Antibody Screening & Identification

Work Shop

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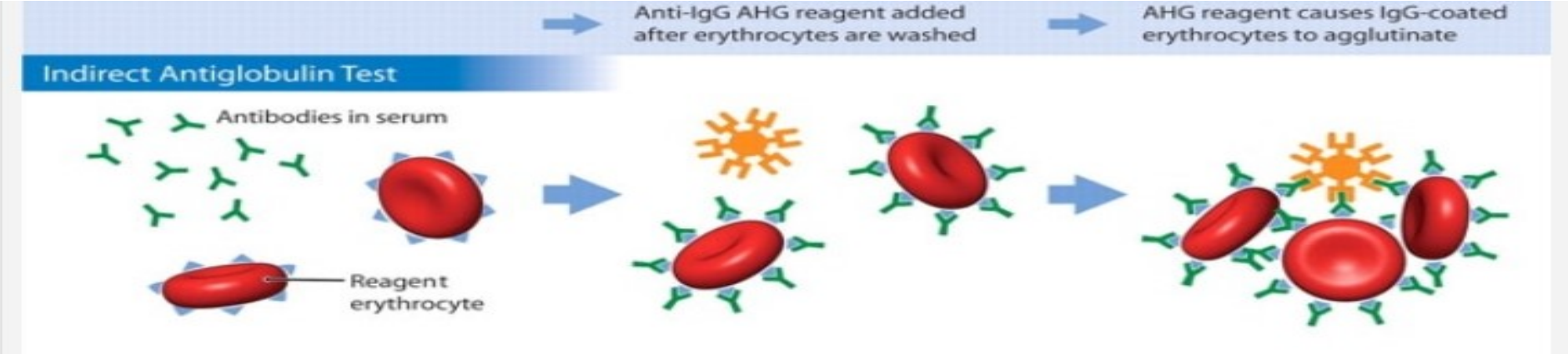
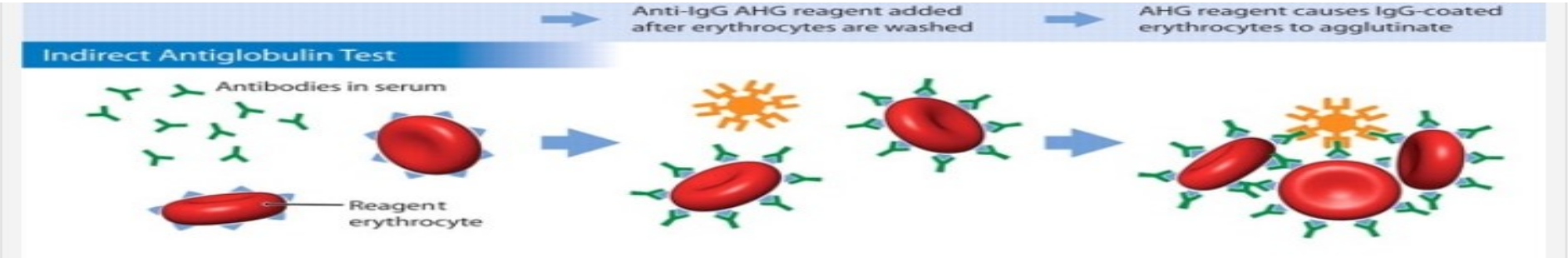


- *The most important tests in pretransfusion and prenatal testing is the antibody screen.*
- *The AABB's Standards for Blood Banks requires the use of the antibody screen test to detect clinically significant antibodies in both the blood donor and transfusion recipient.*
- *Only a small percentage of the healthy population (from 0.02% to 2%) has detectable red cell Abs In contrast, 14% to 50% of individuals in chronically transfused populations, may demonstrate red cell alloantibodies.*

Antibody Screening



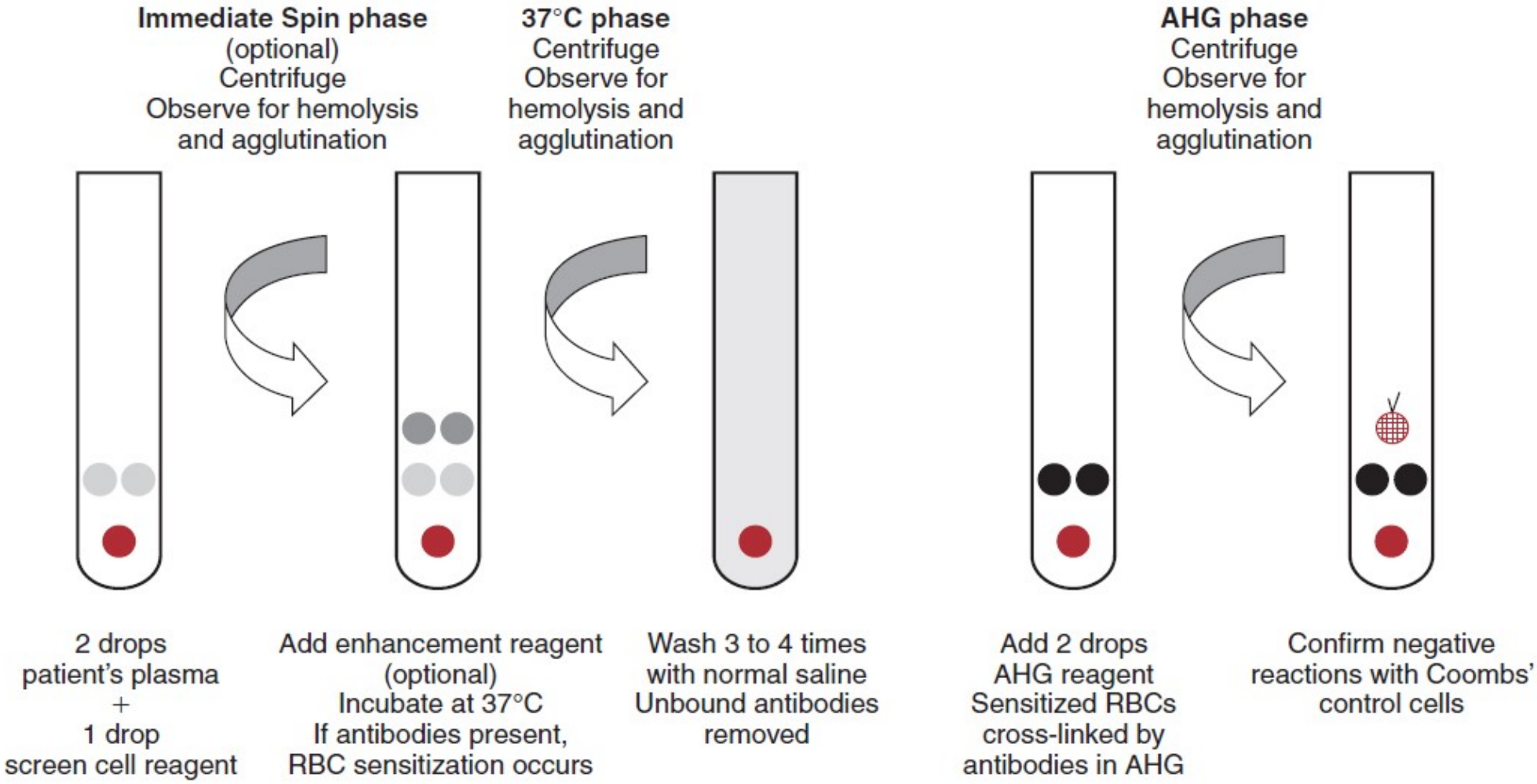
- *Patients requiring transfusion*
- *Patients with suspected transfusion reactions*
- *Women who are pregnant or after delivery*
- *Blood and plasma donors*
- *Patients with autoimmune hemolytic anemia*
- *Patients with graft versus host disease or other complications of organ or stem cell/marrow transplantation*

Specimen Requirements

- *Depending on the test methods used and the hematocrit of the patient, a 5-mL to 10-mL aliquot of whole blood usually contains enough serum or plasma for identifying simple antibody specificities; more whole blood (ie, 10 mL to 20 mL) may be required for complex studies.*

- *The use of samples anticoagulated with EDTA avoids problems associated with the in-vitro uptake of complement components by red cells, which may occur when clotted samples are used.*

- *Serum can be considered interchangeable with plasma*


Immediate Spin phase
(optional)
Centrifuge
Observe for hemolysis
and agglutination
37°C phase
Centrifuge
Observe for
hemolysis and
agglutination
AHG phase
Centrifuge
Observe for
hemolysis and
agglutination
2 drops
patient's plasma
+
1 drop
screen cell reagent
Add enhancement reagent
(optional)
Incubate at 37°C
If antibodies present,
RBC sensitization occurs
Wash 3 to 4 times
with normal saline
Unbound antibodies
removed
Add 2 drops
AHG reagent
Sensitized RBCs
cross-linked by
antibodies in AHG
Confirm negative
reactions with Coombs'
control cells

SCREENING CELLS

	Rh							MNSs				P1	Lewis		Lutheran		Kell		Duffy		Kidd					
Cell	**D**	**C**	**E**	**c**	**e**	**f**	**C^w**	**M**	**N**	**S**	**s**	**P1**	**Le^a**	**Le^b**	**Lu^a**	**Lu^b**	**K**	**k**	**Fy^a**	**Fy^b**	**Jk^a**	**Jk^b**				
I R1R1 (56)	+	+	0	0	+	0	0	+	+	0	+	0	+	0	0	+	+	+	+	0	+	+				
II R2R2 (89)	+	0	+	+	0	0	0	0	+	+	0	+	0	+	0	+	0	+	0	+	+	0				

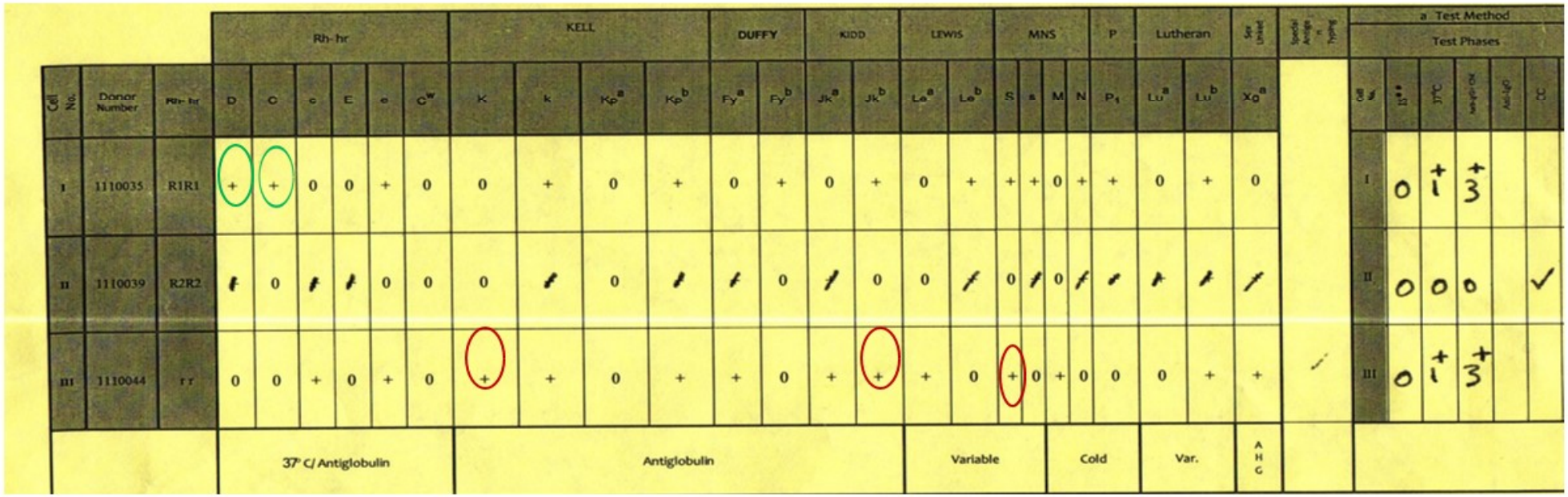
Cell	D	C	c	E	e	C^w	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P_1	M	N	S	s	Lu^a	Lu^b	Xg^a				
R1R1	+	+	0	0	+	0	0	+	0	+	0	+	+	+	+	0	+	0	+	+	+	+	+	0	+	+				
R2R2	+	0	+	+	0	0	+	+	0	+	0	+	+	0	0	+	0	+	0	0	0	+	0	0	+	+				
rr	0	0	+	0	+	0	0	+	0	+	0	+	0	+	+	+	0	+	+	+	+	0	+	0	+	+				

AHG Reagents

- *Monospecific reagents :detect either bound IgG or complement (C3)*
- *Poly-specific reagents that can simultaneously detect IgG and/or C3.*
- *Poly-specific AHG reagent is not often used when performing antibody detection testing since the presence of anticomplement in the AHG reagent may lead to the detection of clinically insignificant anti - bodies.*

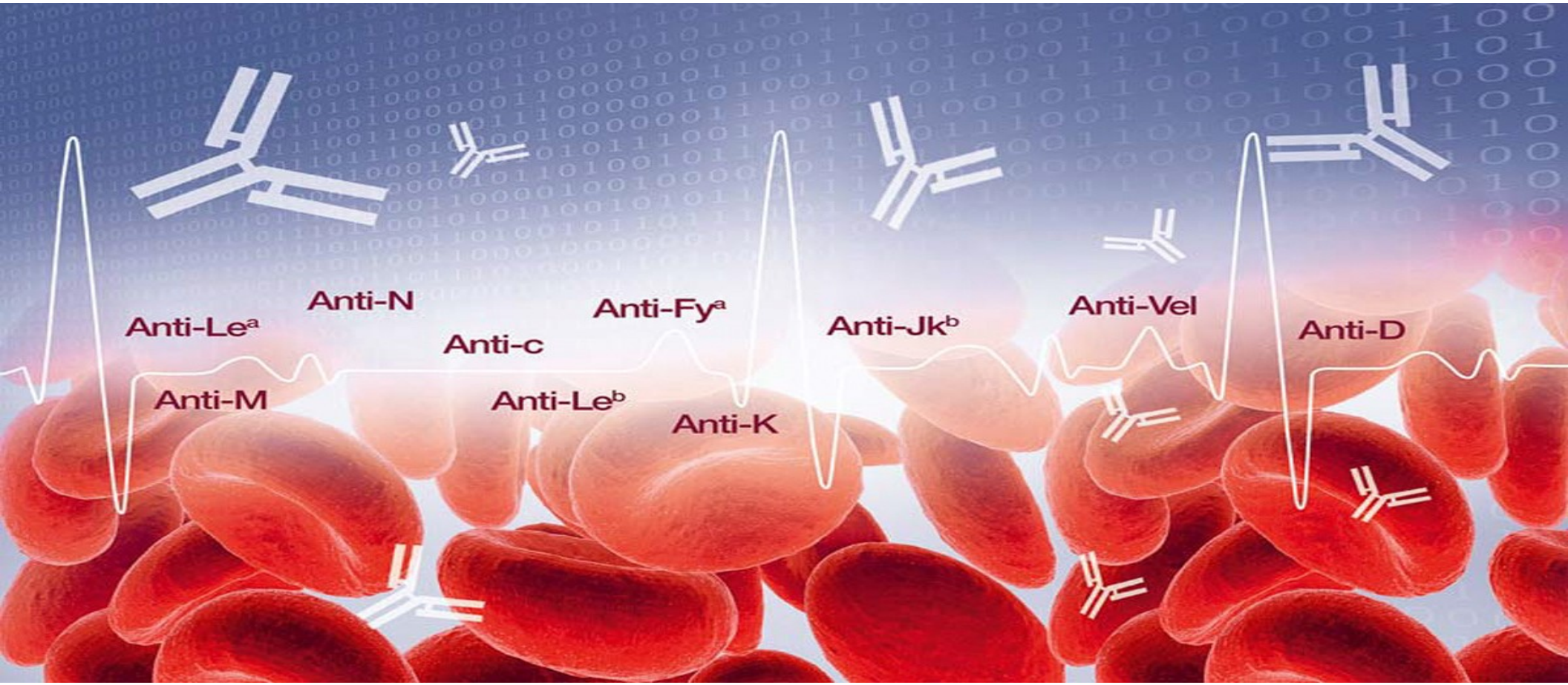
Interpretation of Results

Positive with the 2 cells I and III (3+) in IAT phase



Cell No.	Donor Number	Rh-hr	D	C	c	E	e	C^w	K	k	Kp^a	Kp^b	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	S	s	M	N	P_1	Lu^a	Lu^b	Xo^a	Special Antigen Typing	a Test Method / Test Phases: Cell No.	15	37°C	Anti-IgG	CC
I	1110035	R1R1	+	+	0	0	+	0	0	+	0	+	0	+	0	+	0	+	+	+	0	+	+	0	+	0		I	0	+1	+3		
II	1110039	R2R2	+	0	+	+	0	0	0	+	0	+	+	0	+	0	0	+	0	+	0	+	+	+	+	+		II	0	0	0		✓
III	1110044	r r	0	0	+	0	+	0	+	+	0	+	+	0	+	+	+	0	+	0	+	0	0	0	+	+	✓	III	0	+1	+3		
			37° C/ Antiglobulin						Antiglobulin								Variable		Cold			Var.		AHG							

Antibody Identification



Patient History

- *Transfusions and pregnancy within the last 3 months present the possibility of a mixed red cell population and recent antibody stimulation.*
- *Patient information, such as diagnosis, race, and age, offers additional clues to the nature of the antibody problem.*
- *Some diseases are associated with the development of certain antibodies. For example, a patient with systemic lupus erythematosus or carcinoma is frequently associated with a warm autoantibody, whereas pneumonia may result in a cold autoimmune process.*
- *In addition, knowing if a patient has received IVIG, RhIg, and antilymphocyte globulin is beneficial as these medications may passively transfer antibodies such as anti-A or anti-B, anti-D*
- *Immunotherapeutic agents may also interfere with serologic results. Anti-CD38 [daratumumab] and anti-CD47 treatment for multiple myeloma and other B-cell malignancies*

- *A panel, consists of group O reagent red cells with phenotypes for most common antigen specificities.*
- *Manufacturers prepare panels with various antigen configurations, which may include 10, 11, 15, 16,or 20 cells.*

Donor	Cell number	D	C	c	E	e	C^w	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P_1	M	N	S	s	Lu^a	Lu^b	Xg^a				
R1R1	1	+	+	0	0	+	0	0	+	0	+	0	+	+	+	+	0	0	+	+	+	+	+	+	0	+	+				
R1wR1	2	+	+	0	0	+	+	+	+	0	+	0	+	+	0	0	+	0	+	0	+	0	+	+	0	+	+				
RzR1	3	+	+	0	+	+	0	0	+	0	+	0	+	0	+	+	+	0	+	+	0	+	0	+	0	+	+				
R2R2	4	+	0	+	+	0	0	0	+	0	+	0	+	+	0	+	+	0	+	+	+	0	+	0	0	+	+				
r'r	5	0	+	+	0	+	0	0	+	0	+	0	+	0	0	0	+	+	0	+	+	0	+	+	0	+	0				
r"r	6	0	0	+	+	+	0	0	+	0	+	0	+	+	0	0	+	0	0	+	0	+	0	+	0	+	+				
rr	7	0	0	+	0	+	0	+	+	0	+	0	+	+	0	0	+	0	+	+	0	+	0	+	0	+	+				
Rr	8	0	0	+	0	+	0	0	+	0	+	0	+	0	+	+	0	0	+	0	+	0	+	0	0	+	+				
Rr	9	0	0	+	0	+	0	0	+	0	+	0	+	+	0	+	0	0	+	+	+	+	0	+	0	+	0				
R1r	10	+	+	+	0	+	0	0	+	0	+	0	+	+	0	+	+	0	+	+	+	0	0	+	0	+	+				
R0r	11	+	0	+	0	+	0	0	+	0	+	0	+	+	+	0	+	0	+	+	+	0	0	+	0	+	+				
	Patient Cells																														

Donor	Cell number	D	C	c	E	e	C^w	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P_1	M	N	S	s	Lu^a	Lu^b	Xg^a	IS	37	AHG	CC
R1R1	1	+	+	0	0	+	0	0	+	0	+	0	+	+	+	+	0	0	+	+	+	+	+	+	0	+	+	0	0	2+	
R1wR1	2	+	+	0	0	+	+	+	+	0	+	0	+	0	+	+	0	0	0	+	+	0	+	0	0	+	+	0	0	0	3+
RzR1	3	+	+	0	+	+	0	0	+	0	+	0	+	+	0	+	+	0	+	+	0	+	0	+	0	+	+	0	0	3+	
R2R2	4	+	0	+	+	0	0	0	+	0	+	0	+	+	0	+	+	+	0	+	+	0	+	0	0	+	+	0	0	3+	
r'r	5	0	+	+	0	+	0	0	+	0	+	0	+	0	0	0	+	0	+	+	+	0	+	+	0	+	0	0	0	0	3+
r"r	6	0	0	+	+	+	0	0	+	0	+	0	+	+	+	0	+	0	0	+	0	+	0	+	0	+	+	0	0	2+	
rr	7	0	0	+	0	+	0	+	+	0	+	0	+	+	+	0	+	0	+	+	0	+	0	+	0	+	+	0	0	2+	
rr	8	0	0	+	0	+	0	0	+	0	+	0	+	0	+	+	0	+	0	0	+	0	+	0	0	+	+	0	0	0	3+
r'r"	9	0	+	+	+	+	0	0	+	0	+	0	+	0	+	+	0	0	+	+	0	+	0	+	0	+	0	0	0	0	3+
rr	10	0	0	+	0	+	0	0	+	0	+	0	+	+	0	+	+	0	+	+	+	0	0	+	0	+	+	0	0	3+	
R0r	11	+	0	+	0	+	0	0	+	0	+	0	+	+	+	0	+	0	+	+	+	0	0	+	0	+	+	0	0	2+	
	Patient Cells																											0	0	0	3+

The pattern of antigen expression should be diverse so that it will be possible to distinguish one antibody from another and should include cells with homozygous expression of Rh, Duffy, Kidd, and MNSs antigens.

37/AHG:	Rh
AHG:	K, Fy, Jk, S/s
Variable:	Lu, Xg
IS (RT):	Le, MN, P

37 C and AHG	V	AHG Only	RT	Var.	RT	AHG	RT

	Rh-hr							Xg	Kell						Duffy		Kidd		P	Lutheran		MNSs				Lewis			Re
hr	D	C	E	c	e	f	C^w	Xg^a	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	P_1	Lu^a	Lu^b	M	N	S	s	Le^a	Le^b	Cell	
1	+	+	0	0	+	0	+	+	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	0	+	0	+	1	
1	+	+	0	0	+	0	0	+	0	+	0	+	0	+	+	0	+	+	0	0	+	0	+	0	+	0	+	2	
2	+	0	+	+	0	0	0	+	0	+	0	+	0	+	0	0	+	0	+	0	+	0	+	+	+	0	0	3	
	+	0	0	+	+	+	0	+	0	+	0	+	0	+	0	0	+	0	+	0	+	0	+	0	+	0	0	4	
	0	+	0	+	+	+	0	+	0	+	0	+	0	+	0	+	0	+	+	0	+	+	0	+	0	+	0	5	
	0	0	+	+	+	+	0	+	0	+	0	+	0	+	+	+	+	0	+	0	+	0	+	+	+	+	0	6	
	0	0	0	+	+	+	0	+	+	+	0	+	0	+	0	+	0	+	+	0	+	+	0	+	0	0	+	7	
	0	0	0	+	+	+	0	+	0	+	0	+	0	+	+	+	+	0	+	0	+	+	+	+	0	+	0	8	

Panel 8.3 Multiple Antibodies																									
	Rh							**MNSs**				**P1**	**Lewis**		**Lutheran**		**Kell**		**Duffy**		**Kidd**		***LISS***			
Cell	**D**	**C**	**E**	**c**	**e**	**f**	**C^w**	**M**	**N**	**S**	**s**	**P1**	**Le^a**	**Le^b**	**Lu^a**	**Lu^b**	**K**	**k**	**Fy^a**	**Fy^b**	**Jk^a**	**Jk^b**	***IS***	***37***	***AHG***	***CC***
1 R1R1 (51)	+	+	0	0	+	0	0	+	+	+	0	+	+	0	0	+	+	+	0	+	+	0	*0*	*0*	*0*	✓
2 R1R1 (32)	+	+	0	0	+	0	+	+	0	0	+	+	0	+	0	+	0	+	0	+	+	+	*0*	*0*	*0*	✓
3 R2R2 (64)	+	0	+	+	0	0	0	0	+	0	+	+	+	0	0	+	+	+	0	0	+	+	*0*	*0*	*3+*	
4 r'r (75)	0	+	0	+	+	+	0	+	0	+	+	+	0	+	0	+	0	+	+	0	+	+	*0*	*0*	*2+*	
5 r''r (87)	0	0	+	+	+	+	0	+	+	+	+	+	0	+	0	+	0	+	+	0	+	0	*0*	*0*	*3+*	
6 rr (98)	0	0	0	+	+	+	0	+	+	+	+	+	+	0	0	+	0	+	+	0	+	+	*0*	*0*	*2+*	
7 rr (76)	0	0	0	+	+	+	0	+	0	+	0	0	0	+	0	+	+	0	+	+	0	+	*0*	*0*	*1+*	
8 rr (53)	0	0	0	+	+	+	0	+	0	+	+	+	0	0	0	+	0	+	0	+	0	+	*0*	*0*	*0*	✓
9 rr (23)	0	0	0	+	+	+	0	+	+	+	+	0	+	0	0	+	0	+	+	+	+	0	*0*	*0*	*1+*	
10 R1R1 (34)	+	+	0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	0	+	+	0	*0*	*0*	*0*	✓
Patient cells			0																0				*0*	*0*	*0*	✓

The reaction strengths are variable (1+ to 3+), which suggests:

More than one antibody and/or an antibody that exhibits dosage.

DAT & Auto control

- If the auto control is positive in the antiglobulin phase, a DAT should be performed.
- If the DAT result is negative, antibodies to an additive solution constituent or autoantibodies that are reactive only in the presence of the additive solution should be considered.

- Warm autoantibodies and cold auto“ antibodies, such as anti-I, "IH, or -P, may be reactive in an IAT when certain enhancement techniques are used; therefore, testing should be repeated in another medium.

- If the DAT result is positive, it must be interpreted with careful attention to the transfusion history. Autoantibodies or drugs could explain a positive DAT result.

- If the patient has an alloantibody and recently received blood that expressed the corresponding antigen, the positive DAT result may be caused by coating of the donor red cells with alloantibody.

"Cross-out," "rule-out," or "exclusion" method

Cell	Phenotype	Rhesus					Kell		Duffy		Kidd		Lewis		MNS				P	Test Result		
		D	C	E	c	e	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	M	N	S	s	P_1	IS	37°C	IAT
1	$R_1^wR_1$	+	+	0	0	+	+	+	+	+	+	+	0	+	+	+	0	+	0	0	0	0
2	R_1R_1	+	+	0	0	+	0	+	0	+	+	0	+	0	0	+	0	+	+	0	0	0
3	R_2R_2	+	0	+	+	0	0	+	0	+	+	+	0	+	+	+	+	+	0	2+	3+	4+
4	R_0r	+	0	0	+	+	0	+	+	0	+	0	0	0	+	+	0	+	+	0	0	0
5	r'r	0	+	0	+	+	0	+	+	+	+	+	0	+	+	0	+	0	+	0	0	0
6	r''r	0	0	+	+	+	0	+	+	+	+	+	0	+	+	+	+	+	+	1+	2+	2+
7	rr	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
9	rr	0	0	0	+	+	0	+	+	+	+	0	+	0	+	0	+	0	+	0	0	0
10	rr	0	0	0	+	+	0	+	+	0	+	0	0	+	+	0	0	+	+	0	0	0
11	R_1R_1	+	+	0	0	+	0	+	0	+	0	+	0	0	+	0	+	0	0	0	0	0
Patient Cells																				0	0	0

- *Panel cells that are heterozygous, particularly in the Duffy, Kidd, Rh, and MNS systems, should not be crossed out because the antibody might be too weak to react or is exhibiting dosage.*
- *It is preferable to rule out using homozygous cells whenever possible.*

Cell	Phenotype	Rhesus					Kell		Duffy		Kidd		Lewis		MNS				P	Test Result		
		D	C	E	c	e	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	M	N	S	s	P_1	IS	37°C	IAT
1	$R_1{}^wR_1$	+	+	0	0	+	+	+	+	+	+	+	0	+	+	+	0	+	0	0	0	0
2	R_1R_1	+	+	0	0	+	0	+	0	+	+	0	+	0	0	+	0	+	+	0	0	0
3	R_2R_2	+	0	+	+	0	0	+	0	+	+	+	0	+	+	+	+	+	0	2+	3+	4+
4	R_0r	+	0	0	+	+	0	+	+	0	+	0	0	0	+	+	0	+	+	0	0	0
5	r′r	0	+	0	+	+	0	+	+	+	+	+	0	+	+	0	+	0	+	0	0	0
6	r″r	0	0	+	+	+	0	+	+	+	+	+	0	+	+	+	+	+	+	1+	2+	2+
7	rr	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
9	rr	0	0	0	+	+	0	+	+	+	+	0	+	0	+	0	+	0	+	0	0	0
10	rr	0	0	0	+	+	0	+	+	0	+	0	0	+	+	0	0	+	+	0	0	0
11	R_1R_1	+	+	0	0	+	0	+	0	+	0	+	0	0	+	0	+	0	0	0	0	0
Patient Cells																				0	0	0

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Haematology made simple

- *The next step in panel interpretation is to look at the positive reactions and match the pattern.*
- *When a single antibody is present, the pattern of reactions observed matches one of the antigen columns.*

Confirming Antibody Identification

Cell	Phenotype	Rhesus					Kell		Duffy		Kidd		Lewis		MNS				P	Test Result		
		D	C	E	c	e	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	M	N	S	s	P_1	IS	37°C	IAT
1	$R_1^wR_1$	+	+	0	0	+	+	+	+	+	+	+	0	+	+	+	0	+	0	0	0	0
2	R_1R_1	+	+	0	0	+	0	+	0	+	+	0	+	0	0	+	0	+	+	0	0	0
3	R_2R_2	+	0	+	+	0	0	+	0	+	+	+	0	+	+	+	+	+	0	2+	3+	4+
4	R_or	+	0	0	+	+	0	+	+	0	+	0	0	0	+	+	0	+	+	0	0	0
5	r'r	0	+	0	+	+	0	+	+	+	+	+	0	+	+	0	+	0	+	0	0	0
6	r''r	0	0	+	+	+	0	+	+	+	+	+	0	+	+	+	+	+	+	1+	2+	2+
7	rr	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
9	rr	0	0	0	+	+	0	+	+	+	+	0	+	0	+	0	+	0	+	0	0	0
10	rr	0	0	0	+	+	0	+	+	0	+	0	0	+	+	0	0	+	+	0	0	0
11	R_1R_1	+	+	0	0	+	0	+	0	+	0	+	0	0	+	0	+	0	0	0	0	0
Patient Cells																				0	0	0

Cell	Phenotype	Rhesus					Kell		Duffy		Kidd		Lewis		MNS				P	Test Result		
		D	C	E	c	e	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	M	N	S	s	P_1	IS	37°C	IAT
1	R_1R_1	+	+	0	0	+	0	+	+	0	0	+	0	+	+	+	0	+	0	0	0	0
2	R_2R_2	+	0	+	+	0	0	+	0	+	+	0	+	0	+	0	+	0	0	0	2+	4+
3	rr	0	0	0	+	+	+	+	+	+	0	+	0	+	+	+	+	+	+	0	0	0

- *The standard approach requires two antigen-positive panel cells that agglutinate and two antigen-negative panel cells that do not agglutinate.*
- *AABB supports this current standard practice known as the rule of two.*

Patient's Phenotype

- *Individuals cannot make alloantibodies to antigens they possess on their own RBCs*
- *Phenotyping may be complicated when a patient has a positive DAT*
- *When the IgG DAT is positive, antibody bound to the RBCs prevents the typing serum from reacting with the corresponding antigen. When AHG reagent is added to the test system, it will react with the bound antibody detected in the DAT, yielding false-positive results.*
- *Two reagents useful in stripping antibody from the RBC surface, while leaving the membrane intact to allow phenotyping, are acid glycine/EDTA (EGA) and chloroquine diphosphate.*
- *Recently transfused patients present a different challenge. When phenotyping a recently transfused patient, mixed-field reactions may be observed since two RBC populations are present (i.e., autologous patient RBCs and transfused donor RBCs).*
- *To get an accurate phenotype of autologous patient RBCs, reticulocyte typing can be performed. Usage of molecular technologies, which determine the RBC genotype is more useful.*

Multiple Antibody Resolution: Selected Cell Panel

Panel 8.3 Multiple Antibodies

	Rh							MNSs				P1	Lewis		Lutheran		Kell		Duffy		Kidd		*LISS*			
Cell	**D**	**C**	**E**	**c**	**e**	**f**	**C^w**	**M**	**N**	**S**	**s**	**P1**	**Le^a**	**Le^b**	**Lu^a**	**Lu^b**	**K**	**k**	**Fy^a**	**Fy^b**	**Jk^a**	**Jk^b**	*IS*	*37*	*AHG*	*CC*
1 R1R1 (51)	+	+	0	0	+	0	0	+	+	+	0	+	+	0	0	+	+	+	0	+	+	0	0	0	0	✓
2 R1R1 (32)	+	+	0	0	+	0	+	+	0	0	+	+	0	+	0	+	0	+	0	+	+	+	0	0	0	✓
3 R2R2 (64)	+	0	+	+	0	0	0	0	+	0	+	+	+	0	0	+	+	+	0	0	+	+	0	0	3+	
4 r'r (75)	0	+	0	+	+	+	0	+	0	+	+	+	0	+	0	+	0	+	+	0	+	+	0	0	2+	
5 r''r (87)	0	0	+	+	+	+	0	+	+	+	+	+	0	+	0	+	0	+	+	0	+	0	0	0	3+	
6 rr (98)	0	0	0	+	+	+	0	+	+	+	+	+	+	0	0	+	0	+	+	0	+	+	0	0	2+	
7 rr (76)	0	0	0	+	+	+	0	+	0	+	0	0	0	+	0	+	+	0	+	+	0	+	0	0	1+	
8 rr (53)	0	0	0	+	+	+	0	+	0	+	+	+	0	0	0	+	0	+	0	+	0	+	0	0	0	✓
9 rr (23)	0	0	0	+	+	+	0	+	+	+	+	0	+	0	0	+	0	+	+	+	+	0	0	0	1+	
10 R1R1 (34)	+	+	0	0	+	0	+	0	+	+	0	+	0	+	0	+	0	+	0	+	+	0	0	0	0	✓
Patient cells			0																0				0	0	0	✓

- *The rule of two was not met for confirmation of anti-E. One more E+ Fy(a−) panel cell requires testing.*
- *The patient's red cell phenotype is E-negative and Fy(a−), providing additional support for the presence of anti-E and anti-Fya.*

Panel 8.4 Selected Cell Panel

	Rh							MNSs				P1	Lewis		Lutheran		Kell		Duffy		Kidd		*LISS*		
Cell	**D**	**C**	**E**	**c**	**e**	**f**	**C^w**	**M**	**N**	**S**	**s**	**P1**	**Le^a**	**Le^b**	**Lu^a**	**Lu^b**	**K**	**k**	**Fy^a**	**Fy^b**	**Jk^a**	**Jk^b**	*IS*	*37*	*AHG*
1 r''r (22)	0	+	0	+	+	0	0	+	+	+	0	+	0	+	0	+	+	+	0	+	+	0			
2 R1R1 (72)	+	+	0	0	+	0	+	+	0	0	+	+	0	+	0	+	0	+	0	+	+	+			
3 R2R2 (45)	+	0	(+)	+	0	0	0	0	+	0	+	+	+	0	0	+	+	+	(0)	0	+	+	*0*	*0*	*3+*
4 r''r (28)	0	0	+	+	+	+	0	+	0	+	+	+	0	+	0	+	0	+	+	0	+	+			
5 rr (88)	0	0	0	+	+	+	0	+	+	+	+	+	0	+	0	+	0	+	0	0	+	0			
6 rr (38)	0	0	0	+	+	+	0	+	+	+	+	+	+	0	0	+	0	+	+	0	+	+			
7 rr (74)	0	0	0	+	+	+	0	+	0	+	0	0	0	+	0	+	+	0	+	+	0	+			
8 rr (21)	0	0	0	+	+	+	0	+	0	+	+	+	0	0	0	+	0	+	0	+	0	+			
9 rr (67)	0	0	0	+	+	+	0	+	+	+	+	0	+	0	0	+	0	+	+	+	+	0			
10 R2R2 (92)	+	0	(+)	+	0	0	0	0	+	+	0	+	0	+	0	+	0	+	(0)	+	+	0	*0*	*0*	*3+*

Case study 1

Initial Data:

- A 74-year-old female of European ancestry with myelodysplastic syndrome.
- Her hemoglobin level is 7.5 g/dL.
- Two units of Red Blood Cells Leukocytes Reduced have been ordered
- She last received a transfusion at the facility 2 months ago.
- At that time, an anti-Fya was identified in her plasma.

ABO and RH Typing:

Forward (Cell) Typing			Reverse (Serum) Typing	
Anti-A	Anti-B	Anti-D	A_1 Cells	B Cells
4+	0	4+	0	4+

ABO type: Group A+

The results of forward and reverse typing are consistent.

Antibody Detection Test (Screen):

	RH						MNS				LU		P	Lewis		Kell		Duffy		Kidd		Gel Test
	D	C	E	c	e	f	M	N	S	s	Lu^a	Lu^b	P_1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	IAT
1	+	+	0	0	+	0	+	+	+	+	0	+	+	0	+	0	+	+	0	0	+	**3+**
2	+	0	+	+	0	0	+	0	+	0	0	+	+	+	0	+	+	+	+	+	0	**2+**
3	0	0	0	+	+	+	0	+	0	+	0	+	0	0	+	0	+	0	+	+	+	**0**

- Because both cells that demonstrated reactivity were Fy(a+), the initial antibody detection results suggest that the anti-Fya is still reactive in the patient serum.
- Anti-Fya would be reactive with these cells potentially masking other antibodies present in the serum.

Antibody Identification Panel 1:

	RH							MNS				LU		P	Lewis		Kell		Duffy		Kidd		Gel Test
	D	C	C^w	E	c	e	f	M	N	S	s	Lu^a	Lu^b	P_1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	IAT
1	+	+	0	0	0	+	0	+	+	+	+	0	+	+	0	+	0	+	0	+	+	+	**0**
2	+	+	+	0	0	+	0	+	+	+	0	0	+	0	0	0	0	+	0	+	+	+	**3+**
3	0	0	0	+	+	+	0	0	+	+	+	0	+	0	0	0	0	+	0	+	0	+	**0**
4	0	0	0	0	+	+	+	+	+	0	+	0	+	+	+	0	0	+	0	+	0	+	**0**
5	+	0	0	+	+	0	0	+	+	0	+	0	+	+	+	0	0	+	0	+	+	0	**0**
6	0	0	0	+	+	+	+	+	+	+	+	0	+	+	0	+	+	0	0	+	+	+	**0**
7	0	+	0	0	+	+	+	+	+	0	+	0	+	+	0	+	+	+	0	+	0	+	**0**
AC																							**0**

- The most efficient choice would be assemble a panel of Fy(a-) cells of varying phenotype.
- If Fy(a+) cells were tested, the reaction of anti-Fya would mask the reaction of other antibodies

Interpretation of panel ID

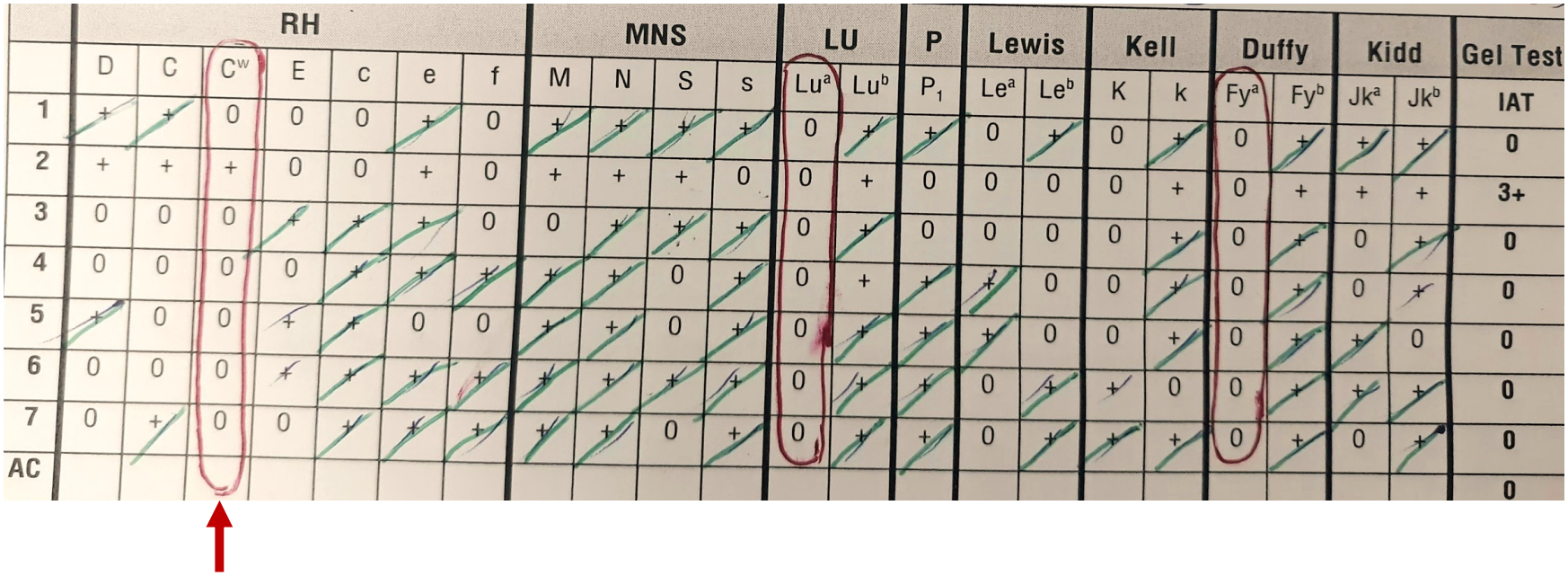
	RH							MNS				LU		P	Lewis		Kell		Duffy		Kidd		Gel Test
	D	C	C^w	E	c	e	f	M	N	S	s	Lu^a	Lu^b	P_1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	IAT
1	+	+	0	0	0	+	0	+	+	+	+	0	+	+	0	+	0	+	0	+	+	+	0
2	+	+	+	0	0	+	0	+	+	+	0	0	+	0	0	0	0	+	0	+	+	+	3+
3	0	0	0	+	+	+	0	0	+	+	+	0	+	0	0	0	0	+	0	+	0	+	0
4	0	0	0	0	+	+	+	+	+	0	+	0	+	+	+	0	0	+	0	+	0	+	0
5	+	0	0	+	+	0	0	+	+	0	+	0	+	+	+	0	0	+	0	+	+	0	0
6	0	0	0	+	+	+	+	+	+	+	+	0	+	+	0	+	+	0	0	+	+	+	0
7	0	+	0	0	+	+	+	+	+	0	+	0	+	+	0	+	+	+	0	+	0	+	0
AC																							0

Anti-C(cell 1) ,E (cell 5) can be ruled out.

Anti-Lea ,Kell, jka can be ruled out.

Anti-S has not be ruled out. Because the only double dose cell is cell 2

Interpretation of panel ID



	RH							MNS				LU		P	Lewis		Kell		Duffy		Kidd		Gel Test
	D	C	C^w	E	c	e	f	M	N	S	s	Lu^a	Lu^b	P_1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	IAT
1	+	+	0	0	0	+	0	+	+	+	+	0	+	+	0	+	0	+	0	+	+	+	0
2	+	+	+	0	0	+	0	+	+	+	0	0	+	0	0	0	0	+	0	+	+	+	3+
3	0	0	0	+	+	+	0	0	+	+	+	0	+	0	0	0	0	+	0	+	0	+	0
4	0	0	0	0	+	+	+	+	+	0	+	0	+	+	+	0	0	+	0	+	0	+	0
5	+	0	0	+	+	0	0	+	+	0	+	0	+	+	+	0	0	+	0	+	+	0	0
6	0	0	0	+	+	+	+	+	+	+	+	0	+	+	0	+	+	0	0	+	+	+	0
7	0	+	0	0	+	+	+	+	+	0	+	0	+	+	0	+	+	+	0	+	0	+	0
AC																							0

Anti-Cw is most consistent with the result of panel, because the only reactive cell was cell2, which is Cw+

Antibody Identification Panel 2:

	RH							MNS				LU		P	Lewis		Kell		Duffy		Kidd		Gel Test
	D	C	C^w	E	c	e	f	M	N	S	s	Lu^a	Lu^b	P_1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	IAT
1	+	+	+	0	0	+	0	+	+	+	+	0	+	+	0	+	0	+	0	+	+	+	**3+**
2	+	+	+	0	0	+	0	+	+	+	0	0	+	0	0	0	0	+	0	+	+	+	**3+**

- One or two additional Cw+ cells that are Fy(a-) should be tested and found reactive.
- Confirmatory test : phenotyping for Cw antigen
- Cw is an antigen in the RH system, a mutation on RHCe protein; therefore, most Cw+ individual are C+

Provide suitable blood for the patient?

Anti-Cw and Anti-Fya are clinically significant antibodies that may cause HTR & HDFN

Selection of Fy(a−), Cw− donor units.

A crossmatch including both 37 C and IAT phases of testing.

No commercial anti-Cw is available in your laboratory to type donor units:

- Type the compatible units with anti-Fya and anti-C.
- Cw is a low prevalence antigen present only in 2% of the population.
- Cw- donor units could be obtained by also selecting C negative units.

References:

1. Technical manual AABB, 20th Edition, 2020

2. Modern Blood Banking & Transfusion Practices, 7th Edition, 2019

3. Basic & Applied Concepts of Blood Banking and Transfusion Practices, 5th Edition, 2021

4. Antibody Identification: Art or Science? A Case Study Approach, Editors Janis R. Hamilton, MS, MT(ASCP)SBB Susan, 2013 by AABB

Thank you for your attention

Initial Data:

JP is a 58-year-old male with a history of cardiovascular disease for the past 10 years. He is scheduled for his second cardiac bypass surgery. For his first surgery, 7 years ago, he received 3 units of Red Blood Cells (RBCs) postoperatively. The transfusions were unremarkable.

An order for type and crossmatch of 4 RBC units has been received in preparation for this surgery.

ABO and RH Typing:

Forward (Cell) Typing			Reverse (Serum) Typing	
Anti-A	**Anti-B**	**Anti-D**	**A_1 Cells**	**B Cells**
0	4+	4+	4+	0

Antibody Detection Test (Screen):

	RH						MNS				LU		P	Lewis		Kell		Duffy		Kidd		Gel Test
	D	C	E	c	e	f	M	N	S	s	Lu^a	Lu^b	P_1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	IAT
1	+	+	0	0	+	0	+	+	+	+	0	+	+	+	0	+	+	+	0	0	+	**1+**
2	+	0	+	+	0	0	+	0	0	+	0	+	+	0	+	0	+	+	+	+	0	**0**

Antibody Identification Panel:

	RH						MNS				LU		P	Lewis		Kell		Duffy		Kidd		Gel Test
	D	C	E	c	e	f	M	N	S	s	Lu^a	Lu^b	P_1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	IAT
1	+	+	0	0	+	0	+	+	+	+	0	+	+	0	+	0	+	0	+	+	+	**0**
2	+	+	0	0	+	0	+	+	+	+	0	+	0	0	0	0	+	+	+	+	+	**0**
3	+	0	+	+	0	0	0	+	+	+	0	+	0	0	0	0	+	+	+	0	+	**0**
4	+	0	0	+	+	+	+	+	0	+	0	+	+	+	0	0	+	0	0	0	+	**0**
5	0	+	0	+	+	+	+	+	0	+	0	+	+	+	0	0	+	+	0	+	0	**0**
6	0	0	+	+	+	+	+	+	+	+	0	+	+	0	+	0	+	0	+	+	+	**0**
7	0	0	0	+	+	+	+	+	+	0	0	+	+	+	0	+	+	0	+	0	+	**1+**
8	0	0	0	+	+	+	+	+	+	+	0	+	0	0	+	0	+	+	0	+	0	**0**
9	0	0	0	+	+	+	+	0	+	+	0	+	+	+	0	0	+	0	+	+	+	**0**
10	0	0	0	+	+	+	0	+	0	+	0	+	+	0	+	0	+	+	0	0	+	**0**
11	+	+	0	0	+	0	+	+	+	+	0	+	0	+	0	+	+	0	+	+	+	**1+**
AC																						**0**