCREEN II	SCREEN III	AUTO	DAT
0	0	0	0	0

Challenge with the Current Presentation



There are a few challenges in this current presentation which need to be solved:

1. Cell and serum blood groupings do not match: Blood group in initial testing is O RhD Positive and in extended tests is A_{weak} RhD Positive.

Possibility of A_{weak} subtype that needs to be confirmed.

2. Group discrepancy detected on detailed testing from tube sample needs to be confirmed from bag sample to rule out any technical error during taking donation from the donor.

Possible Answers and Next Steps



A_{weak} RhD positive blood group of donor needs to be confirmed by:

1. Cold adsorption technique using anti-A from pooled serum of B donors followed by heat elution, for confirmation of A_{weak} phenotype.
2. Blood group extended testing and cold adsorption to be performed from sample obtained from donor bag (from bag tube segment post stripping) as well for final confirmation of donor blood group.

Further Work



Cold adsorption and Heat Elution techniques to be performed on Donor tube Sample:

Washed Donor Cells



Cold Adsorption of washed cells with anti-A from pooled type B donor serum at 4°C for 90mins

Washed 5 to 6 times



Supernatant of last wash preserved



Packed cells mixed with equal volume of 4% Bovine serum



albumin



Further Work (Continued)



Heat elution of the mixture done at 56⁰C for 10 mins
in a water bath with intermittent shaking



Centrifuged at 3000 rpm for 3 mins and
supernatant (Eluate) obtained



Eluate subjected to further testing with ABO
pooled reagent cells as follows:



Further Work - Results



Plain Card	Eluate	Last Wash
A cell	3+	0
B cell	0	0
O cell	0	0

Result of cold adsorption and heat elution steps:
Positive reactivity of A cells with eluate confirms presence of adsorbed anti-A in the eluate obtained after heat elution of cold adsorbed donor cells thus confirming A_{weak} status of donor cells.

Updated Clinical Information



1. Telephone contact with the donor – saying thank you for blood donation and explaining him the problem being faced in confirmation of his blood group.
2. Apologies for delay in dispatching of his donor card.
3. Take history of any previous donation if any, and any previous documentation of his blood group or any similar difficulty faced in confirmation of his blood group in the past.

Further Testing Results and Interpretations



Final confirmation of donor blood group from donor bag –

Cold adsorption and heat elution steps from bag samples (from bag tube segment post stripping)

as done previously with donor tube Sample.

Conclusions



Final Donor Blood group is:

A_{weak} RhD Positive

and donor given final blood group report.

Donor to be considered as A positive
and if required to be transfused;
to receive O positive RBC units and
A positive/AB positive FFP/PC.

Of note, this transfusion recommendation may
change depending on countries.

Summary of Case Challenges



1. A_{weak} ABO typing confirmation.
2. Use of cold adsorption technique.
3. Use of heat elution technique.

Other possibility to further study this case: Molecular investigation of the *ABO* gene

Lessons Learned by the Case



1. Cell grouping and serum grouping must be performed by Tube method as a Gold standard.
2. Serum grouping step is very important in blood group confirmation and cell and serum grouping results must correlate.
3. Cell washing step is very important in tube testing
4. Final blood group confirmation in case of donor blood group discrepancy must be done by a specialized immunohematology lab with special tests like cold adsorption techniques and heat elution techniques.