CASE OF THE MONTH: NOVEMBER, 2019
(Case study by Jim Perkins, © 2019)

Indian Immunohematology Initiative

History: A 73 year old woman with spondylolisthesis (slippage of one vertebra on another) was scheduled for L4-5 and L5-S1 fusion and L5-S1 laminectomies with autologous iliac bone grafting. She did not have a history of transfusion at our institution including at the time of laminectomy at the same site 3 years earlier. In anticipation of significant operative bleeding autologous blood collection was ordered, and two units were drawn, 13 and 9 days before surgery; the antibody detection test ("antibody screen") performed on both units was negative.

On admission a type-and-screen was performed and two additional units of allogeneic RBCs were ordered by the surgeon with results as below. Since the antibody screen was negative, 3 abbreviated ("immediate spin") crossmatches were performed initially.

ABO and Rh Typing

<A	<B	A1 cells	B cells	6% alb	<D	<D/AHG	CCC	Interp
0	4+	4+	0		4+			

Antibody Screen

	Gel
SCI	0
SCII	0

Crossmatches

	IS
Unit #1; B-pos	0
Unit #2; B-pos	0
Unit #3; B-pos	4+

Question:

1. What do you think might be going on here? Based on your hypothesis(es) how might you proceed

The technologist performed the following tests with results as shown below:

- confirmed the forward ("cell") typing on the three units labeled as group B,
- extended the crossmatches by incubating and testing the crossmatches at 37^{o} and adding a Coombs test,
- crossmatching two group O RBCs,
- performed a DAT on the unit yielding the positive crossmatch ("index donor"),
- performed a DAT on the patient's RBCs and on the index donor RBCs.

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Crossmatches; LISS/Tube method **Prewarmed technique** **DAT** **Repeat forward typing**

	IS	37ºC/15'	AHG	37ºC/15'	AHG	Poly-AHG	Anti-IgG	Anti-C3d	Anti-A	Anti-B
Unit #1; B-pos	0	0	0√						0	4+
Unit #2; B-pos	0	0	0√						0	4+
Unit #3; B-pos	4+	4+	2+	0	w+	0√	0√		0	4+
Unit #4; O-pos	0	0	0√							
Unit #5; O-pos	0	0	0√							
Patient						0√	0√	0√		

Question:

2. Now what do you think might be going on? Based on your new hypothesis how might you proceed?

At this point the technologist pursued the possibility that the patient had a cold-reactive allo- or auto-antibody by performing the following.

Initial panel, LISS/tube

Lot #37953			Rh system						Kell						Duffy		Kidd		Lewis		P	MNSs				Lutheran		Xg		LISS/tube			
Cell	**Special type**	**Rh**	**D**	**C**	**c**	**E**	**e**	**V**	**K**	**k**	**Kp^a**	**Kp^b**	**Js^a**	**Js^b**	**Fy^a**	**Fy^b**	**Jk^a**	**Jk^b**	**Le^a**	**Le^b**	**P1**	**M**	**N**	**S**	**s**	**Lu^a**	**Lu^b**	**Xg^a**	**Cell**	**IS**	**15' RT**	**37º**	**IgG**
1	**Lu:14**	**R1R1**	+	+	0	0	+	0	+	0	0	+	0	+	+	0	+	+	+	0	+	+	0	0	+	0	+	+	1	0	vw+	0	0√
2	**Bg^a+, Co^b+**	**R1wR1**	+	+	0	0	+	0	+	+	+	+	0	+	0	+	+	0	0	+	0	+	0	+	+	0	+	+	2	0	0	0	0√
3	**Yt^b+**	**R2R2**	+	0	+	+	0	0	0	+	0	+	0	+	0	+	0	+	0	+	0	0	+	+	+	0	+	+	3	0	0	0	0√
4	**He+**	**Ror**	+	0	+	0	+	+	0	+	0	+	0	+	0	0	+	0	0	+	+	0	+	+	+	0	+	0	4	w+	2+	w+	w+
5		**r'r**	0	+	+	0	+	0	0	+	0	+	0	+	0	+	0	+	0	+	+	+	0	+	+	0	+	0	5	0	0	0	0√
6	**Co^b+**	**r"r**	0	0	+	+	+	0	0	+	0	+	0	+	+	+	+	0	0	+	+	+	+	+	+	0	+	+	6	vw+	1+	0	0√
7	**Co^b+**	**rr**	0	0	+	0	+	0	+	+	0	+	0	+	0	+	+	+	0	+	+	+	+	+	+	0	+	0	7	0	w+	0	0√
8		**rr**	0	0	+	0	+	0	0	+	0	+	0	+	+	0	+	0	+	0	+	+	+	0	+	0	+	+	8	0	w+	0	0√
9		**rr**	0	0	+	0	+	0	0	+	0	+	+	+	0	0	+	+	0	0	0	+	+	0	+	0	+	0	9	0	0	0	0√
10		**R1R1**	+	+	0	0	+	0	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	+	0	+	0	10	0	0	0	0√
11	**I-, Yt^b+**	**R1R1**	+	+	0	0	+	0	0	+	0	+	0	+	+	0	+	+	+	0	0	+	+	0	+	0	+	+	11	0	w+	0	0√
Patient																													AC				

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Extended Phenotype

	Rh system				Kell				Kidd		Duffy		Lewis		MNSs							
	C	E	c	e	K	k	Kp^a	Js^a	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	S	s	M	N	P1	I	H	A_1	
Patient	4+	4+	4+	3+									0	0			4+	2+	0				
Pos control	3+	4+	4+	3+									2+	2+			3+	2+	3+				
Neg control	0	0	0	0									0	0			0	0	0				

Questions:

3. Do the new findings suggest a hypothesis that can be tested? What tests would you like to do base on your impression from the available results? Is there any testing you think should have been done above?

At this point the initial technologist went to lunch and the problem was handed off to a colleague. The latter individual diluted the cells in the initial panel and tested it with the patient's plasma by the gel method, by which all the cells were nonreactive. Three additional gel panels were completely non-reactive with the patient's plasma. However, RBCs from the index donor cells reacted 3+ by the gel method. In addition, enzyme treatment of the of the index donor RBCs eliminated their reactivity with the patient's plasma.

The second technologist also tested a "cold panel" and performed a P1 neutralization with the following results.

"Cold panel"

	Cell phenotype					Saline/tube technique			
Cell ID	**M**	**N**	**Le^a**	**Le^b**	**P1**	**IS**	**RT, 15'**	**15ºC, 15'**	**4ºC, 15'**
Panel 37953 - TC	+	+	+	0	0	0	0	w+	2+
Panel 40001 - #8	0	+	0	+	0	0	0	w+	2+
Panel 39981 - #8	+	0	0	0	0	0	0	1+	2+
Cord cell I						0	0	0	1+
Cord cell I						0	0	1+	2+
Cord cell I						0	0	0	1+
B cells						0	0	0	1+
Auto control						0	0	1+	2+

P1 neutralization

	P1 substance				Saline control			
Cell ID	**IS**	**RT**	**37º**	**IgG**	**IS**	**RT**	**37º**	**IgG**
Panel 37953 - #3, P1 neg	0	0	0	0√	0	0	0	0√
Panel 37953 - #4, P1 pos	vw+	w+	0	0√	vw+	1+	0	0√
Panel 37953 - #6, P1 pos	0	0	0	0√	0	0	0	0√
Panel 37953 - #7, P1 pos	0	0	0	0√	0	0	0	0√

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Questions:

4. What is your impression now? Are there any data which you would discard from consideration at this point? What testing would you try now?

It was now the end of the day shift, and an evening shift technologist took over the case. This individual tested the patient plasma with a second panel by the saline/tube method, as well as a panel of cells selected from the laboratory's library of rare frozen RBCs. The panels and the corresponding results were as follows.

Repeat panel, saline/tube

Lot #39981			Rh system						Kell						Duffy		Kidd		Lewis		P	MNSs				Lutheran		Xg		LISS/tube			
Cell	Special type	Rh	D	C	c	E	e	V	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P1	M	N	S	s	Lu^a	Lu^b	Xg^a	Cell	IS	15' RT	37°	IgG
1		R1R1	+	+	0	0	+	0	0	0	0	+	0	+	+	+	+	0	+	0	+	+	+	0	+	0	+	+	1	0	0	0	0√
2		R1wR1	+	+	0	0	+	0	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	+	0	+	+	2	0	0	0	0√
3		R2R2	+	0	+	+	0	0	0	+	0	+	0	+	0	+	+	0	0	+	+	0	+	0	+	0	+	0	3	0	0	0	0√
4	Bg^a+	Ror	+	0	+	0	+	+	0	+	0	+	0	+	0	0	+	0	0	0	+	+	0	0	0	0	+	0	4	0	0	0	0√
5		r'r	0	+	+	0	+	0	0	+	0	+	0	+	+	0	+	+	0	+	+	+	+	+	0	0	+	+	5	0	0	0	0√
6		r"r	0	0	+	+	+	0	+	0	0	+	0	+	+	0	0	+	0	+	+	+	+	0	+	0	+	+	6	0	0	0	0√
7		rr	0	0	+	0	+	0	+	+	0	+	0	+	0	+	+	0	0	+	+	+	+	0	+	0	+	+	7	0	0	0	0√
8		rr	0	0	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	0	0	+	0	0	+	0	+	0	8	0	0	0	0√
9		rr	0	0	+	0	+	0	0	+	0	+	0	+	0	0	+	+	+	0	+	0	+	0	0	0	+	+	9	0	0	0	0√
10		R1R1	+	+	0	0	+	0	+	+	0	+	0	+	+	0	+	+	+	0	0	+	+	+	0	0	+	+	10	0	0	0	0√
11	He+	rr	0	0	+	0	+	0	+	+	0	+	0	+	+	0	0	+	0	+	+	0	+	+	+	0	+	+	11	0	0	0	0√
Patient																													AC	0	0	0	0√

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Selected cells, saline/tube, 2 drops of plasma

Lot #39981			Rh system						Kell						Duffy		Kidd		Lewis		P	MNSs				Lutheran		Xg		Saline/tube			
Cell	Special type*	Rh	D	C	c	E	e	V	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P1	M	N	S	s	Lu^a	Lu^b	Xg^a	Cell	IS	15' RT	37°	IgG
1	Wr^a +	R2r	+	0	+	+	+	0	0	+	0	+	0	+	+	+	+	+	+	0	+	+	+	+	+	0	+	0	1	0	0	0	0√
2	Wr^a +	R2R2	+	0	+	+	0		0	+					+	+	+	0	0	+	+	+	0	+	+				2	0	0	0	0√
3	Hil+	R1r	+	+	+	0	+		0	+	0	+	0	+	+	+	+	+	0	+	0	+	0	+	+	0	+		3	0	0	0	0√
4	Mi VIII	R1r	+	+	+	0	+	0	0	+	0	+	0	+	+	+	0	+			+	+	+	0	+	0		+	4	0	0	0	0√
5	Mg+	R1R1	+	+	0	0	+		+	+	0	+			+	+					+	0	+	0	+	0	+		5	0**	0**	0	0√
Repeat testing of cell #5 by the saline/tube method using 4 drops of patient plasma.																													5	$1+^w$	1+	0	0√
6	Mt^a	rr	0	0	+	0	+		0	+	0				+	0	+	+				+	+	+	+				6	0	0	0	0√
7	St^a	R2r	+	0	+	+	+		0	+					+	0	+	0	0	+	+	+	+	+	+				7	0	0	0	0√
8	Mi III	R1R1	+	+	0	0	+		0						+	0	+	0	0	+		+	+	0	+				8	0	0	0	0√
9	Mi^a+	R1r	+	+	+	0	+	0	+	+	0	+	0	+	+	+	0	+	+	0	+	+	+	+	+	0	+	0	9	0	0	0	0√
10	Mg+	R2r	+	0	+	+	+	0	0	+	0	+			+	+	+	0	0	+		0	+	0	+	0			10	3+		1+	1+
11	Mg+	R2r	+	0	+	+	+		0	+	0	+			+	+	+	+	0	+		0	+	0	+				11	2+		w+	1+
12	He+	rr	0	0	+	0	+	0	+	+	0	+	0	+	+	0	0	+	0	+	+	0	+	+	+	0	+	+	12	0	0	0	0√
13	He+	R1R1	+	+	0	0	+	0	0	+	0	+	0	+	0	+	+	0	0	+	+	+	+	+	+	0	+	0	13	0	0	0	0√
14	Hut +	Ror	+	0	+	0	+	+	0	+	0	+	0	+	0	0	+	+	0	0	0	+	+	+	+	0	+	0	14	0	0	0	0√
15	He+	Ror	+	0	+	0	+	+	0	+	0	+	0	+	0	0	+	0	0	+	+	0	+	0	+	0	+	0	15	0	1+	0	0√
16	Mg+	R1R2	+	+	+	+	+	0	0	+	0	+	0	+	+	+	0	+	0	+	0	0	+	0	+	0	+	0	16	3+		1+	w+

***Special types are as listed by the submitting laboratory (SCARF). **Initially read as "rough".**

5. What is your impression now? Are the final data conclusive?

6. How would you select RBCs that are safe to transfuse?

7. What do we know about the antigen the patient's antibody is directed against?