American Red Cross

It’s **N**ot What You Think!

Initial presentation

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Missouri and Arkansas Region

- Black female, roughly 70 years old
- Hgb: 13.2g/dL
- Transfusion, surgical, and OB history unclear

Patient Transfusion History	If transfused, provide number of units/dates and facility.
Patient has been transfused at another facility	Unknown. No history available in system or at other local hospital blood banks.

Additional Patient Information:
Transfusion/pregnancy history is unknown.

- Complaint: STAT presurgical admit type and screen
  - Surgery scheduled in 6 days
  - No units ordered
  - Limited specimen sent

Initial presentation

American Red Cross
Missouri and Arkansas Region

Gather more information if necessary

- Contacted hospital, requested additional information
- Questions:
  - Transfused or not? When? Where?
  - Known antibody history?
  - Any relevant diagnoses, medications, etc?

- Hospital offered to contact patient and/or patient's doctor's office
  - "It might take a while"

Blood type

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Anti-A	Anti-B	Anti-A,B	Anti-D	Rh control	A_1 cells	A_2 cells	B cells
4+	0	4+	3+	0	4+	4+	4+

Type: NTD positive

DAT: Polyspecific tube testing

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Missouri and Arkansas Region

Poly IS	Control IS	Poly 5’ RT	Control 5’ RT
0	0	0 √	0

DAT: Negative

Initial panel: IS

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Missouri and Arkansas Region

	Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
			D	C	E	c	e	f	V	C^w	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		IS		
1	Immucor 37832	B3785 R1wR1 #1	+	+	0	0	+			+	0	+	0	+	0	+	+	+	+	0	+	0	+	0	+	+	+	0	+	+		4+		
2	Immucor 37832	B9783 R1R1 #3	+	+	0	0	+			0	0	+	+	+	+	w	0	0	+	0	+	0	+	+	0	0	+	0	+	+		4+		
3	Immucor 37832	B11193 R1R1 #5	+	+	0	0	+			0	+	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	0	+	+		4+		
4	Immucor 37832	C5066 R2R2 #7	+	0	+	+	0			0	0	+	+	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	+	+		4+		
5	Immucor 37832	C7328 R2R2 #9	+	0	+	+	0			0	0	+	0	+	0	+	+	+	+	0	+	0	+	+	+	0	+	0	+	0		4+		
6	Immucor 37832	R2421 R1r #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	+	0	+	+	+	+	0	0	+	+		4+		
7	Immucor 37832	E967 r'r #11	0	+	0	+	+			0	+	+	+	0	+	0	+	0	+	0	+	0	+	+	0	+	+	0	+	0		4+		
8	Immucor 37832	F956 r"r #12	0	0	+	+	+			0	+	+	0	+	+	0	+	0	+	0	+	0	+	+	+	+	+	0	+	+		4+		
9	Immucor 37832	F1079 r"r #15	0	0	+	+	+			0	+	+	0	+	+	0	0	0	+	0	+	0	+	+	+	+	+	0	+	+		4+		
10	Immucor 37832	N5173 rr #16	0	0	0	+	+			0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	+	+	0	+	+		4+		
11	Immucor 37832	D563 Ror #20	+	0	0	+	+			0	0	+	+	+	0	0	+	0	+	0	+	+	+	0	0	+	0	0	+	0	He+	4+		
	Auto Control																															0		

Initial panel: IS + 15’ RT

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Missouri and Arkansas Region

	Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
			D	C	E	c	e	f	V	C^{W}	M	N	S	s	Le^{a}	Le^{b}	P1	K	k	Kp^{a}	Kp^{b}	Js^{a}	Js^{b}	Fy^{a}	Fy^{b}	Jk^{a}	Jk^{b}	Lu^{a}	Lu^{b}	Xg^{a}		IS	15’ RT	
1	Immucor 37832	B3785 R1wR1 #1	+	+	0	0	+			+	0	+	0	+	0	+	+	+	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	
2	Immucor 37832	B9783 R1R1 #3	+	+	0	0	+			0	0	+	+	+	+	w	0	0	+	0	+	0	+	+	0	0	+	0	+	+		4+	4+	
3	Immucor 37832	B11193 R1R1 #5	+	+	0	0	+			0	+	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	
4	Immucor 37832	C5066 R2R2 #7	+	0	+	+	0			0	0	+	+	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	
5	Immucor 37832	C7328 R2R2 #9	+	0	+	+	0			0	0	+	0	+	0	+	+	+	+	0	+	0	+	+	+	+	0	+	0	+	0		4+	4+	
6	Immucor 37832	R2421 R1r #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	+	0	+	+	+	+	+	0	+	+		4+	4+	
7	Immucor 37832	E967 r'r #11	0	+	0	+	+			0	+	+	+	0	+	0	+	0	+	0	+	0	+	+	+	0	+	+	0	+	0		4+	4+	
8	Immucor 37832	F956 r"r #12	0	0	+	+	+			0	+	+	0	+	+	0	+	0	+	0	+	0	+	+	+	+	+	+	0	+	+		4+	4+	
9	Immucor 37832	F1079 r"r #15	0	0	+	+	+			0	+	+	0	+	+	0	0	0	+	0	+	0	+	+	+	+	+	+	0	+	+		4+	4+	
10	Immucor 37832	N5173 rr #16	0	0	0	+	+			0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	
11	Immucor 37832	D563 Ror #20	+	0	0	+	+			0	0	+	+	+	0	0	+	0	+	0	+	+	+	0	0	+	0	0	+	0	He+	4+	4+	
	Auto Control																															0	0	

Initial panel: IS + 15’ RT + 4°C

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Missouri and Arkansas Region

	Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
			D	C	E	c	e	f	V	C^{W}	M	N	S	s	Le^{a}	Le^{b}	P1	K	k	Kp^{a}	Kp^{b}	Js^{a}	Js^{b}	Fy^{a}	Fy^{b}	Jk^{a}	Jk^{b}	Lu^{a}	Lu^{b}	Xg^{a}		IS	15’ RT	15’ 4C
1	Immucor 37832	B3785 R1wR1 #1	+	+	0	0	+			+	0	+	0	+	0	+	+	+	+	0	+	0	+	0	+	+	+	+	0	+	+		4+	4+	4+
2	Immucor 37832	B9783 R1R1 #3	+	+	0	0	+			0	0	+	+	+	+	w	0	0	+	0	+	0	+	+	0	0	+	0	+	+		4+	4+	4+
3	Immucor 37832	B11193 R1R1 #5	+	+	0	0	+			0	+	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	4+
4	Immucor 37832	C5066 R2R2 #7	+	0	+	+	0			0	0	+	+	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	4+
5	Immucor 37832	C7328 R2R2 #9	+	0	+	+	0			0	0	+	0	+	0	+	+	+	+	0	+	0	+	+	+	+	0	+	0	+	0		4+	4+	4+
6	Immucor 37832	R2421 R1r #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	+	0	+	+	+	+	+	0	0	+	+		4+	4+	4+
7	Immucor 37832	E967 r'r #11	0	+	0	+	+			0	+	+	+	0	+	0	+	0	+	0	+	0	+	0	+	+	+	+	0	+	0		4+	4+	4+
8	Immucor 37832	F956 r"r #12	0	0	+	+	+			0	+	+	0	+	+	0	+	0	+	0	+	0	+	+	+	+	+	+	0	+	+		4+	4+	4+
9	Immucor 37832	F1079 r"r #15	0	0	+	+	+			0	+	+	0	+	+	0	0	0	+	0	+	0	+	+	+	+	+	+	0	+	+		4+	4+	4+
10	Immucor 37832	N5173 rr #16	0	0	0	+	+			0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	0	+	+	0	+	+		4+	4+	4+
11	Immucor 37832	D563 Ror #20	+	0	0	+	+			0	0	+	+	+	0	0	+	0	+	0	+	+	+	0	0	+	0	0	+	0	He+	4+	4+	4+
	Auto Control																															0	0	2+

Initial panel: PeG / AHG

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Missouri and Arkansas Region

	Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
			D	C	E	c	e	f	V	C^w	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		PeG AHG		
1	Immucor 37832	B3785 R1wR1 #1	+	+	0	0	+			+	0	+	0	+	0	+	+	+	+	0	+	0	+	0	+	+	+	0	+	+		4+		
2	Immucor 37832	B9783 R1R1 #3	+	+	0	0	+			0	0	+	+	+	+	w	0	0	+	0	+	0	+	+	0	0	+	0	+	+		4+		
3	Immucor 37832	B11193 R1R1 #5	+	+	0	0	+			0	+	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	0	+	+		3+		
4	Immucor 37832	C5066 R2R2 #7	+	0	+	+	0			0	0	+	+	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	+	+		4+		
5	Immucor 37832	C7328 R2R2 #9	+	0	+	+	0			0	0	+	0	+	0	+	+	+	+	0	+	0	+	+	+	0	+	0	+	0		4+		
6	Immucor 37832	R2421 R1r #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	+	0	+	+	+	+	0	0	+	+		4+		
7	Immucor 37832	E967 r'r #11	0	+	0	+	+			0	+	+	+	0	+	0	+	0	+	0	+	0	+	+	+	0	+	+	0	+	0		4+		
8	Immucor 37832	F956 r"r #12	0	0	+	+	+			0	+	+	0	+	+	0	+	0	+	0	+	0	+	+	+	+	+	+	0	+	+		4+		
9	Immucor 37832	F1079 r"r #15	0	0	+	+	+			0	+	+	0	+	+	0	0	0	+	0	+	0	+	+	+	+	+	+	0	+	+		4+		
10	Immucor 37832	N5173 rr #16	0	0	0	+	+			0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	+	+	0	+	+		4+		
11	Immucor 37832	D563 Ror #20	+	0	0	+	+			0	0	+	+	+	0	0	+	0	+	0	+	+	+	0	0	+	0	0	+	0	He+	4+		
	Auto Control																														0√		

Initial panel: LISS/37°C + LISS/AHG

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Missouri and Arkansas Region

	Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
			D	C	E	c	e	f	V	C^w	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		LISS 37C	LISS AHG	
1	Immucor 37832	B3785 R1wR1 #1	+	+	0	0	+			+	0	+	0	+	0	+	+	+	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	
2	Immucor 37832	B9783 R1R1 #3	+	+	0	0	+			0	0	+	+	+	+	w	0	0	+	0	+	0	+	+	0	0	+	0	+	+		4+	4+	
3	Immucor 37832	B11193 R1R1 #5	+	+	0	0	+			0	+	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	3+	
4	Immucor 37832	C5066 R2R2 #7	+	0	+	+	0			0	0	+	+	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	
5	Immucor 37832	C7328 R2R2 #9	+	0	+	+	0			0	0	+	0	+	0	+	+	+	+	0	+	0	+	+	+	0	+	0	+	0		4+	4+	
6	Immucor 37832	R2421 R1r #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	+	0	+	+	+	+	0	0	+	+		4+	4+	
7	Immucor 37832	E967 r'r #11	0	+	0	+	+			0	+	+	+	0	+	0	+	0	+	0	+	0	+	+	+	0	+	+	0	+	0		4+	4+	
8	Immucor 37832	F956 r"r #12	0	0	+	+	+			0	+	+	0	+	+	0	+	0	+	0	+	0	+	+	+	+	+	+	0	+	+		4+	4+	
9	Immucor 37832	F1079 r"r #15	0	0	+	+	+			0	+	+	0	+	+	0	0	0	+	0	+	0	+	+	+	+	+	+	0	+	+		4+	4+	
10	Immucor 37832	N5173 rr #16	0	0	0	+	+			0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	
11	Immucor 37832	D563 Ror #20	+	0	0	+	+			0	0	+	+	+	0	0	+	0	+	0	+	+	+	0	0	+	0	0	+	0	He+	4+	4+	
	Auto Control																															0	0%	

Initial possibilities

Consider common problems, presenting unusually

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- Auto-anti-H?
  - A_1 patients have minimal H substance on their red cells- autoantibody mimicking alloantibody
    - $O > A_2 > B > A_2B > A_1 > A_1B$ [1]
  - Evidence for: 2+ AC reaction at 4°C
  - Try XM vs random A units
    - Assuming at least one is type A_1, should be compatible if anti-H

Initial possibilities

Consider common problems, presenting unusually

Testing performed		
CROSSMATCH	IS	PeG AHG
Random A pos units		
W181124168187	4+	4+
W181124147815	4+	4+

Initial possibilities

Consider common problems, presenting unusually

- Auto-anti-H?
  - A_1 patients have minimal H substance on their red cells- autoantibody mimicking alloantibody
    - $O > A_2 > B > A_2B > A_1 > A_1B$ [1]
  - Evidence for: 2+ AC reaction at 4°C
  - Evidence against: both units reactive
    - Possible both units were A_2
    - Remember reverse type discrepancy

A_1 cells	A_2 cells	B cells
4+	4+	4+

More possibilities

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Make a list

- ~~Auto-anti-H~~
  - Evidence for: 2+ AC reaction at 4°C
  - Evidence against: 4+ reactive with A_1 cells
- Multiple cold-reactive common alloantibodies, unrelated auto-anti-I/H at 4°C?
  - Evidence for: 3+ reaction at AHG vs other 4+
- Allo-anti-I, unrelated auto-anti-H at 4°C?
  - Evidence for: 4+ reactions including IS, negative AC and DAT
- Cold-reactive antibody to a high prevalence antigen, unrelated auto-anti-I/H at 4°C?
  - Evidence for: 4+ reactions including IS, negative AC and DAT
- Atypical HTLA-like antibody, unrelated auto-anti-I/H at 4°C?
  - Evidence for: none

Papain treated adsorption cells, plasma adsorbed 1x @ 4°C

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	Rh-Hr								MNS				Lewis		P	Kell				Duffy		Kidd		Luth		Additional antigens	Test Results		
	D	C	E	c	e	f	V	C^W	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Js^a	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		IS	LISS 37C	LISS AHG
R_1R_1 W18112413651500*	+	+	0	0	+				0	+	0	+	+	0	+	0				0	+	+	+						
R_2R_2 W18112414520200H	+	0	+	+	0				+	0	+	0	0	0	+	0				+	0	+	0						
rr W18112411972300*	0	0	0	+	+				+	0	+	+	0	+	+	0				0	+	0	+						
R_1R_1																													
B 9440011-00 304320 #2	+	0	+	+	0	0		0	+	0	+	0	+	0	s	0	+	0	0	+	0	+	0	+	+		4+	4+	3+
W 37813 G478 #7	0	0	0	+	+			0	+	0	+	0	0	+	0	+	+	0	0	0	+	+	+	0	+				
R_2R_2																													
W 37832 B3785 #1	+	+	0	0	+			0	0	+	0	+	0	+	+	+	+	0	0	0	+	+	+	0	+		4+	4+	4+
B 9440011-00 305405 #7	0	0	0	+	+	+		0	0	+	0	+	0	+	+	+	+	0	0	+	0	0	+	0	w				
rr																													
B 9440011-00 305606 #1	+	+	0	0	+	0		0	+	+	0	+	+	0	0	0	+	0	0	+	0	+	0	0	+		4+	4+	4+
W 37832 R2421 #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	0	+	+	+	0	0	+				

Papain treated adsorption cells, plasma adsorbed 1x @ 4°C

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Target antigen(s) destroyed by papain treatment?

	Rh-Hr								MNS				Lewis		P	Kell				Duffy		Kidd		Luth		Additional antigens	Test Results		
	D	C	E	c	e	f	V	C^w	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Js^a	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		IS	LISS 37C	LISS AHG
R_1R_1 W18112413651500*	+	+	0	0	+				0	+	0	+	+	0	+	0				0	+	+	+						
R_2R_2 W18112414520200H	+	0	+	+	0				+	0	+	0	0	0	+	0				+	0	+	0						
rr W18112411972300*	0	0	0	+	+				+	0	+	+	0	+	+	0				0	+	0	+						
R_1R_1																													
B 9440011-00 304320 #2	+	0	+	+	0	0		0	+	0	+	0	+	0	s	0	+	0	0	+	0	+	0	+	+		4+	4+	3+
W 37813 G478 #7	0	0	0	+	+			0	+	0	+	0	0	+	0	+	+	0	0	0	+	+	+	0	+				
R_2R_2																													
W 37832 B3785 #1	+	+	0	0	+			0	0	+	0	+	0	+	+	+	+	0	0	0	+	+	+	0	+		4+	4+	4+
B 9440011-00 305405 #7	0	0	0	+	+	+		0	0	+	0	+	0	+	+	+	+	0	0	+	0	0	+	0	w				
rr																													
B 9440011-00 305606 #1	+	+	0	0	+	0		0	+	+	0	+	+	0	0	0	+	0	0	+	0	+	0	0	+		4+	4+	4+
W 37832 R2421 #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	0	+	+	+	0	0	+				

Papain treated adsorption cells, plasma adsorbed 1x @ 4°C
Untreated adsorption cells, plasma adsorbed 1x @ 4°C

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	Rh-Hr								MNS				Lewis		P	Kell				Duffy		Kidd		Luth		Additional antigens	Test Results		
	D	C	E	c	e	f	V	C^w	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Js^a	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		IS	LISS 37C	LISS AHG
R_1R_1 W18112413651500*	+	+	0	0	+				0	+	0	+	+	0	+	0				0	+	+	+						
R_2R_2 W18112414520200H	+	0	+	+	0				+	0	+	0	0	0	+	0				+	0	+	0						
rr W18112411972300*	0	0	0	+	+				+	0	+	+	0	+	+	0				0	+	0	+						
R_1R_1																													
B 9440011-00 304320 #2	+	0	+	+	0	0		0	+	0	+	0	+	0	s	0	+	0	0	+	0	+	0	+	+		0	0	0/0√
W 37813 G478 #7	0	0	0	+	+			0	+	0	+	0	0	+	0	+	+	0	0	0	+	+	+	0	+		0	0	0/0√
R_2R_2																													
W 37832 B3785 #1	+	+	0	0	+			0	0	+	0	+	0	+	+	+	+	0	0	0	+	+	+	0	+		0	0	0/m+
B 9440011-00 305405 #7	0	0	0	+	+	+		0	0	+	0	+	0	+	+	+	+	0	0	+	0	0	+	0	w				
rr																													
B 9440011-00 305606 #1	+	+	0	0	+	0		0	+	+	0	+	+	0	0	0	+	0	0	+	0	+	0	0	+		0	0	0/m+
W 37832 R2421 #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	0	+	+	+	0	0	+				

Papain treated adsorption cells, plasma adsorbed 1x @ 4°C
Untreated adsorption cells, plasma adsorbed 2x @ 4°C

American Red Cross
Missouri and Arkansas Region

	Rh-Hr								MNS				Lewis		P	Kell				Duffy		Kidd		Luth		Additional antigens	Test Results		
	D	C	E	c	e	f	V	C^w	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Js^a	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		IS	LISS 37C	LISS AHG
R_1R_1 W18112413651500*	+	+	0	0	+				0	+	0	+	+	0	+	0				0	+	+	+						
R_2R_2 W18112414520200H	+	0	+	+	0				+	0	+	0	0	0	+	0				+	0	+	0						
rr W18112411972300*	0	0	0	+	+				+	0	+	+	0	+	+	0				0	+	0	+						
R_1R_1																													
B 9440011-00 304320 #2	+	0	+	+	0	0		0	+	0	+	0	+	0	s	0	+	0	0	+	0	+	0	+	+		0	0	0/0 √
W 37813 G478 #7	0	0	0	+	+			0	+	0	+	0	0	+	0	+	+	0	0	0	+	+	+	0	+		0	0	0/0 √
R_2R_2																													
W 37832 B3785 #1	+	+	0	0	+			0	0	+	0	+	0	+	+	+	+	0	0	0	+	+	+	0	+		0	0	0/0 √
B 9440011-00 305405 #7	0	0	0	+	+	+		0	0	+	0	+	0	+	+	+	+	0	0	+	0	0	+	0	w		0	0	0/0 √
rr																													
B 9440011-00 305606 #1	+	+	0	0	+	0		0	+	+	0	+	+	0	0	0	+	0	0	+	0	+	0	0	+		0	0	0/0 √
W 37832 R2421 #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	0	+	+	+	0	0	+		0	0	0/0 √

Papain treated adsorption cells, plasma adsorbed 1x @ 4°C
Untreated adsorption cells, plasma adsorbed 2x @ 4°C

American Red Cross
Missouri and Arkansas Region

	Rh-Hr								MNS				Lewis		P	Kell				Duffy		Kidd		Luth		Additional antigens	Test Results		
	D	C	E	c	e	f	V	C^W	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Js^a	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		IS	LISS 37C	LISS AHG
R_1R_1 W18112413651500*	+	+	0	0	+				0	+	0	+	+	0	+	0				0	+	+	+						
R_2R_2 W18112414520200H	+	0	+	+	0				+	0	+	0	0	0	+	0				+	0	+	0						
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B 9440011-00 304320 #2	+	0	~~+~~	~~+~~	0	0		0	~~+~~	0	~~+~~	0	+	0	s	0	+	0	0	~~+~~	0	+	0	+	+		0	0	0/0 √
W 37813 G478 #7	0	0	0	+	+			0	+	0	+	0	0	~~+~~	0	+	+	0	0	0	+	+	+	0	+		0	0	0/0 √
R_2R_2																													
W 37832 B3785 #1	+	~~+~~	0	0	~~+~~			0	0	~~+~~	0	~~+~~	0	+	+	+	+	0	0	0	~~+~~	+	+	0	+		0	0	0/0 √
B 9440011-00 305405 #7	0	0	0	+	+	~~+~~		0	0	+	0	+	0	+	+	+	+	0	0	+	0	0	~~+~~	0	w		0	0	0/0 √
rr																													
B 9440011-00 305606 #1	~~+~~	+	0	0	+	0		0	+	+	0	+	~~+~~	0	0	0	+	0	0	+	0	+	0	0	+		0	0	0/0 √
W 37832 R2421 #10	+	+	0	+	+			0	+	+	+	+	0	+	+	~~+~~	0	0	0	+	+	+	0	0	+		0	0	0/0 √

Confirming enzyme sensitivity

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Lewis		P	Kell							Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^w	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b		Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		FIC AHG		
Imm-10F 35799	B7367 R1R1 #1	+	+	0	0	+			0	+	+	0	+	0	+	+	+	0	0	+	0	+		0	+	+	0	0	+	+	Co(b+)	0√		
Imm-10F 35799	C3496 R2R2 #3	+	0	+	+	0			0	+	+	+	+	+	0	0	0	+	0	+	0	+		+	+	+	+	0	+	+		0√		
Imm-10F 35799	H642 rr #8	0	0	0	+	+			0	+	0	+	+	0	+	+	0	+	0	+	0	+		+	0	+	0	0	+	+	Co(b+)	0√		
Auto Control																																		

Blood type

American Red Cross
Missouri and Arkansas Region

Anti-A	Anti-B	Anti-A,B	Anti-D	Rh control	A_1 cells	A_2 cells	B cells
4+	0	4+	3+	0	4+	4+	4+

Type: NTD positive

Blood type with adsorbed plasma

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Missouri and Arkansas Region

Anti-A	Anti-B	Anti-A,B	Anti-D	Rh control	A_1 cells	A_2 cells	B cells
4+	0	4+	3+	0	0	0	3+

Type: A positive

Fewer possibilities

American Red Cross
Missouri and Arkansas Region

Make a list, cross things off

- ~~Auto-anti-H~~
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  - Evidence for: 4+ reactions including IS, negative AC and DAT
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Missouri and Arkansas Region

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  - Evidence against: adsorption should have been enhanced with papain treated adsorbing cells
- Cold-reactive antibody to a high prevalence antigen, unrelated auto-anti-I/H at 4°C?
  - Evidence for: 4+ reactions including IS, negative AC and DAT; adsorbed out, no underlying
  - Evidence against: none
- Atypical HTLA-like antibody, unrelated auto-anti-I/H at 4°C?
  - Evidence for: none

Fewer possibilities

American Red Cross
Missouri and Arkansas Region

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American Red Cross
Missouri and Arkansas Region

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  - Evidence for: 4+ reactions including IS, negative AC and DAT; adsorbed out, no underlying
  - Evidence against: none
- Atypical HTLA-like antibody, unrelated auto-anti-I/H at 4°C?
  - Evidence for: none
  - Evidence against: strong reactivity, adsorbed easily

Fewer possibilities

American Red Cross
Missouri and Arkansas Region

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  - Evidence for: none
  - Evidence against: strong reactivity, adsorbed easily

Fewer possibilities

American Red Cross
Missouri and Arkansas Region

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  - Evidence for: none
  - Evidence against: strong reactivity, adsorbed easily

Investigating an antibody to a high prevalence antigen

American Red Cross
Missouri and Arkansas Region

Gather information, list what you know

- What do we know?
  - Patient is of African descent
    - Possible direction for high prevalence antigens
  - Antibody is reactive at wide thermal range- strong IgM component
    - Reactive at IS, RT, 4°C, 37°C/LISS, LISS/AHG, PeG/AHG
    - Can be adsorbed out at 4°C
  - Antigen sensitive to ficin and papain treatment
    - Confirmed with neat plasma + enzyme treated reagent cells

Investigating an antibody to a high prevalence antigen

American Red Cross
Missouri and Arkansas Region

Make another list [2]

- ~~Allo-anti-I~~
- Anti-Pr
- Anti-Vel
- Anti-PP1Pk
- Anti-En^{a}
- Various antibodies to high prevalence Rh antigens
- Anti-Fy3
- Anti-Js^{b}
- Anti-U
- Anti-Cr^{a}
- Anti-At^{a}
- Anti-Hy
- Anti-Jo^{a}
- Anti-CD47

Investigating an antibody to a high prevalence antigen

American Red Cross
Missouri and Arkansas Region

Make another list [2], cross things off

- ~~Allo-anti-I~~ : usually IgM, antigen enhanced by papain
- Anti-Pr : usually IgM, antigen sensitive to papain, always described as autoantibody[3]
- Anti-Vel : usually IgM/IgG mix, antigen enhanced by papain
- Anti-PP1Pk : usually IgM and IgG, antigen enhanced by papain
- Anti-En^{a} : usually IgM and IgG, some regions of antigen enhanced, some sensitive to papain, incredibly rare
- Various antibodies to high prevalence Rh antigens : usually more IgG than IgM, antigens enhanced by papain
- Anti-Fy3 : usually IgG, antigen resistant to papain
- Anti-Js^{b} : usually IgG, antigen resistant to papain
- Anti-U : usually IgG, antigen resistant to papain
- Anti-Cr^{a} : usually IgG, antigen resistant to papain
- Anti-At^{a} : usually IgG, antigen resistant to papain
- Anti-Hy : usually IgG, antigen resistant to papain
- Anti-Jo^{a} : usually IgG, antigen resistant to papain
- Anti-CD47 : doesn't adsorb out cleanly, should have been less or nonreactive at AHG with Werfen AHG

Investigating an antibody to a high prevalence antigen

American Red Cross
Missouri and Arkansas Region

Make another list [2], cross things off

- ~~Allo-anti-I~~ : usually IgM, antigen enhanced by papain
- ~~Anti-Pr~~ : usually IgM, antigen sensitive to papain, always described as autoantibody[3]
- ~~Anti-Vel~~ : usually IgM/IgG mix, antigen enhanced by papain
- ~~Anti-PP1Pk~~ : usually IgM and IgG, antigen enhanced by papain
- ~~Anti-En^{a}~~ : usually IgM and IgG, some regions of antigen enhanced, some sensitive to papain, incredibly rare
- ~~Various antibodies to high prevalence Rh antigens~~ : usually more IgG than IgM, antigens enhanced by papain
- ~~Anti-Fy3~~ : usually IgG, antigen resistant to papain
- ~~Anti-Js^{b}~~ : usually IgG, antigen resistant to papain
- ~~Anti-U~~ : usually IgG, antigen resistant to papain
- ~~Anti-Cr^{a}~~ : usually IgG, antigen resistant to papain
- ~~Anti-At^{a}~~ : usually IgG, antigen resistant to papain
- ~~Anti-Hy~~ : usually IgG, antigen resistant to papain
- ~~Anti-Jo^{a}~~ : usually IgG, antigen resistant to papain
- ~~Anti-CD47~~ : doesn't adsorb out cleanly, should have been less or nonreactive at AHG with Werfen AHG

Additional history

If the history is unclear, start investigating

- Contacted hospital, requested additional history
  - Patient stated:
    - History of multiple outpatient surgeries, but never transfused
    - Recalled being told she had anti-N
    - Recalled being told she “had a unique blood issue”
  - Planned surgery was total knee arthroplasty
    - May require transfusion intra- or post-operatively

- Does this make sense?
  - No evidence of anti-N in adsorption
  - No history of dialysis (anti-Nf)

Knee joint replacement - series—Procedure https://medlineplus.gov/ency/presentations/100088_3.htm

Phenotype

American Red Cross
Missouri and Arkansas Region

C	E	c	e	K	Fy^a	Fy^b	Jk^a	Jk^b	M	N	S	s	Le^a	Le^b	P1

Phenotype

American Red Cross
Missouri and Arkansas Region

C	E	c	e	K	Fy^a	Fy^b	Jk^a	Jk^b	M	N	S	s	Le^a	Le^b	P1
						+	+	+							

Unlikely anti-Fy3

Phenotype

American Red Cross
Missouri and Arkansas Region

C	E	c	e	K	Fy^a	Fy^b	Jk^a	Jk^b	M	N	S	s	Le^a	Le^b	P1
0	0	+	+			+	+	+							

Unlikely some Rh antibodies

Unlikely anti-Fy3

Phenotype

American Red Cross
Missouri and Arkansas Region

C	E	c	e	K	Fy^a	Fy^b	Jk^a	Jk^b	M	N	S	s	Le^a	Le^b	P1
0	0	+	+			+	+	+	+	0			+	0	+

Unlikely some Rh antibodies

Unlikely anti-Fy3

GPA present, unlikely En(a-) or M^kM^k phenotype

Unlikely anti-PP1Pk

Phenotype

C	E	c	e	K	Fy^a	Fy^b	Jk^a	Jk^b	M	N	S	s	Le^a	Le^b	P1
0	0	+	+	0	0	+	+	+	+	0	0	0	+	0	+

Unlikely some Rh antibodies

Unlikely anti-Fy3

GPA present, unlikely En(a-) or M^kM^k phenotype

Unlikely anti-PP1Pk

Phenotype: S- s-

- Anti-U?
  - usually IgG, antigen resistant to papain

Additional results

American Red Cross
Missouri and Arkansas Region

Test with trypsin, 0.2M DTT treated cells

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^w	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		FIC AHG	TRY AHG	DTT PeG AHG
Imm-10F 35799	B7367 R1R1 #1	+	+	0	0	+			0	+	+	0	+	0	+	+	+	0	0	+	0	+	0	+	+	0	+	0	+	+	Co(b+)	0√	4+	4+
Imm-10F 35799	C3496 R2R2 #3	+	0	+	+	0			0	+	+	+	+	+	0	0	0	+	0	+	0	+	+	+	+	+	0	+	+		0√	4+	4+
Imm-10F 35799	H642 rr #8	0	0	0	+	+			0	+	0	+	+	0	+	+	0	+	0	+	0	+	+	0	+	0	0	+	+	Co(b+)	0√	4+	4+
Auto Control																																	

Investigating an antibody to a high prevalence antigen

American Red Cross
Missouri and Arkansas Region

Gather information, list what you know

- Now what do we know?
  - Patient is of African descent
    - Possible direction for high prevalence antigens
  - Antibody is reactive at wide thermal range- strong IgM component
    - Reactive at IS, RT, 4°C, 37°C/LISS, LISS/AHG, PeG/AHG
    - Can be adsorbed out at 4°C
  - Antigen sensitive to ficin and papain treatment
    - Confirmed with neat plasma + enzyme treated reagent cells
  - Antigen resistant to trypsin and DTT treatment
  - S- s- probably relevant?

Investigating an antibody to a high prevalence antigen

American Red Cross
Missouri and Arkansas Region

Gather information, list what you know

- Now what do we know?
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  - Antigen resistant to trypsin and DTT treatment
  - S- s- probably relevant?

“…history of anti-N…”

“…unique blood issue…”

Academic interlude

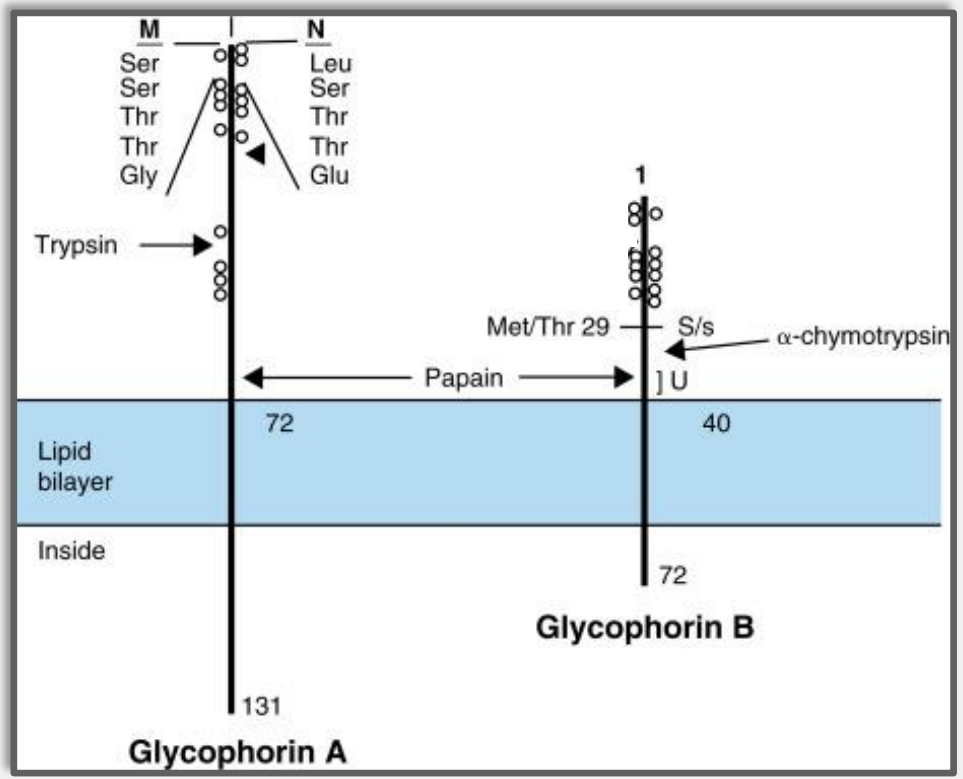
Anti-N

American Red Cross
Missouri and Arkansas Region

What is this antibody?

- Rare antibody [2]
  - Usually IgM
  - Antigen destroyed by papain and trypsin, resistant to DTT
  - Not generally considered clinically significant
  - Classically, found in dialysis patients
    - Anti-Nf
    - Due to use of formaldehyde sterilized equipment

- <u>Was not seen in adsorbed plasma</u>



Judith Aeschlimann, Connie M. Westhoff, Chapter 28 - MNS and Duffy Blood Group Systems, *Transfusion Medicine and Hemostasis (Third Edition)*, Elsevier, 2019

Papain treated adsorption cells, plasma adsorbed 1x @ 4°C
Untreated adsorption cells, plasma adsorbed 2x @ 4°C

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	Rh-Hr								MNS				Lewis		P	Kell				Duffy		Kidd		Luth		Additional antigens	Test Results		
	D	C	E	c	e	f	V	C^w	M	**N**	S	s	Le^a	Le^b	P1	K	k	Kp^a	Js^a	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		IS	LISS 37C	LISS AHG
R_1R_1 W18112413651500*	+	+	0	0	+				0	+	0	+	+	0	+	0				0	+	+	+						
R_2R_2 W18112414520200H	+	0	+	+	0				+	**0**	+	0	0	0	+	0				+	0	+	0						
rr W18112411972300*	0	0	0	+	+				+	**0**	+	+	0	+	+	0				0	+	0	+						
R_1R_1																													
B 9440011-00 304320 #2	+	0	+	+	0	0		0	+	0	+	0	+	0	s	0	+	0	0	+	0	+	0	+	+		0	0	0/0√
W 37813 G478 #7	0	0	0	+	+			0	+	0	+	0	0	+	0	+	+	0	0	0	+	+	+	0	+		0	0	0/0√
R_2R_2																													
W 37832 B3785 #1	+	+	0	0	+			0	0	**+**	0	+	0	+	+	+	+	0	0	0	+	+	+	0	+		**0**	**0**	**0/0√**
B 9440011-00 305405 #7	0	0	0	+	+	+		0	0	**+**	0	+	0	+	+	+	+	0	0	+	0	0	+	0	w		**0**	**0**	**0/0√**
rr																													
B 9440011-00 305606 #1	+	+	0	0	+	0		0	+	**+**	0	+	+	0	0	0	+	0	0	+	0	+	0	0	+		**0**	**0**	**0/0√**
W 37832 R2421 #10	+	+	0	+	+			0	+	**+**	+	+	0	+	+	+	0	0	0	+	+	+	0	0	+		**0**	**0**	**0/0√**

‘N’ antigen

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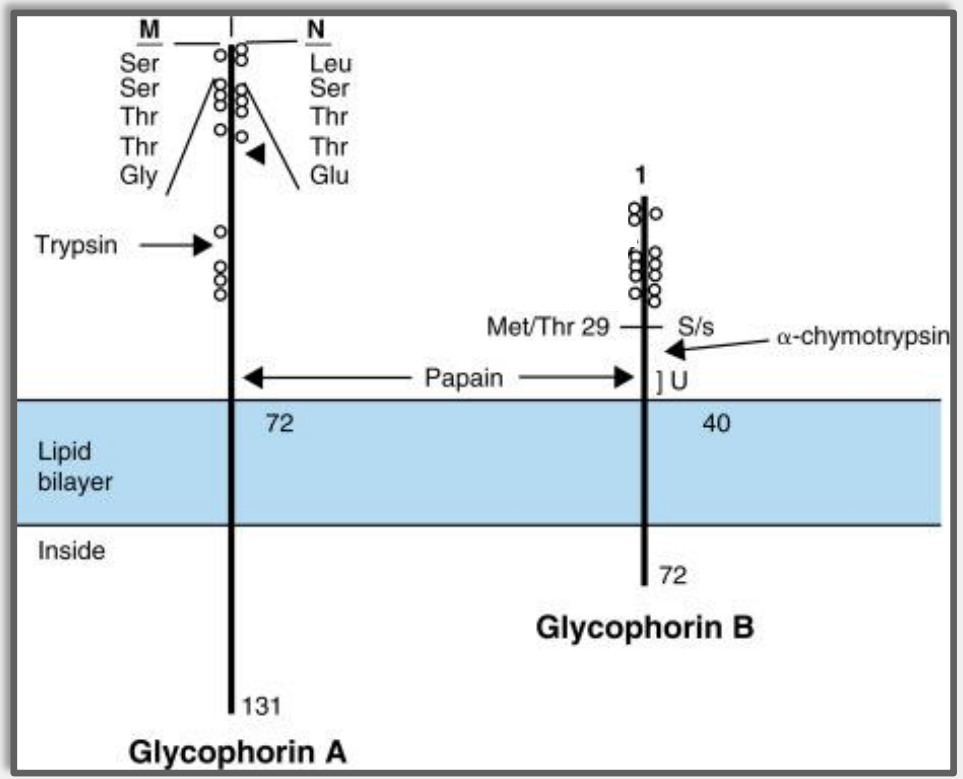
Missouri and Arkansas Region

What is like N, but not N?

- First 26 amino acids of GPB identical to N antigenic form of GPA [2, 4]
- A second N on GPB!
- Distinguished from GPA N as ‘N’
- People with normal/intact GPB are ‘N’ positive [5]
  - Anti-N from N- ‘N’+ rare due to immune tolerance
    - Does not normally react with ‘N’ largely due to steric factors, number of antigenic sites, possible low binding affinity
- Reagents formulated to only detect N, not ‘N’ [6]
- Antigen is sensitive to papain treatment, resistant to trypsin and DTT treatment
- Fits what we know about our patient’s antibody



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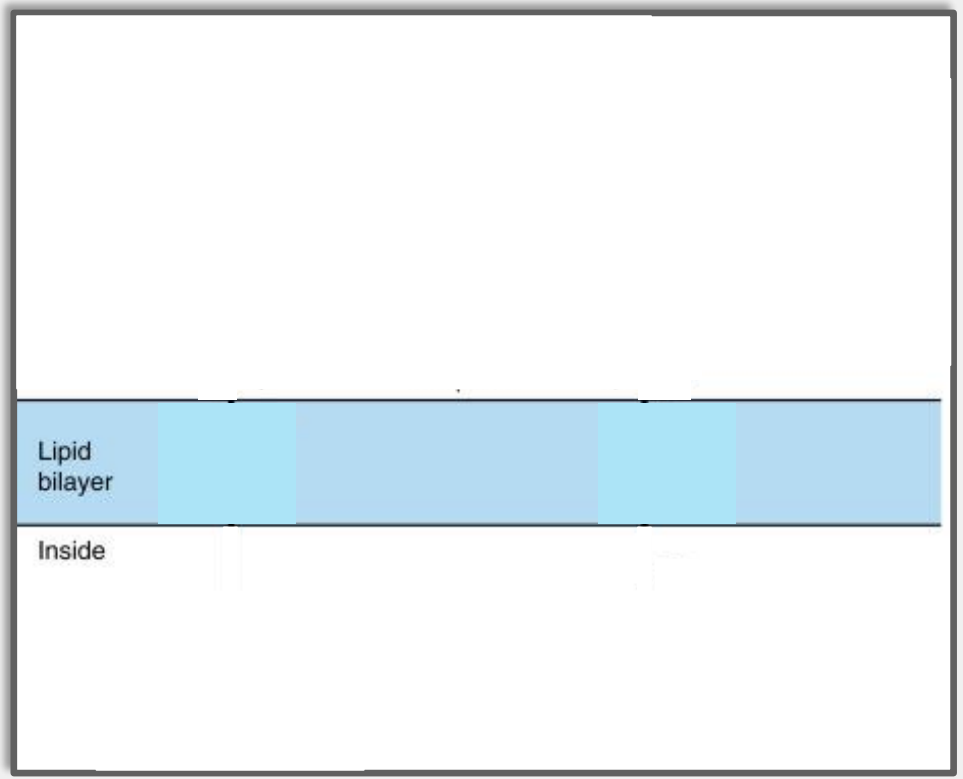
‘N’ antigen

What is like N, but not N?

- Usually absent on S- s- cells [2, 3]
  - U_{var}
  - U –
  - M^kM^k phenotype
    - GPA and GPB absent



(some liberties taken in visually representing the listed phenotypes)

Judith Aeschlimann, Connie M. Westhoff, Chapter 28 - MNS and Duffy Blood Group Systems, *Transfusion Medicine and Hemostasis (Third Edition)*, Elsevier, 2019

‘N’ antigen

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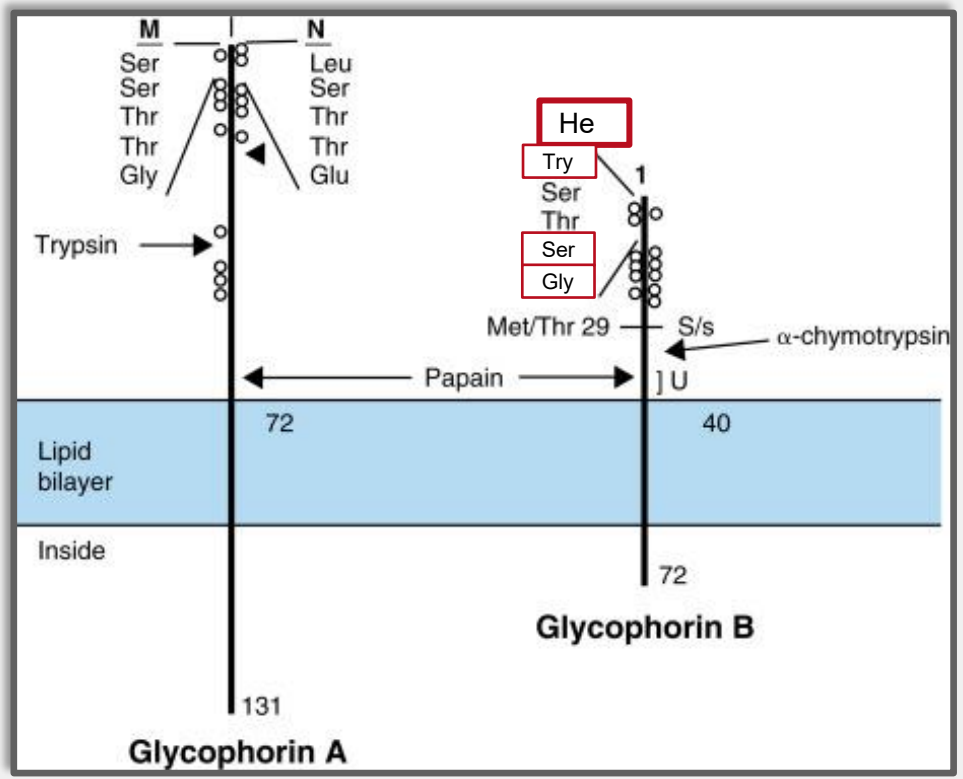
’N’ variants

- Rare He antigen [6]
  - Antithetical to ‘N’, roughly 3% prevalence in Black Americans
  - He can be present on S+ and/or s+ cells, as well as S- s- U_{var} or U- cells [9]
    - Roughly 70+% of S- s- U_{var} reacting with anti-U/GPB, are He+
    - Remainder are ‘N’+

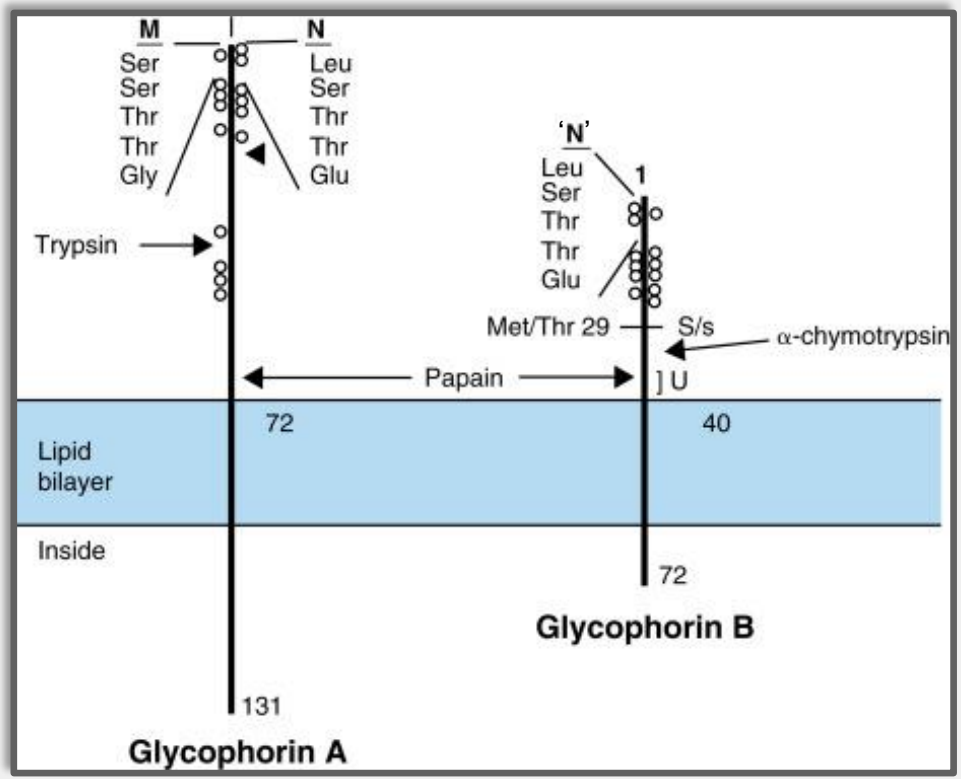


Judith Aeschlimann, Connie M. Westhoff, Chapter 28 - MNS and Duffy Blood Group Systems, *Transfusion Medicine and Hemostasis (Third Edition)*, Elsevier, 2019

Anti-N (from N- ‘N’-)

Antibody characteristics

- No human anti-’N’ identified [6]
  - If anti-N visibly reacts with N- ‘N’+ cells, every example found also reacts with N+ ‘N’- cells
- No simple term to distinguish regular anti-N, vs anti-N from N- ‘N’- people
- Anti-N from N- ‘N’- individuals can cause HTR [7]
  - ^{51}Cr labeled N+ cells showed decreased *in vivo* survival in one patient studied
  - MMA predicted clinically significant
- Minimal reports of transfusing antigen positive blood to these patients [6]



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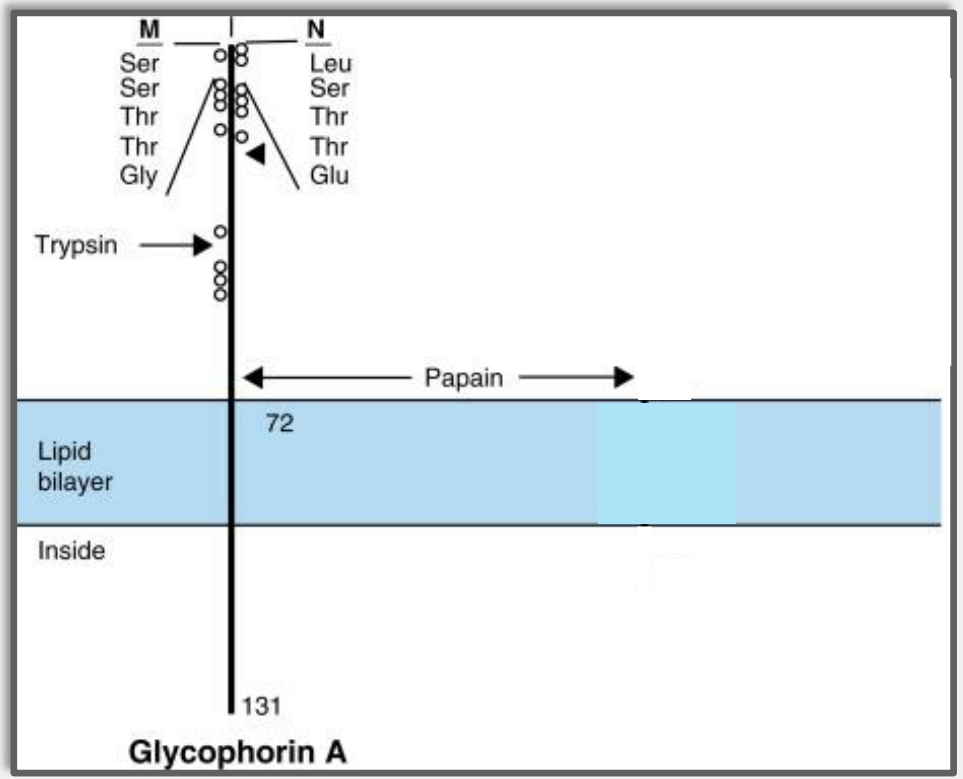
Transfusion considerations

- M+ N- S- s- U- [8]
  - Most straightforward way to ensure ‘N’-
- M+ N- S- s- U_{var} [8]
  - Majority are ‘N’-



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Back to patient testing

Evaluate previous results- do they help confirm antibody ID?

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Untreated cells

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS						Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^W	M	**N**	S	s	**'N'**	**U**	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a				
Imm-10F 35799	B7367 R1R1 #1	+	+	0	0	+			0	+	**+**	0	+	**+**	**+**	0	+	+	+	0	0	+	0	+	0	+	+	0	0	+	+	Co(b+)			
Imm-10F 35799	C3496 R2R2 #3	+	0	+	+	0			0	+	**+**	+	+	**+**	**+**	+	0	0	0	+	0	+	0	+	+	+	+	+	0	+	+				
Imm-10F 35799	H642 rr #8	0	0	0	+	+			0	+	**0**	+	+	**+**	**+**	0	+	+	0	+	0	+	0	+	+	0	+	0	0	+	+	Co(b+)			
Auto Control																																		

Evaluate previous results- do they help confirm antibody ID?

American Red Cross
Missouri and Arkansas Region

Ficin treated cells

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS						Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^W	M	**N**	S	s	**'N'**	**U**	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		**FIC** **AHG**		
Imm-10F 35799	B7367 R1R1 #1	+	+	0	0	+			0	0	**0**	0	0	**0**	**+**	0	+	+	+	0	0	+	0	+	0	0	+	0	0	+	0	Co(b+)	**0√**		
Imm-10F 35799	C3496 R2R2 #3	+	0	+	+	0			0	0	**0**	0	0	**0**	**+**	+	0	0	0	+	0	+	0	+	0	0	+	+	0	+	0		**0√**		
Imm-10F 35799	H642 rr #8	0	0	0	+	+			0	0	**0**	0	0	**0**	**+**	0	+	+	0	+	0	+	0	+	0	0	+	0	0	+	0	Co(b+)	**0√**		
Auto Control																																		

N and 'N' antigens destroyed

U antigen unaffected

Evaluate previous results- do they help confirm antibody ID?

American Red Cross
Missouri and Arkansas Region

Ficin treated cells

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS						Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^W	M	**N**	S	s	**'N'**	**U** ⚔	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		**FIC** **AHG**		
Imm-10F 35799	B7367 R1R1 #1	+	+	0	0	+			0	0	**0**	0	0	**0**	✗	0	+	+	+	0	0	+	0	+	0	0	+	0	0	+	0	Co(b+)	**0**√		
Imm-10F 35799	C3496 R2R2 #3	+	0	+	+	0			0	0	**0**	0	0	**0**	✗	+	0	0	0	+	0	+	0	+	0	0	+	+	0	+	0		**0**√		
Imm-10F 35799	H642 rr #8	0	0	0	+	+			0	0	**0**	0	0	**0**	✗	0	+	+	0	+	0	+	0	+	0	0	+	0	0	+	0	Co(b+)	**0**√		
Auto Control																																		

Able to rule out anti-U

Evaluate previous results- do they help confirm antibody ID?

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Trypsin treated cells

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS						Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^W	M	**N**	S	s	**'N'**	**U**	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		FIC AHG	**TRY** **AHG**	
Imm-10F 35799	B7367 R1R1 #1	+	+	0	0	+			0	0	**0**	0	+	**+**	✗	0	+	+	+	0	0	+	0	+	0	+	+	0	0	0	0	Co(b+)	0√	**4+**	
Imm-10F 35799	C3496 R2R2 #3	+	0	+	+	0			0	0	**0**	+	+	**+**	✗	+	0	0	0	+	0	+	0	+	+	+	+	+	0	0	0		0√	**4+**	
Imm-10F 35799	H642 rr #8	0	0	0	+	+			0	0	**0**	+	+	**+**	✗	0	+	+	0	+	0	+	0	+	+	0	+	0	0	0	0	Co(b+)	0√	**4+**	
Auto Control																																		

N antigen destroyed

'N' and U antigens unaffected

Evaluate previous results- do they help confirm antibody ID?

American Red Cross
Missouri and Arkansas Region

Trypsin treated cells

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS						Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^W	M	**N**	S	s	**'N'**	**U**	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		FIC AHG	**TRY** **AHG**	
Imm-10F 35799	B7367 R1R1 #1	+	+	0	0	+			0	0	**0**	0	+	+	≠	0	+	+	+	0	0	+	0	+	0	+	+	0	0	0	0	Co(b+)	0√	**4+**	
Imm-10F 35799	C3496 R2R2 #3	+	0	+	+	0			0	0	**0**	+	+	+	≠	+	0	0	0	+	0	+	0	+	+	+	+	+	0	0	0		0√	**4+**	
Imm-10F 35799	H642 rr #8	0	0	0	+	+			0	0	**0**	+	+	+	≠	0	+	+	0	+	0	+	0	+	+	0	+	0	0	0	0	Co(b+)	0√	**4+**	
Auto Control																																		

Anti-N reacting with N- 'N'+ cells

Evaluate previous results- do they help confirm antibody ID?

American Red Cross
Missouri and Arkansas Region

0.2M DTT treated cells

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS						Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^W	M	N	S	s	'N'	U	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		FIC AHG	TRY AHG	DTT PeG AHG
Imm-10F 35799	B7367 R1R1 #1	+	+	0	0	+			0	+	+	0	+	+	✗	0	+	+	0	0	0	0	0	0	0	+	+	0	0	0	+	Co(b+)	0√	4+	**4+**
Imm-10F 35799	C3496 R2R2 #3	+	0	+	+	0			0	+	+	+	+	+	✗	+	0	0	0	0	0	0	0	0	+	+	+	+	0	0	+		0√	4+	**4+**
Imm-10F 35799	H642 rr #8	0	0	0	+	+			0	+	+	+	+	+	✗	0	+	+	0	0	0	0	0	0	+	0	+	0	0	0	+	Co(b+)	0√	4+	**4+**
Auto Control																																		

N, 'N', and U antigens unaffected

Nothing additional ruled in or out

Final confirmation

Neat testing

American Red Cross
Missouri and Arkansas Region

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS						Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^W	M	**N**	S	s	**'N'**	**U**	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		IS	LISS 37C	LISS AHG
Imm-10 37813	D1387 Ror #TC	+	0	0	+	+			0	0	**+**	0	0	**0**	**0**	0	0	+	0	+	0	+	0	+	0	0	+	+	0	+	+	U-	**2+**	**3+**	**4+**
Imm-20 34796	C3723 R2R2 #8	+	0	+	+	0			0	+	**0**	0	0	**0**	**0**	0	+	+	0	+	0	+	0	+	0	+	+	+	0	+	+	U-	**0**	**0**	**0/0**
Auto Control																																		

Final confirmation

Neat testing

American Red Cross
Missouri and Arkansas Region

Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS						Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
		D	C	E	c	e	f	V	C^W	M	N	S	s	'N'	U	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		IS	LISS 37C	LISS AHG
Imm-10 37813	D1387 Ror #TC	+	0	0	+	+			0	0	+	0	0	**0**	**0**	0	0	+	0	+	0	+	0	+	0	0	+	+	0	+	+	U-	**2+**	**3+**	**4+**
Imm-20 34796	C3723 R2R2 #8	+	0	+	+	0			0	+	**0**	0	0	**0**	**0**	0	+	+	0	+	0	+	0	+	0	+	+	+	0	+	+	U-	**0**	**0**	**0/0**
Auto Control																																		

Reactive with N+ 'N'- cell, confirming N/'N' specificity

Nonreactive with neat N- 'N'- cell

A few interesting things to note

He+ cell tested- why did it react?

	Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Le wis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
			D	C	E	c	e	f	V	C^w	M	N	S	s	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		IS	LISS 37C	LISS AHG
1	Immucor 37832	B3785 R1wR1 #1	+	+	0	0	+			+	0	+	0	+	0	+	+	+	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	4+
2	Immucor 37832	B9783 R1R1 #3	+	+	0	0	+			0	0	+	+	+	+	w	0	0	+	0	+	0	+	+	0	0	+	0	+	+		4+	4+	4+
3	Immucor 37832	B11193 R1R1 #5	+	+	0	0	+			0	+	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	3+
4	Immucor 37832	C5066 R2R2 #7	+	0	+	+	0			0	0	+	+	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	4+
5	Immucor 37832	C7328 R2R2 #9	+	0	+	+	0			0	0	+	0	+	0	+	+	+	+	0	+	0	+	+	+	0	+	0	+	0		4+	4+	4+
6	Immucor 37832	R2421 R1r #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	+	0	+	+	+	+	0	0	+	+		4+	4+	4+
7	Immucor 37832	E967 r'r #11	0	+	0	+	+			0	+	+	+	0	+	0	+	0	+	0	+	0	+	+	0	+	+	0	+	0		4+	4+	4+
8	Immucor 37832	F956 r''r #12	0	0	+	+	+			0	+	+	0	+	+	0	+	0	+	0	+	0	+	+	+	+	+	0	+	+		4+	4+	4+
9	Immucor 37832	F1079 r''r #15	0	0	+	+	+			0	+	+	0	+	+	0	0	0	+	0	+	0	+	+	+	+	+	0	+	+		4+	4+	4+
10	Immucor 37832	N5173 rr #16	0	0	0	+	+			0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	4+
11	Immucor 37832	D563 Ror #20	+	0	0	+	+			0	0	+	+	+	0	0	+	0	+	0	+	+	+	0	0	+	0	0	+	0	0	He+	**4+**	**4+**	**4+**
	Auto Control																															0	0	0/0

A few interesting things to note

He+ cell tested- why did it react?

	Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
			D	C	E	c	e	f	V	C^w	M	**N**	S	s	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		IS	LISS 37C	LISS AHG
1	Immucor 37832	B3785 R1wR1 #1	+	+	0	0	+			+	0	+	0	+	0	+	+	+	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	4+
2	Immucor 37832	B9783 R1R1 #3	+	+	0	0	+			0	0	+	+	+	+	w	0	0	+	0	+	0	+	+	0	0	+	0	+	+		4+	4+	4+
3	Immucor 37832	B11193 R1R1 #5	+	+	0	0	+			0	+	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	3+
4	Immucor 37832	C5066 R2R2 #7	+	0	+	+	0			0	0	+	+	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	4+
5	Immucor 37832	C7328 R2R2 #9	+	0	+	+	0			0	0	+	0	+	0	+	+	+	+	0	+	0	+	+	0	+	0	+	0	+	0		4+	4+	4+
6	Immucor 37832	R2421 R1r #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	+	0	+	+	+	+	+	0	0	+	+		4+	4+	4+
7	Immucor 37832	E967 r'r #11	0	+	0	+	+			0	+	+	+	0	+	0	+	0	+	0	+	0	+	+	0	+	+	+	0	+	0		4+	4+	4+
8	Immucor 37832	F956 r''r #12	0	0	+	+	+			0	+	+	0	+	+	0	+	0	+	0	+	0	+	+	+	+	+	0	+	+		4+	4+	4+
9	Immucor 37832	F1079 r''r #15	0	0	+	+	+			0	+	+	0	+	+	0	0	0	+	0	+	0	+	+	+	+	+	0	+	+		4+	4+	4+
10	Immucor 37832	N5173 rr #16	0	0	0	+	+			0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	4+
11	Immucor 37832	D563 Ror #20	+	0	0	+	+			0	0	+	+	+	0	0	+	0	+	0	+	+	+	+	0	0	+	0	0	+	0	He+	4+	4+	4+
	Auto Control																															0	0	0/0

- Cell is N+
- Likely heterozygous for *He* allele
  - If so, 'N' also present

A few interesting things to note

Weaker reaction at LISS/AHG on N- cell

	Supplier/ Lot	Donor / RhHr - Vial	Rh-Hr								MNS				Lewis		P	Kell						Duffy		Kidd		Luth		X	Additional antigens	Test Results		
			D	C	E	c	e	f	V	C^w	M	**N**	S	s	Le^a	Le^b	P1	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b	Xg^a		IS	LISS 37C	LISS AHG
1	Immucor 37832	B3785 R1wR1 #1	+	+	0	0	+			+	0	+	0	+	0	+	+	+	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	4+
2	Immucor 37832	B9783 R1R1 #3	+	+	0	0	+			0	0	+	+	+	+	w	0	0	+	0	+	0	+	+	0	0	+	0	+	+		4+	4+	4+
3	Immucor 37832	B11193 R1R1 #5	+	+	0	0	+			0	+	**0**	+	0	+	0	+	0	+	0	+	0	+	+	0	+	0	+	0	+	+		4+	4+	**3+**
4	Immucor 37832	C5066 R2R2 #7	+	0	+	+	0			0	0	+	+	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	+	+		4+	4+	4+
5	Immucor 37832	C7328 R2R2 #9	+	0	+	+	0			0	0	+	0	+	0	+	+	+	+	0	+	0	+	+	+	0	+	0	+	0		4+	4+	4+
6	Immucor 37832	R2421 R1r #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	+	0	+	+	+	0	+	0	+	+		4+	4+	4+
7	Immucor 37832	E967 r'r #11	0	+	0	+	+			0	+	+	+	0	+	0	+	0	+	0	+	0	+	+	0	+	+	0	+	0		4+	4+	4+
8	Immucor 37832	F956 r''r #12	0	0	+	+	+			0	+	+	0	+	+	0	+	0	+	0	+	0	+	+	+	+	+	0	+	+		4+	4+	4+
9	Immucor 37832	F1079 r''r #15	0	0	+	+	+			0	+	+	0	+	+	0	0	0	+	0	+	0	+	+	+	+	+	0	+	+		4+	4+	4+
10	Immucor 37832	N5173 rr #16	0	0	0	+	+			0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	+	+	0	+	+		4+	4+	4+
11	Immucor 37832	D563 Ror #20	+	0	0	+	+			0	0	+	+	+	0	0	+	0	+	0	+	+	+	0	0	+	0	0	+	0	0	He+	4+	4+	4+
	Auto Control																															0	0	0/0

Papain treated adsorption cells, plasma adsorbed 1x @ 4°C
Untreated adsorption cells, plasma adsorbed 1x @ 4°C

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	Rh-Hr								MNS				Lewis		P	Kell				Duffy		Kidd		Luth		Additional antigens	Test Results		
	D	C	E	c	e	f	V	C^W	**M**	**N**	S	s	Le^a	Le^b	P1	K	k	Kp^a	Js^a	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		IS	LISS 37C	LISS AHG
R_1R_1 W18112413651500*	+	+	0	0	+				**0**	**+**	0	+	+	0	+	0				0	+	+	+						
R_2R_2 W18112414520200H	+	0	+	+	0				+	0	+	0	0	0	+	0				+	0	+	0						
rr W18112411972300*	0	0	0	+	+				+	0	+	+	0	+	+	0				0	+	0	+						
R_1R_1																													
B 9440011-00 304320 #2	+	0	+	+	0	0		0	+	**0**	+	0	+	0	s	0	+	0	0	+	0	+	0	+	+		0	0	**0/0**
W 37813 G478 #7	0	0	0	+	+			0	+	**0**	+	0	0	+	0	+	+	0	0	0	+	+	+	0	+		0	0	**0/0**
R_2R_2																													
W 37832 B3785 #1	+	+	0	0	+			0	0	+	0	+	0	+	+	+	+	0	0	0	+	+	+	0	+		0	0	0/m+
B 9440011-00 305405 #7	0	0	0	+	+	+		0	0	+	0	+	0	+	+	+	+	0	0	+	0	0	+	0	w				
rr																													
B 9440011-00 305606 #1	+	+	0	0	+	0		0	+	+	0	+	+	0	0	0	+	0	0	+	0	+	0	0	+		0	0	0/m+
W 37832 R2421 #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	0	+	+	+	0	0	+				

Double dose N+,'N'+ cell likely adsorbed antibody most efficiently

- Nonreactive against N-, 'N'+ cells
- N+, 'N'+ might have reacted

Papain treated adsorption cells, plasma adsorbed 1x @ 4°C
Untreated adsorption cells, plasma adsorbed 1x @ 4°C

American Red Cross
Missouri and Arkansas Region

	Rh-Hr								MNS				Lewis		P	Kell				Duffy		Kidd		Luth		Additional antigens	Test Results		
	D	C	E	c	e	f	V	C^W	M	**N**	S	s	Le^a	Le^b	P1	K	k	Kp^a	Js^a	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		IS	LISS 37C	LISS AHG
R_1R_1 W18112413651500*	+	+	0	0	+				0	+	0	+	+	0	+	0				0	+	+	+						
R_2R_2 W18112414520200H	+	0	+	+	0				+	**0**	+	0	0	0	+	0				+	0	+	0						
rr W18112411972300*	0	0	0	+	+				+	**0**	+	+	0	+	+	0				0	+	0	+						
R_1R_1																													
B 9440011-00 304320 #2	+	0	+	+	0	0		0	+	0	+	0	+	0	s	0	+	0	0	+	0	+	0	+	+		0	0	0/0√
W 37813 G478 #7	0	0	0	+	+			0	+	0	+	0	0	+	0	+	+	0	0	0	+	+	+	0	+		0	0	0/0√
R_2R_2																													
W 37832 B3785 #1	+	+	0	0	+			0	0	**+**	0	+	0	+	+	+	+	0	0	0	+	+	+	0	+		0	0	**0/m+**
B 9440011-00 305405 #7	0	0	0	+	+	+		0	0	+	0	+	0	+	+	+	+	0	0	+	0	0	+	0	w				
rr																													
B 9440011-00 305606 #1	+	+	0	0	+	0		0	+	**+**	0	+	+	0	0	0	+	0	0	+	0	+	0	0	+		0	0	**0/m+**
W 37832 R2421 #10	+	+	0	+	+			0	+	+	+	+	0	+	+	+	0	0	0	+	+	+	0	0	+				

Double dose N+,'N'+ cell likely adsorbed antibody most efficiently

- Nonreactive against N-, 'N'+ cells
- N+, 'N'+ might have reacted

N-, 'N'+ cells likely adsorbed antibody less efficiently

- Both cells tested happened to be N+, 'N'+
- N-, 'N'+ might not have reacted

Second adsorption removed reactivity- this is NOT anti-N plus anti-'N'

Are we done?

Make yet another list

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- Blood type: **A pos**
- DAT: **Negative**
- Common alloantibodies? **None**
- Anti-U? **No**
- Anti-N from N- S- s-? **Yes**

Discrepancy resolved with adsorbed plasma

Ruled out with adsorbed plasma

Ruled out with ficin treated reagent cells

Reacted with:
- One N+ 'N'- cell (neat)
- Two N- 'N'+ cells (neat)
- Three N- 'N'+ cells (trypsin treated)
- All N+ 'N'+ cells

Did not react with:
- One N- 'N'- cell (neat)
- Three N- 'N'- cells (ficin treated)

Are we done?

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Yes!

(mostly)

Transfusion recommendations

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- Crossmatch compatible red blood cell components negative for the N and ‘N’ antigens
  - No anti-’N’ antisera
  - Solution: U neg units

- Crossmatch compatible red blood cell components negative for the N and U antigens
  - N- U_{var} vs N- U-
    - Anti-U ruled out
    - Easier to find U_{var} units
    - Most U_{var} are ‘N’ neg
    - Consider prescreening U_{var} units with patient plasma before sending to hospital

Outcome

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- 2 units N- U- pRBC requested by hospital
- ARDP request placed, units located within 4-5 days
- Hospital decided to go ahead with surgery without units on hand- order canceled
- No further contact regarding patient- presumed uneventful surgery and recovery

Molecular results

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Blood Group	Antigen	Result	
Rh	c	+	
	C	0	
	e	+	
	E	0	
	V	+	
	VS	+	
Kell	K	0	
	k	+	
	Kp^a	0	
	Kp^b	+	
	Js^a	0	
	Js^b	+	
Duffy	Fy^a	0	
	Fy^b	+	
Kidd	Jk^a	+	
	Jk^b	+	
MNS	M	+	
	N	0	
	S	0	
	s	0	
	U	var	

COMMENTS: The sample is homozygous for the *GYPB* c.230C>T variant that is associated with exon 5 skipping.
Predicted phenotype: S-s-U^{var}

Presumed *GYP*He(NY) (GYPB*03N.02)*

No further testing requested

Takeaways

- Reactive AC at 4°C not always relevant
  - 4°C-only CAA quite common
- Get a good history
- Overwhelmed by options? Make lists
- Don’t forget to try enzymes and DTT treatment

- N and ‘N’ antigens- insignificant except when they aren't (very rare)
  - Most ‘common' situation is N- S- s- patient
  - Homozygous for some combo of *SHe* and/or *sHe* allele(s)

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Thank you

American Red Cross