



Bits and Pieces: When Missed Information Makes an Impact

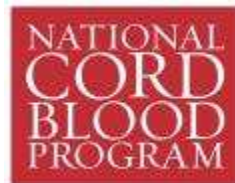
Megan Dupont MLS(ASCP)^{CM}SBB^{CM}





Expanding our organization to meet clinical, cellular and transfusion product and service needs for patients.

Now providing almost one million blood products, over 450,000 laboratory and multi-assay infectious disease tests, and over 12,500 specialty clinical procedures annually to hospitals nationwide.



CASE BACKGROUND

Here's what we know...

- LB is a 15 year old male trauma victim
- O Pos
- Screening Cells II & III both 2+ (Gel)
- Negative DAT
- Multiply transfused 8 days previously
- Client is requesting antibody identification and 4 units of RBCs to have available for AM surgery



OUR WORKUP- ABORH & DAT

	ABO Group				Rh Type	
	Anti-A	Anti-B	A ₁ Cells	B Cells	Anti-D	Control
IS	0	0	4+	4+	r	0
5'RT					1+ ^w	0
15'RT					2+ ^{mf*}	0



*Hospital reported that the patient is O Pos and had received O Neg RBCs recently

Direct Antiglobulin Test

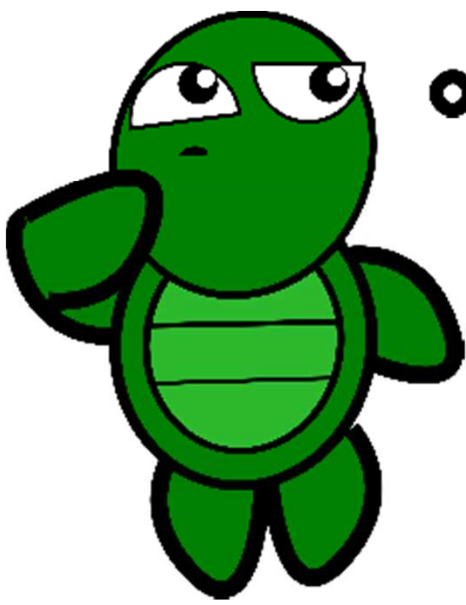
Poly	IgG	C'	Saline
(+)	(+ ^w)	(0) ✓	(0)

OUR WORKUP – 1ST PANEL

		Rh					Kell		Duffy		Kidd		MNS				Results		
		D	C	E	c	e	K	k	Fy^a	Fy^b	Jk^a	Jk^b	M	N	S	s	5 min RT	PEG IAT	
1	R ₁ R ₁	+	+	0	0	+	+	0	+	0	0	+	0	+	0	+	0	0	(0) ✓
2	R ₂ R ₂	+	0	+	+	0	0	+	0	+	0	+	0	+	0	+	0	0	(0) ✓
3	R ₁ R ₁	+	+	0	0	+	0	+	0	0	0	+	We will come back to this reactivity!				0	(0) ✓	
4	rr	0	0	0	+	+	0	+	0	0	0	+					0	0	+
5	R ₁ R ₁	+	+	0	0	+	0	+	0	+	0	+	+	0	+	+	0	0	(+ ^w)
6	R ₁ R ₁	+	+	0	0	+	0	+	0	+	+	0	0	+	0	+	0	0	(0) ✓
7	R ₁ R ₁	+	+	0	0	+	0	+	+	0	0	+	0	+	0	+	0	0	(0) ✓
8	rr	0	0	0	+	+	0	+	+	+	0	+	+	0	+	0	0	0	(0) ✓
Auto																	0	0	(0) ✓

**Not quite what
we expected...**

What testing
methodology was
our client using
again?



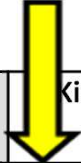
CASE BACKGROUND

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OUR WORKUP - 1ST PANEL MTS GEL TESTING

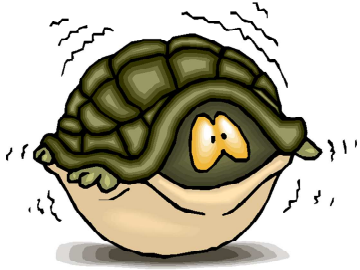


		Rh					Kell		Duffy		Kidd		MNS				Results		
		D	C	E	c	e	K	k	Fy^a	Fy^b	Jk ^a	Jk^b	M	N	S	s	5 min RT	PEG IAT	MTS GEL
1	R ₁ R ₁	+	+	0	0	+	+	0	0	+	0	+	0	+	0	+	0	(0) ✓	0
2	R ₂ R ₂	+	0	+	+	0	0	+	0	+	0	+	0	+	0	+	0	(0) ✓	0
3	R ₁ R ₁	+	+	0	0	+	0	+	0	0	0	+	0	+	0	0	0	(0) ✓	0
4	rr	0	0	0	+	+	0	+	0	0	0	+	+	0	0	+	0	(0) ✓	0
5	R ₁ R ₁	+	+	0	0	+	0	+	0	+	0	+	+	0	+	+	0	(+ ^w)	0
6	R ₁ R ₁	+	+	0	0	+	0	+	0	+	+	0	0	+	0	+	0	(0) ✓	2+
7	R ₁ R ₁	+	+	0	0	+	0	+	+	0	0	+	0	+	0	+	0	(0) ✓	0
8	rr	0	0	0	+	+	0	+	+	+	0	+	+	0	+	0	0	(0) ✓	0
Auto																	0	(0) ✓	0

OUR WORKUP-2ND GEL PANEL



		Rh					Kell		Duffy		Kidd		MNS				Results
		D	C	E	c	e	K	k	Fy ^a	Fy ^b	Jk ^a	Jk ^b	M	N	S	s	
1	rr	0	0	0	+	+	+	+	0	+	+	0	+	0	+	0	2+
2	rr	0	0	0	+	+	0	+	+	+	+	0	+	0	+	0	2+
3	R ₁ R ₁	+	+	0	0	+	0	+	+	0	+	0	+	0	0	+	2+
4	R ₂ R ₂	+	0	+	+	0	+	+	+	0	+	0	0	+	0	+	2+
5	rr	0	0	0	+	+	0	+	+	0	+	+	0	+	0	+	1+ ^w
6	R ₁ R ₁	+	+	0	0	+	+	0	+	+	+	+	+	+	0	+	1+
7	R ₂ R ₂	+	0	+	+	0	0	+	+	0	0	+	+	+	+	+	0
8	rr	0	0	0	+	+	+	+	+	0	0	+	+	0	+	0	0



ELUSIVE KIDD BLOOD GROUP ANTIBODIES...

Kay B, Poisson JL, Tuma CW, Shulman IA (2016). Anti-Jk^a that are detected by solid-phase red blood cell adherence but missed by gel testing can cause hemolytic transfusion reactions. *Transfusion*, 56(12), 2973-2979.

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Villa MA, Moulds M, Coluccio EB, Pizzo MN, Paccapel C, Revelli, N, Morelati F, Truglio F, Manera MC, Tedeschi A, Marconi M (2007). An acute haemolytic transfusion reaction due to anti-Jka. *Blood Transfusion*, 5(2), 102-106.

DOES THE ANTIGEN TYPING SUPPORT THE RESULTS?

	Anti-Jk ^a
Pos	2+ ^s
Neg	0
Patient Cells	1+ ^{mf}



- O Pos
- Screening Cells II & III both 2+ (Gel)
- Negative DAT
- Patient multiply transfused 8 days previously
- Client is requesting antibody identification and 4 units of RBCs to have available for AM surgery



DENSITY GRADIENT CELL SEPARATION

Washed PRBCs
into microhct
tubes

- ≥ 3 days since transfusion
- Sample <24 hours old
- Mix continuously while filling tubes

Centrifuge for
15 minutes

- This method will only be effective if patient is producing reticulocytes
- Cells containing Hgb S are not effectively separated by this method

Cut tube 5mm
below the top of
the column of
cells

- Newly formed autologous red cells generally have a lower specific gravity than transfused red cells and will therefore concentrate at the top of the column of red cells when blood is centrifuged in a microhematocrit tube

WHAT DO THE RETICS TELL US?

	Anti-Jk ^a
Pos	2+ ^s
Neg	0
Whole Blood	1+ ^{mf}
Patient Reticulocytes	(0)

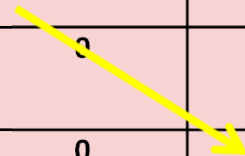


What we know so far:

- O Positive
- Positive DAT
- Anti-Jk^a in plasma, retics type Jk(a-)

WAIT...WHAT ABOUT CELL #5?

		Rh					Kell		Duffy		Kidd		MNS				Results						
		D	C	E	c	e	K	k	Fy ^a	Fy ^b	Jk ^a	Jk ^b	M	N	S	s	5 min RT	PEG IAT					
1	R ₁ R ₁	+	+	0	0	+	+	0	0	+	0	+	0	+	0	+	0	0	0	0	0	(0) ✓	
2	R ₂ R ₂	+	0	+	+	0	0	+	0	+	0	+	0	+	0	+	0	0	0	0	0	(0) ✓	
3	R ₁ R ₁	+	+	0	0	+	0	+	0	0	0	+	0	+	0	0	0	0	0	0	0	(0) ✓	
4	rr	0	0	0	+	+	0	+	0	0	0	+	+	0	0	+	0	0	0	0	0	(0) ✓	
5	R ₁ R ₁	+	+	0	0	+	0	+	0	+	0	+	+	0	+	+	0	0	0	0	0	(+ ^w)	
6	R ₁ R ₁	+	+	0	0	+	0	+	0	+	+	0	0	+	0	+	0	0	0	0	0	(0) ✓	
7	R ₁ R ₁	+	+	0	0	+	0	+	+	0	0	+	0	+	0	+	0	0	0	0	0	(0) ✓	
8	rr	0	0	0	+	+	0	+	+	+	0	+	+	0	+	+	0	0	0	0	0	(0) ✓	
Aut o																						0	(0) ✓



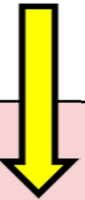
HLA ANTIBODY OR ANTIBODY TO LOW INCIDENCE ANTIGEN?

- Suspect when only one/two cells reactive
- All common specificities have been excluded



- Resolution:
 - **EGA treat reactive cells**
 - Test selective cells positive for low incidence antigens
 - Consider dosage or carryover of cold reactive antibody; evaluate reactivity at RT & 4C
 - Treat reactive cells with DTT, trypsin and/or ficin and repeat testing

WAIT...WHAT ABOUT CELL #5?



		Rh					Kell		Duffy		Kidd		MNS				Results		
		D	C	E	c	e	K	k	Fy ^a	Fy^b	Jk ^a	Jk^b	M	N	S	s	5 min RT	PEG IAT	EGA PEG IAT
1	R ₁ R ₁	+	+	0	0	+	+	0	0	+	0	+	0	+	0	+	0	(0) ✓	/
2	R ₂ R ₂	+	0	+	+	0	0	+	0	+	0	+	0	+	0	+	0	(0) ✓	/
3	R ₁ R ₁	+	+	0	0	+	0	+	0	0	0	+	0	+	0	0	0	(0) ✓	/
4	rr	0	0	0	+	+	0	+	0	0	0	+	+	0	0	+	0	(0) ✓	/
5	R ₁ R ₁	+	+	0	0	+	0	+	0	+	0	+	+	0	+	+	0	(+ ^w)	(0) ✓
6	R ₁ R ₁	+	+	0	0	+	0	+	0	+	0	0	+	0	+	+	0	(0) ✓	/
7	R ₁ R ₁	+	+	0	0	+	0	+	+	0	0	+	0	+	0	+	0	(0) ✓	/
8	rr	0	0	0	+	+	0	+	+	+	0	+	+	0	+	0	0	(0) ✓	/
Auto																	0	(0) ✓	/

OUR WORKUP - ELUATE

Direct Antiglobulin Test

Poly	IgG	C'	Saline
(+) ^{mf}	(+ ^w) ^{mf}	(0)	(0)

		Rh					Kell		Duffy		Kidd		MNS				Results
		D	C	E	c	e	K	k	Fy^a	Fy^b	Jk^a	Jk^b	M	N	S	s	PEG IAT
1	R ₂ R ₂	+	0	+	+	0	0	+	0	+	0	+	0	+	0	+	(0) ✓
2	R ₀ R ₀	+	0	+	+	0	0	+	0	+	+	0	+	+	+	0	(0) ✓
3	rr	0	0	0	+	+	+	+	0	+	+	0	+	0	+	0	(0) ✓
4	R ₁ R ₁	+	+	0	0	+	0	+	+	0	+	0	+	0	0	+	(0) ✓
5	rr	0	0	0	+	+	0	+	+	0	+	0	0	+	0	+	(0) ✓
6	R ₁ R ₁	+	+	0	0	+	0	+	+	+	+	+	+	0	0	+	(0) ✓
7	R ₂ R ₂	+	0	+	+	0	+	+	+	0	+	0	0	+	0	+	(0) ✓
8	rr	0	0	0	+	+	+	+	0	+	0	+	0	+	0	+	(0) ✓

FINAL REPORT

- **O Pos**
- **Positive DAT (IgG): Cause unknown**
- **Eluate: No alloantibodies detected**
- **Plasma: Anti-Jk^a, patient's retics type Jk(a-)**
- **Plasma: HLA antibody**
- **Transfusion recommendation:**
 - Jk(a-)
 - Donor blood should be nonreactive with patient's plasma
- **Sent 4 O Pos, Jk(a-) LRPC that were nonreactive with patient's plasma**

3 DAYS LATER...



**Client requested 4
additional O Pos, Jk(a-)
units for surgery.**

7 DAYS LATER...

- Client submitted sample for antibody identification and has requested 2 units STAT
- Patient has received a total of 13 Red Cell transfusions, the most recent being 2 days prior to specimen collection
- All samples submitted are hemolyzed



WORKUP #2 – ABORH & DAT

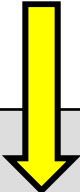
	ABO Group				Rh Type	
	Anti-A	Anti-B	A ₁ Cells	B Cells	Anti-D	Control
IS	0	0	4+	4+	2+ ^{mf}	0



Direct Antiglobulin Test

Poly	IgG	C'	Saline
(0) ✓	(+)	(0) ✓	(0)

WORKUP #2 – 1ST PANEL

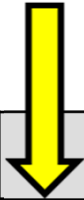


		Rh					Kell		Duffy		Kidd		MNS				Results	
		D	C	E	c	e	K	k	Fy^a	Fy ^b	Jk ^a	Jk^b	M	N	S	s	5 min RT	PEG IAT
1	R ₁ R ₁	+	+	0	0	+	+	+	0	+	0	+	+	+	+	0	0	(+)
2	rr	0	0	0	+	+	0	+	+	+	0	+	+	0	+	0	0	(0) ✓
3	R ₂ R ₂	+	0	+	+	0	0	+	+	0	0	+	+	0	+	+	0	1+
4	R ₁ R ₁	+	+	0	0	+	+	0	0	+	0	+	0	+	0	+	0	1+
5	R ₂ R ₂	+	0	+	+	0	0	+	0	+	0	+	0	+	0	+	0	1+
6	R ₂ r'	+	+	+	0	+	0	+	+	0	0	+	+	0	0	+	0	(+)
7	rr	0	0	0	+	+	0	+	+	0	0	+	+	0	0	+	0	(0) ✓
Auto																	0	(+)

History of anti-Jk^a and HLA antibody



WORKUP #2 – 2ND PANEL



		Rh					Kell		Duffy		Kidd		MNS				Results
		D	C	E	c	e	K	k	Fy^a	Fy^b	Jk ^a	Jk^b	M	N	S	s	PEG IAT
1	R ₁ R ₁	+	+	0	0	+	0	+	+	0	0	+	+	0	+	+	(+)
2	R ₂ R ₂	+	0	+	+	0	0	+	+	0	0	+	+	0	+	+	1+
3	r'r	0	+	0	+	+	0	+	0	+	0	+	0	+	+	+	(0) ✓
4	rr	0	0	0	+	+	0	+	0	+	0	+	+	+	0	+	(0) ✓
5	rr	0	0	0	+	0	0	+	+	0	0	+	0	+	0	+	(0) ✓
6	rr	0	0	0	+	+	+	+	+	+	0	+	+	0	+	+	(0) ✓
7	rr	0	0	0	+	+	0	+	+	+	0	+	+	0	+	0	(0) ✓
8	rr	0	0	0	+	+	+	+	0	+	0	+	+	+	+	+	(0) ✓
9	r'r'	0	+	0	0	+	+	+	+	+	0	+	0	+	0	+	(0) ✓
10	r'r'	0	+	0	0	+	0	+	+	+	0	+	+	0	+	+	(0) ✓
11	r''r''	0	0	+	+	0	0	+	+	+	0	+	+	+	0	+	(0) ✓
12	r''r''	0	0	+	+	0	0	+	0	+	0	+	+	0	0	+	(0) ✓

WORKUP #2 - ELUATE

Direct Antiglobulin Test

Poly	IgG	C'	Saline
(0)	(+)	(0)	(0)



		Rh					Kell		Duffy		Kidd		MNS				Results
		D	C	E	c	e	K	k	Fy^a	Fy^b	Jk^a	Jk^b	M	N	S	s	PEG IAT
1	R ₁ R ₁	+	+	0	0	+	0	+	+	0	0	+	+	0	+	+	(+)
2	R ₀ R ₀	+	0	0	+	+	0	+	+	0	0	+	+	+	+	0	(+)
3	rr	0	0	0	+	+	0	+	+	+	0	+	0	+	0	0	(0) ✓
4	rr	0	0	0	+	+	+	+	0	+	0	+	+	+	+	+	(0) ✓
5	rr	0	0	0	+	+	+	+	+	0	0	+	0	+	0	+	(0) ✓
6	rr	0	0	0	+	+	0	+	+	+	0	+	0	+	0	0	(0) ✓
7	R ₂ R ₂	+	0	+	+	0	0	+	+	0	0	+	+	0	+	+	1+
8	rr	0	0	0	+	+	0	+	+	0	0	+	0	+	0	+	(0) ✓
9	r'r'	0	+	0	0	+	+	+	+	0	+	0	+	0	+	+	(0) ✓
10	r'r'	0	+	0	0	+	0	+	+	0	+	+	0	+	+	+	(0) ✓
11	r''r''	0	0	+	+	0	0	+	+	0	+	+	+	0	+	+	(0) ✓
12	r''r''	0	0	+	+	0	0	+	0	+	0	+	+	0	0	+	(0) ✓



So you're telling me our patient appears to have anti-D in both his plasma and his eluate? But...isn't he O Pos???

WORKUP CONCLUSIONS

- Appears as if there is anti-D in the eluate & plasma
- Patient has historically been O Positive
- Is the anti-D autoantibody or alloantibody?
 - Partial D perhaps?



TIME FOR SOME INVESTIGATING

ABO Group					Rh Type	
	Anti-A	Anti-B	A ₁ Cells	B Cells	Anti-D	Control
IS	0	0	4+	4+	2+ ^{mf}	0

	Anti-D
Retics from Cell Sep*	r ^{mf}



LET'S TRY TO GET SOME MORE INFO...



1. Contacted facility requesting workup:
 - Patient urgently transfused 2 O+ & 1 O- RBCs during LifeFlight to facility
 - Patient's cells typed weakly with anti-D (1+^w), reported as O Positive
 - Obtained facility information where patient was originally treated

2. Contacted facility where patient was 1st treated:
 - ABORh: **O Negative**
 - Negative antibody screen
 - Transfused 2 O+ & 1 O- RBCs during LifeFlight to transfer facility

FINAL REPORT

- **ABORh: O NEGATIVE**
- **DAT: Positive (IgG)**
- **Plasma & Eluate: Anti-D**
 - **Previously identified anti-Jk^a & HLA antibody**
- **Transfusion Recommendation:**
 - **D-negative**
 - **Jk(a-)**
 - **Nonreactive with the patient's plasma (HLA)**
- **Sent 2 D-negative, C-negative, E-negative, Jk(a-) units prior to completion of workup for emergency use in surgery**

“Unity is strength... when there is teamwork and collaboration, wonderful things can be achieved.”

Mattie Stepanek



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Kay B, Poisson JL, Tuma CW, Shulman IA (2016). Anti-Jk^a that are detected by solid-phase red blood cell adherence but missed by gel testing can cause hemolytic transfusion reactions. *Transfusion*, 56(12), 2973-2979.

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QUESTIONS???





Community Blood Center

 **New York**
Blood Center

 INNOVATIVE
BLOOD
RESOURCES

Rhode Island
**Blood
Center**

 **Blood Bank**
of Delmarva

NATIONAL
CENTER FOR
BLOOD GROUP
GENOMICS



GENOMICS FOR THE
BLOOD GROUP

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