Usefulness of the effect of enzymes and DTT on antigens in antibody identification[1,2]

The following table shows *general* patterns of reactions; for more detail, see individual antigen sheets. It is important to control for "anti-enzyme" reactivity, i.e., by testing autologous RBCs treated in parallel or by testing an eluate. The patterns given are a useful guide in antibody identification, but remember that not all antibodies read the FactsBook!

Ficin/ Papain	Trypsin	α-Chymo-trypsin	200 mM DTT/AET	Possible specificity
Negative	Negative	Negative	Positive	Bp^a; Ch/Rg; XG
Negative	Negative	Negative	Negative	IN; JMH
Negative	Negative	Positive	Positive	M, N, En^aTS; Ge2, Ge4
Negative	Positive	Negative	Positive	'N'; Fy^a, Fy^b
Variable	Positive	Negative	Positive	S, s
Variable	Positive	Negative	Weak or negative	YT
Negative	Positive	Positive	Positive	En^aFS
Positive	Negative	Negative	Weak or negative	LU, MER2
Positive – Papain Weak or negative – Ficin	Negative	Negative	Negative	KN
Positive	Negative	Weak	Negative	DO
Positive	Positive	Negative	Weak	CROM
Positive	Positive	Negative	Positive	Some DI (3^{rd} loop)
Positive	Positive	Positive/weak	Negative	LW
Positive	Positive/weak	Positive/weak	Positive	SC
Positive	Positive^	Positive^	Negative	KEL^ (except KALT, which is trypsin sensitive)
Positive	Positive	Positive	Positive	ABO; En^aFR, U; P1PK; RH; LE; Fy3; JK; most DI; CO; H; Ge3; OK; I/i; P, FORS; JR; LAN, Cs^a; ER; LKE, PX2; Vel,† ABTI; At^a; Emm; AnWj; Sd^a; PEL; MAM
Positive	Positive	Positive	Enhanced	Kx

^Kell blood group system antigens are sensitive to treatment with a mixture of trypsin and α-chymotrypsin.
†DTT may be variable.

Effect of acid on antigen expression

EDTA/glycine/acid-treated RBCs do not express antigens in the KEL blood group system, the ER collection or Bg antigens, and antigens of the JK blood group system may be weakened.

Effect of chloroquine diphosphate on antigen expression

A modified technique of treating RBCs with chloroquine for 30 mins at 37°C weakens Mt^a, Lu^b, Fy^b, Yt^a, Bg^a, and antigens of the RH, DO, KN, and JMH blood group systems.

Substrate specificity of selected enzymes for peptide and CHO bonds

Classification	Enzyme (source)	Substrate specificity (in order of preference)
Thiol endoprotease has an essential cysteine in the active site and may require a sulfhydryl compound to activate it	Bromelin (Pineapple) Ficin (Fig tree latex) Papain (Papaya)	Hydrolyzes C-terminal peptide bond of Lys, Ala, Tyr, Gly Hydrolyzes C-terminal peptide bond of Lys, Ala, Tyr, Gly, Asp, Leu, Val Hydrolyzes C-terminal peptide bond of Arg, Lys, and bond next but one to Phe
Metallo endoprotease requires a specific metal ion in the active site	Pronase (*Streptomyces griseus*)	Hydrolyzes C-terminal peptide bond of any hydrophobic amino acid
Serine endoprotease requires serine and histidine residues at the enzyme site for enzymatic activity	α-chymotrypsin (Bovine pancreas) Proteinase K (*Tritirachium album*) Trypsin (Bovine or Porcine pancreas) V8 protease (*Staphylococcus aureus* strain V8)	Hydrolyzes C-terminal peptide bond of Phe, Trp, Tyr, Leu Hydrolyzes C-terminal peptide bond of aromatic or hydrophobic amino acids Hydrolyzes C-terminal peptide bond of Arg, Lys Hydrolyzes C-terminal peptide bond of Glu, Asp

(*Continued*)

(Continued)

Classification	Enzyme (source)	Substrate specificity (in order of preference)
Carboxyl endoprotease has an essential COOH in the active site	Pepsin (Porcine stomach mucosa)	Hydrolyzes C-terminal peptide bond of Phe, Leu, Trp, Tyr, Asp, Glu
Exoglycosidase	Sialidase/Neuraminidase (*Vibrio cholerae*) α-Galactosidase (GH^27/36 from e.g., coffee bean) A-zyme (GH^109, bacterial α3-*N*-acetylgalactosaminidase) B-zyme (GH^110, bacterial α3-galactosidase)	Hydrolyzes glycosidic bond between terminal NeuAc in any linkage to any sugar Hydrolyzes glycosidic bond of α-linked terminal Gal in B, P^k, and P1 antigens Hydrolyzes glycosidic bond of α3-linked terminal GalNAc in A antigen, leaving H antigen Hydrolyzes glycosidic bond of α3-linked terminal Gal in B antigen, leaving H antigen
Endoglycosidase	Endo F (*Flavobacterium meningosepticum*)	Hydrolyzes the glycosidic bond between the two core GlcNAc residues in biantennary N-glycans, and leaves one GlcNAc attached to the Asn residue of the protein

Endo = Internal substrate bonds. Exo = Terminal substrate bonds.
Note: Bacterial deacetylases may modify the side-chains of sugars, e.g., the acquired B phenomenon results from deacetylation of *N*-acetylgalactosamine to galactosamine. Organisms such as *E. coli*, *Clostridium tertium*, and *Proteus mirabilis* have been implicated in this phenomenon.
^GH = glycoside hydrolase family, see www.cazy.org.

Cord RBCs are

Negative for	Le^a, Le^b (sometimes); Ch, Rg; AnWj; Sd^a
Weak for	A, B; H; I; P1; Lu^a, Lu^b; Yt^a; JMH; sometimes Xg^a; Vel; Bg; KN; and DO antigens; Fy3 as detected by anti-Fy3 made by Blacks
Strong for	LW system antigens; i antigen

Antigens with variable expression on different RBCs in the same sample and on RBCs from different donors (presumed to be due to different antigen copy number)

Carbohydrate antigens	A, B; FORS1; H; I; Le^a, Le^b; P1, P^k; P; Sd^a
Protein antigens	Lu^a, Lu^b; Xg^a; KN; MER2; JMH; Jr^a; Vel; Lan; AnWj; Ch/Rg
Plasma adsorbed antigens	Le^a, Le^b; Ch, Rg

Mixed Field Agglutination may be observed in:

Transfused patients
Maternal-fetal hemorrhage or fetal-maternal hemorrhage
Stem cell transplant recipients
Chimera (genetic)
Genetic variants of antigen, e.g., A_3, A_{finn}, A_{mos}, B_{mos}
Chromosomal abnormalities resulting in two populations of RBCs, e.g., ABO and RH in leukemia
Low density of antigen sites, e.g., Xg^a, Sd^a, Lu^a
Polyagglutination, e.g., Tn
Modification by bacterial enzymes, e.g., deacetylation in acquired B
X-inactivation, Kx in female carriers.

Blood group antigens absent (altered) on selected RBC phenotypes

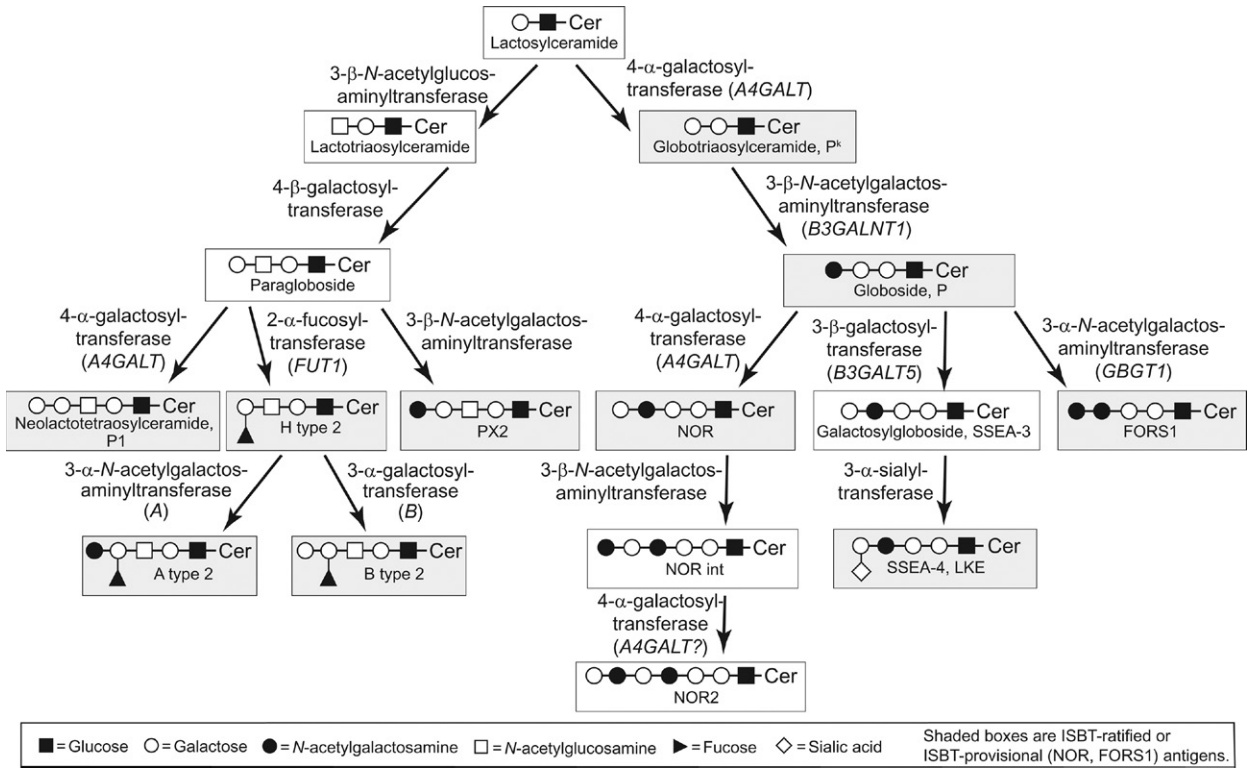
Phenotype	Absent or altered, usually reduced (in parentheses) antigens
O_h (Bombay)	ABO and H systems; rarely LE
A_h/B_h (para-Bombay)	(Very weak A and/or B antigens)
En(a–)Fin	M, N, GPA-associated; Wr^a, Wr^b
U–	S, s, U, He, and GPB-associated
M^kM^k	MNS; Wr^a, Wr^b (some antigens, not in the MNS system, may appear to be enhanced due to reduced sialic acid)
p [previously Tj(a–)]	P; P1, P^k, NOR; LKE (PX2 elevated)

(*Continued*)

(Continued)

Phenotype	Absent or altered, usually reduced (in parentheses) antigens
P_1^k	P; LKE, PX2
P_2^k	P; P1; LKE, PX2
Rh_{null}	RH; LW; RHAG; Fy5 (S, s, U may be weak)
Rh_{mod}	(Weak RH; LW; RHAG; S, s, U; Fy5)
Recessive Lu(a–b–)	LU system
Dominant Lu(a–b–) [*In(Lu)*]	(Weak LU; KN; IN; P1; MER2; AnWj)
X-Linked Lu(a–b–)	(Weak LU system; I; strong i)
K_0	KEL system (Kx increased)
K_{mod}	(Weak KEL system; Kx increased)
Kp(a + b–)	(Weak KEL system; Kx slightly increased)
Fy^X	(Weak Fy^b often requires adsorption/elution for detection, Fy3, Fy6 weak in homozygotes)
Recessive Jk(a–b–)	JK system
Dominant Jk(a–b–) [*In(Jk)*]	(Very weak JK antigens)
Gy(a–)	DO system
Hy–	Hy, Jo^a (Weak Gy^a, Do^b, DOYA, DOMR, and sometimes Jo^a)
McLeod	Kx (Weak KEL system)
Leach (Ge:–2,–3,–4)	GE system (Weak KEL system)
Gerbich (Ge:–2,–3,4)	Ge2, Ge3 [Weak KEL system (some)]
Yus (Ge:–2,3,4)	Ge2
Inab	CROM system
Dr(a–)	Dr^a (dramatically weak CROM system)
Helgeson	KN system, Cs^a
Vel–	ABTI (can be weak)
ABTI–	(Vel)

Biosynthetic pathways



Antigens with lactosylceramide as a precursor

High prevalence antigens absent (and selected phenotypes) in certain ethnic populations

Phenotype	Population (Any = may be found in any population; >=more prevalent than)
AnWj–	Transient in any >>Israeli Arabs (inherited type)
At(a–)	Blacks
Cr(a–)	Blacks
Di(b–)	South Americans > Native Americans > Japanese
DISK–	Dutch > Europeans > Any
Dr(a–)	Jews from Bukhara > Japanese
En(a–)	Finns > Canadians > English > Japanese
Es(a–)	Mexicans, South Americans, Blacks
Fy(a–b–)	Blacks >> Arabs/Jews > Mediterraneans >> Caucasians
Ge:–2,–3 (Gerbich phenotype)	Papua New Guineans >> Melanesians >> Caucasians > Any
Ge:–2,3 (Yus phenotype)	Mexicans > Israelis > Mediterraneans > Any
Ge:–2,–3,–4 (Leach phenotype)	Any
GUTI–	Chileans
Gy(a–)	Eastern Europeans (Romany) > Japanese
hr^B–	Blacks
hr^S–	Blacks
Hy–	Blacks
IFC (Cr_{null}, Inab)	Japanese > Any
In(b–)	Indians > Iranians > Arabs
Jk(a–b–)	Polynesians >> Finns > Japanese > Any
Jo(a–)	Blacks

(*Continued*)

(Continued)

Phenotype	Population (Any = may be found in any population; >=more prevalent than)
Jr(a–)	Japanese > Asians > Europeans > Bedouin Arabs > Any
Js(b–)	Blacks
k–	Caucasians >> Any
K_0 (K_{null})	Reunion Islanders > Finns > Japanese > Any
K12 –	Caucasians
K14–	French-Cajuns
K22–	Israelis
KCAM–	Blacks >>> Any
Kn(a–)	Caucasians > Blacks > Any
Kp(b–)	Caucasians > Japanese
KUCI–	Native Americans
Lan–	Caucasians > Japanese > Blacks > Any
Lu(a–b–)	Any
Lu20–	Israelis
Lu21–	Israelis
LW(a–b–)	Transient in any >> inherited type in Canadians
LW(a–)	Balts
MAM–	Arabs > Any
MAR–	Finns > Any
McC(a–)	Blacks > Caucasians > Any
MER2–	Indian Jews, Turks, Portugese
M^kM^k	Swiss > Japanese
O_h (Bombay)	Indians > Japanese > Any
Ok(a–)	Japanese
P–	Japanese > Finns > Israelis > Any
Para-Bombay	Reunion Islanders > Indians > Any

(*Continued*)

(Continued)

Phenotype	Population (Any = may be found in any population; >=more prevalent than)
PEL–	French-Canadians
PP1P^{k}–	Swedes > Amish > Israelis > Japanese > Any
Sl(a–)	Blacks >> Caucasians > Any
Tc(a–b+c–)	Blacks
Tc(a–b–c+)	Caucasians
SERF–	Thais
U– and S–s–U+^{var}	Blacks
UMC–	Japanese
Vel–	Swedes > Any
WES(b–)	Finns > Blacks > Any
Yk(a–)	Caucasians > Blacks > Any
Yt(a–)	Arabs > Jews > Any

Low-prevalence antigens present in certain ethnic populations

Phenotype	Population (Any = may be found in any population; >= more prevalent than)
An(a+)	Finns
Be(a+)	Germans > Poles
Bp(a+)	English, Italians
Cl(a+)	Scottish, Irish
Crawford+	Blacks > Hispanic
C^{W}+	Latvians > Finns > Caucasians
C^{X}+	Finns > Caucasians > Somalis
DAK+	Blacks >>> Caucasians
DANE+	Danes > Europeans

(*Continued*)

(Continued)

Phenotype	Population (Any = may be found in any population; >= more prevalent than)
Dantu+	Blacks
Di(a+)	South American Indians > Japanese > Native Americans > Chinese, Poles
Evans+	Celts >>> Any
E^W+	Germans > Any
FPTT+	Any
FORS1+	English
Fr(a+)	Mennonites
Go(a+)	Blacks
HAG+	Israelis
He+	Xhosas > Blacks
Hg(a+)	Welsh > Australians
Hil+	Chinese > Any
Hop+	Thais > Any
In(a+)	Arabs > Iranians > Indians > Any
JAL+	English, French-speaking Swiss, Brazilians, Blacks
Jn(a+)	Poles, Slovaks
Js(a+)	Blacks
K+	Arabs > Iranian Jews > Caucasians > Any
K24+	French-Cajuns
Kn(b+)	Caucasians > Blacks
Kp(a+)	Caucasians > Any
Kp(c+)	Japanese
KREP+	Poles
Ls(a+)	Blacks > Finns > Any
Lu14+	English > Danes > Any
LW(b+)	Estonians > Finns > Balts > Europeans
MARS+	Choctaw tribe of Native Americans
M^c+	Europeans
M^g+	Swiss > Sicilians > Any
Mi(a+)	Thais > Taiwanese > Chinese > Any
MINY+	Thais > Taiwanese > Chinese > Any
Mit+	Western Europeans

(*Continued*)

(Continued)

Phenotype	Population (Any = may be found in any population; >= more prevalent than)
Mo(a+)	Belgians, Norwegians
Mt(a+)	Thais > Swiss > Caucasians > Blacks
Mur+	Thais > Taiwanese > Chinese > Any
MUT+	Chinese > Any
NFLD+	French Canadians, Japanese
NOR+	Poles
Ny(a+)	Norwegians > Swiss > Any
Or+	Japanese, Australians, Blacks, Jamaicans
Os(a+)	Japanese
Rd+	Danes > Canadians > Jews > Blacks > Any
Rh32+	Blacks > Caucasians > Japanese
Rh33+	Germans > Caucasians
Rh35+	Danes
Rh42+	Blacks
SAT+	Japanese
Sc2+	Mennonites > Northern Europeans
St(a+)	Japanese > Asian > Caucasians >>> Any
STEM+	Blacks
Tc(b+)	Blacks
Tc(c+)	Caucasians
TSEN+	Thais > Any
Ul(a+)	Finns > Japanese
V+	Blacks
Vr+	Dutch
VS+	Bantus > Blacks
Vw+	Swiss > Caucasians
WARR+	Native Americans
Wb+	Welsh > Australians
Wd(a+)	Hutterites
WES(a+)	Finns > Blacks
Wu+	Scandinavians > Dutch > Blacks
Yt(b+)	Arabs > Jews > Europeans > Any

Clinical significance of some alloantibodies to blood group antigens [3,4]

Usually clinically significant	Sometimes clinically significant	Clinically insignificant if not reactive at 37°C	Generally clinically insignificant
A and B	AnWj	A1	Chido/Rodgers
Diego	At^a	H	Cost
Duffy	Colton	Le^a	JMH
H in O_h	Cromer	Lutheran	HLA/Bg
Kell	Dombrock	M, $N^{\dagger}$	Knops
Kidd	Gerbich	P1	Le^b
P	Indian	Sd^a	Xg^a
PP1P^k	Jr^a		
Rh	Kx		
S, s, U	Lan		
Vel	Landsteiner-Wiener		
	Scianna		
	Yt^a		

†Rule out that 37°C reactivity is not due to carry-over agglutination.

Characteristics of some blood group alloantibodies

Antibody specificity	IgM (direct)	IgG (indirect)	Clinical transfusion reaction	HDFN
ABO	Most	Some	Immediate Mild to severe	Common Mild to moderate
Rh	Some	Most	Immediate/delayed Mild to severe	Common Mild to severe
Kell	Some	Most	Immediate/delayed	Sometimes

(*Continued*)

(Continued)

Antibody specificity	IgM (direct)	IgG (indirect)	Clinical transfusion reaction	HDFN
			Mild to severe	Mild to severe
Kidd	Few	Most	Immediate/delayed Mild to severe	Rare; mild
Duffy	Rare	Most	Immediate/delayed Mild to severe	Rare; mild
M	Some	Most	Delayed (rare)	Rare; usually mild
N	Most	Rare	None	None
S	Some	Most	Delayed/mild	Rare; mild to severe
s	Rare	Most	Delayed/mild	Rare; mild to severe
U	Rare	Most	Immediate/delayed Mild to severe	Rare; mild to severe
P1	Most	Rare	None (rare)	None
Lutheran	Some	Most	Delayed	Rare; mild
Le^a	Most	Few	Immediate (rare)	None
Le^b	Most	Few	None	None
Diego	Some	Most	Delayed; None to severe	Mild to severe
Colton	Rare	Most	Delayed; mild	Rare; mild to severe
Dombrock	Rare	Most	Immediate/delayed Mild to severe	Rare; mild
LW	Rare	Most	Delayed; none to mild	Rare; mild
Yt^a	Rare	Most	Delayed (rare); mild	None
I	Most	Rare	None	None
Ch/Rg	Rare	Most	Anaphylactic (3)	None
JMH	Rare	Most	Delayed (rare)	None
Knops	Rare	Most	None	None
Xg^a	Rare	Most	None	None

Antigen-negative prevalence for some polymorphic antigens

System	Antigen	Prevalence of antigen-negativity	
		Caucasian	Black
Rh	D	0.15	0.08
	C	0.32	0.73
	E	0.71	0.78
	c	0.20	0.04
	e	0.02	0.02
	f	0.35	0.08
	C^w	0.98	0.99
	V	>0.99	0.70
	VS	>0.99	0.73
MNS	M	0.22	0.26
	N	0.30	0.25
	S	0.48	0.69
	s	0.11	0.06
	M–S–	0.15	0.19
	M–s–	0.01	0.02
	N–S–	0.10	0.16
	N–s–	0.06	0.02
P1PK	P1	0.21	0.06
Lewis	Le^a	0.78	0.77
	Le^b	0.28	0.45
Lutheran	Lu^a	0.92	0.95
	Lu^b	<0.01	<0.01
Kell	K	0.91	0.98
	k	0.002	<0.001
	Kp^a	0.98	>0.99
	Kp^b	<0.01	<0.01
	Js^a	>0.99	0.80
	Js^b	<0.001	0.01
Duffy	Fy^a	0.34	0.90
	Fy^b	0.17	0.77
Kidd	Jk^a	0.23	0.08
	Jk^b	0.26	0.51
Dombrock	Do^a	0.33	0.45
	Do^b	0.18	0.11
Colton	Co^a	<0.001	<0.001
	Co^b	0.90	0.90

To determine the average number of blood donor samples to screen when searching for antigen-negative units, multiply the antigen-negative prevalence for each antigen, and divide the resultant percentage into 100.
For example, to screen for blood for a patient with anti-Fy^a + anti-K + anti-S:

$(0.34) \times (0.91) \times (0.48) = 0.148521$
$0.148521 \times 100 = 14.851$
$100 \div 14.851 = 6.7$ rounds up to 7

Thus, approximately 1 in 7 (or 14 in 100) donor samples will lack Fy^a, K, and S antigens.

Potentially useful information for problem-solving in immunohematology

Available information	Considerations
Patient demographics	Diagnosis, age, sex, ethnicity, transfusion, and/or pregnancy history, drugs, IV fluids (Ringer's lactate, IV-IgG, Rh-immune globulin, other plasma-containing products, anti-lymphocyte globulin (ALG), anti-thymocyte globulin (ATG), infections, malignancies, hemoglobinopathies, stem cell transplantation
Initial serological results	ABO, Rh, DAT, phenotype, antibody detection results, autologous control, cross-match results
Hematology/chemistry values	Hemoglobin, hematocrit, bilirubin, LDH, reticulocyte count, haptoglobin, hemoglobinuria, albumin:globulin ratio, RBC morphology
Sample characteristics	Site and technique of collection, age of sample, anticoagulant, hemolysis, lipemic, color of serum/plasma, agglutinates/aggregates in the sample
Other	Check records in current and previous institutions for previously identified antibodies
Antibody identification	Auto control, phase of reactivity, potentiator (saline, albumin, LISS, PEG), reaction strength, effect of chemicals on antigen (proteases, thiol reagents), pattern of reactivity (single antibody or mixture of antibodies), characteristics of reactivity (mixed field, rouleaux), hemolysis, preservatives/antibiotics in reagents

Alloantibodies that may have *in vitro* hemolytic properties

Anti-A, -B, -A,B, -H (in O_h people), -I, -i, -Le^a, -Le^b, -$PP1P^k$, -P, -Jk^a, -Jk^b, -Jk3, -Ge3, -Vel, and rare examples of anti-Sc1, -Lan, -Jr^a, -Co3, -Emm, and -Milne.

Conditions associated with suppression (sometimes total) or with alteration of antigen expression

Condition	Antigens affected
Pregnancy	ABO; H; I; LE; LW; P1; JMH; Sd^a; some Jk^a; Gy^a; AnWj
Carcinoma	ABO; H; I; P1; KN
Leukemia	ABO; H; I; RH; Yt^a; CO (chromosome 7 rearrangements)
Infection	ABO; A with appearance of Tn, A with appearance of acquired B; T activation; H; I; K
Hodgkin's lymphoma	ABO; H; LW
LADII (CDG-II)	ABO; H; LE
PNH	CROM; YT; DO; MER2; JMH; Emm
CDA	CO; LW; IN
AIHA	En^a, U; RH; KEL; JK; DI; LW; SC; GE; Vel; AnWj
SLE	CH/RG; KN; Yt^a
Hemopoietic stress	ABO; H; I (concomitant increased expression of i)
Diseases with increased clearance of immune complexes (e.g. AIDS)	KN
Old age	ABO; H; JMH
South-East Asian ovalocytes	En^a, S, s, U; Di^b, Wr^b; D, C, e; Kp^b; Jk^a, Jk^b; Xg^a; LW; $Sc1^5$

Causes of apparent *in vivo* hemolysis

Immune

ABO incompatibility
Clinically significant alloantibody
Anamnestic alloantibody response
Autoimmune hemolytic anemia
Cold agglutinin disease
HDFN
Drug-induced hemolytic anemia

Polyagglutination (sepsis T-active plasma)
Paroxysmal cold hemoglobinuria
TTP/HUS*-microangiopathic process

*Thrombotic thrombocytopenic purpura/hemolytic uremic syndrome.

Non-immune

Mechanical
- Poor sample collection
- Small-bore needle used for transfusion
- Excessive pressure during transfusion
- Malfunctioning blood warmer
- Donor blood exposed to excessive heat or cold
- Urinary catheter
- Crush trauma
- Prosthetic heart valves
- Aortic stenosis
- March hemoglobinuria

Microbial
- Sepsis
- Malaria
- Contamination of donor blood

Chemical
- Inappropriate solutions infused
- Drugs infused
- Serum phosphorus <0.2 mg/dL
- Water irrigation of bladder
- Azulfidine
- Dimethyl sulfoxide
- Venom (snake, bee, Brown Recluse spider)[6]
- Certain herbal preparations, teas, enemas

Inherent RBC abnormalities
- Paroxysmal nocturnal hemoglobinuria
- Sickle cell anemia
- Spherocytosis
- Hemoglobin H
- G6PD deficiency (in recipient or donor)

Warm autoantibodies to the following blood group antigens have been described[7,8] (listed alphabetically)

A, B	JMH
AnWj	K, k, Kp^b, Js^b, K13, Kell protein
Co3	Kx
Di^b, Wr^b	LW^a, LW^{ab}
En^a, U, M, N, S, Pr	Rh, in particular e, Rh17
Fy^b	Rx

Ge2, Ge3	Sc1
H	Vel
I^T	Yt^a
Jk^a, Jk^b, Jk3	Xg^a

Target antigen suppression

In some autoimmune cases, the target antigen may be weakened to the extent that the patient's RBCs are negative in the DAT. The following antigens have been implicated[9] (listed alphabetically):

AnWj	Ge3	JMH	Rh	U
Co3	Jk^a	Kp^b	Sc1	Vel
En^a	Jk^b	LW	Sc3	

Drugs associated with immune hemolytic anemia and/or positive DAT in which drug-dependent antibodies were detected[8,10–13]

Drugs can cause the production of antibodies that may be against the drug itself, RBC membrane components or an antigen formed by the drug and the RBC membrane. Such antibodies may cause a positive DAT, immune hemolytic anemia or both. A drug may also cause a positive DAT through non-immunologic protein adsorption onto the RBC. The mechanisms involved eliciting an immune response to drugs are not well-understood, and various theories have been proposed.
In the table[10], when an antibody is indicated to react by two methods, it does not necessarily mean that all examples of antibodies to that drug were detected by both methods.

Drug (alternative name)	Therapeutic category	HA	Positive DAT	Method of detecting serum antibody		Reactive without drug added *in vitro*
				Drug-coated RBCs	Serum + drug + RBCs	
Aceclofenac	NSAID	✓	✓	–	✓	–
Acetaminophen (Paracetamol)	NSAID	✓	✓	–	✓	–
Acyclovir	Anti-viral	✓	✓	✓	–	–
Aminopyrine (Piramidone)	NSAID	✓	–	✓	–	–
Amoxicillin	Anti-microbial	✓	✓	✓	–	–

(*Continued*)

(Continued)

Drug (alternative name)	Therapeutic category	HA	Positive DAT	Method of detecting serum antibody		Reactive without drug added *in vitro*
				Drug-coated RBCs	Serum+ drug + RBCs	
Amphotericin B	Anti-microbial	✓	✓	–	✓†	–
Ampicillin	Anti-microbial	✓	✓	✓	✓	–
Antazoline	Anti-histamine	✓	✓	–	✓	–
Aspirin	Analgesic, antipyretic, anti-inflammatory	✓	–	–	✓	–
Azapropazone (Apazone)	Anti-inflammatory, analgesic	✓	✓	✓	–	✓
Buthiazide (Butizide)	Diuretic, anti-hypertensive	✓	✓	–	✓†	–
Carbimazole	Anti-thyroid	✓	✓	✓	✓	✓
Carboplatin	Anti-neoplastic	✓	✓	✓	✓	✓
Carbromal	Sedative, hypnotic	–	✓	✓	–	–
Catechin [(+)-Cyanidanol-3] (Cianidanol)	Anti-diarrheal	✓	✓	✓	✓†	✓
Cefamandole	Anti-microbial	✓	✓	✓	–	–
Cefazolin	Anti-microbial	✓	✓	✓	–	–
Cefixime	Anti-microbial	✓	–	✓	✓	–
Cefotaxime	Anti-microbial	✓	✓	✓	✓	✓^
Cefotetan	Anti-microbial	✓	✓	✓¶	✓	✓
Cefoxitin	Anti-microbial	✓	✓	✓	✓	✓
Cefpirome	Anti-bacterial	–	✓	–	✓	–
Ceftazidime	Anti-microbial	✓	✓	✓	✓	✓
Ceftizoxime	Anti-microbial	✓	✓	✓	✓	✓^
Ceftriaxone	Anti-microbial	✓	✓	–	✓†	✓^
Cefuroxime	Anti-bacterial	✓	✓	✓	–	–

(*Continued*)

(Continued)

Drug (alternative name)	Therapeutic category	HA	Positive DAT	Method of detecting serum antibody		Reactive without drug added *in vitro*
				Drug-coated RBCs	Serum + drug + RBCs	
Cephalexin	Anti-microbial	✓	✓	✓¶	–	–
Cephalothin	Anti-microbial	✓	✓	✓¶	✓	–
Chloramphenicol	Anti-bacterial	✓	✓	✓	–	✓
Chlorinated hydrocarbons	Insecticides	✓	✓	✓	✓	✓
Chlorpromazine	Anti-emetic, anti-psychotic	✓	✓	✓	–	✓
Chlorpropamide	Anti-diabetic	✓	✓	–	✓	✓^
Cimetidine[14]	Anti-ulcerative	✓	✓	✓	✓	–
Ciprofloxacin	Anti-bacterial	✓	✓	–	✓	✓
Cisplatin (Cisdiaminodi-chloroplatinum)	Anti-neoplastic	✓	✓	✓¶	✓	–
Cloxacillin	Anti-bacterial	–	✓	–	–	✓
Cyclofenil	Gonad-stimulating principle	✓	✓	–	✓	✓
Cyclosporin (Cyclosporine)	Immuno-suppressant	✓	✓	✓	–	✓
Dexchlor-pheniramine maleate (Chlorpheniramine)	Anti-histaminic	✓	✓	–	✓	–
Diclofenac	NSAID	✓	✓	✓	✓†	✓^
Diethylstilbestrol (Stilboestrol)	Estrogen	✓	✓	–	✓	–
Dipyrone	NSAID	✓	✓	✓	✓	–
Erythromycin	Anti-microbial	✓	✓	✓	–	–
Etodolac	NSAID	✓	✓	–	✓†	–
Ethambutol	Anti-bacterial	✓	✓	✓	✓	–
Fenoprofen	NSAID	✓	✓	–	✓	✓^

(*Continued*)

(Continued)

Drug (alternative name)	Therapeutic category	HA	Positive DAT	Method of detecting serum antibody		Reactive without drug added *in vitro*
				Drug-coated RBCs	Serum+ drug + RBCs	
Fluconazole	Anti-fungal	✓	✓	✓	✓	–
Fluorescein	Injectable dye	✓	✓	✓	✓	✓^
Fluorouracil	Anti-neoplastic	✓	✓	–	✓	–
Furosemide	Diuretic	–	✓	–	✓	–
Glafenine (Glaphenine)	Analgesic	✓	✓	–	–	✓
Hydralazine	Anti-hypertensive	✓	✓	✓	–	–
Hydro chlorothiazide	Diuretic	✓	✓	✓	✓	✓^
Hydrocortisone[15]	Glucocorticoid	✓	✓	✓	✓	–
9-Hydroxy-methyl-ellipticinium (Elliptinium acetate)	Anti-neoplastic	✓	✓	–	✓	–
Ibuprofen	NSAID	✓	✓	–	✓	✓
Imatinib mesylate	Anti-neoplastic	✓	✓	✓	–	–
Insulin	Anti-diabetic	✓	✓	✓	–	–
Isoniazid	Anti-microbial	✓	✓	✓	✓	–
Latamoxef (Moxalactam)	Anti-microbial	✓	✓	–	–	✓
Levofloxacin (Ofloxacin)	Anti-bacterial	✓	✓	✓	✓	✓
Mefloquine	Anti-microbial	✓	✓	✓	✓	✓^
Melphalan	Anti-neoplastic	✓	–	–	✓	–
6-Mercaptopurine	Anti-neoplastic	✓	✓	✓	–	–
Methadone	Analgesic	–	✓	✓	–	–
Methotrexate	Anti-neoplastic, anti-rheumatic	✓	✓	✓	✓	✓

(*Continued*)

(Continued)

Drug (alternative name)	Therapeutic category	HA	Positive DAT	Method of detecting serum antibody		Reactive without drug added *in vitro*
				Drug-coated RBCs	Serum + drug + RBCs	
Metrizoate-based radiographic contrast media		✓	✓	✓	✓	✓
Minocycline	Anti-bacterial	✓	✓	–	✓	–
Nabumetone analgesic	Anti-inflammatory	✓	✓	–	✓†	✓
Nafcillin	Anti-microbial	✓	✓	✓	✓–	–
Naproxen	Anti-inflammatory, analgesic, anti-pyretic	✓	✓	–	✓	–
Nifedipine[16]	Anti-hypertensive	✓	✓	–	✓	–
Nitrofurantoin	Anti-bacterial	✓	–	–	✓	–
Nomifensine§	Anti-depressant	✓	✓	–	✓†	✓^
Norfloxacin	Anti-microbial	–	✓	✓	–	–
Oxaliplatin	Anti-neoplastic	✓	✓	✓¶	✓	✓^
p-Aminosalicylic acid (PAS) (para-aminosalicylsaure)	Anti-microbial	✓	✓	–	✓	–
Penicillin G	Anti-microbial	✓	✓	✓	✓	–
Phenacetin (Acetophenetidin)	NSAID	✓	✓	–	✓	✓
Phenytoin (Fenitoine)	Anti-convulsant, anti-arrhythmic	✓	✓	✓	–	–
Piperacillin	Anti-microbial	✓	✓	✓	✓	✓^
Probenecid	Uricosuric	✓	✓	–	✓	✓^
Propyphenazone	NSAID	✓	✓	–	✓	–
Pyrazinamide	Anti-bacterial	✓	✓	✓	✓	–
Pyrimethamine (Pirimetamine)	Anti-microbial	✓	✓	✓	–	–
Quinidine	Anti-arrhythmic, anti-microbial	✓	✓	✓	✓	✓^

(*Continued*)

(Continued)

Drug (alternative name)	Therapeutic category	HA	Positive DAT	Method of detecting serum antibody		Reactive without drug added *in vitro*
				Drug-coated RBCs	Serum+ drug + RBCs	
Quinine	Anti-microbial	✓	–	–	✓	✓
Ranitidine	Anti-ulcerative	✓	✓	✓	✓	–
Rifabutin	Anti-bacterial	✓	✓	–	✓	–
Rifampin (Rifampicin)	Anti-bacterial	✓	✓	✓	✓	✓^
Stibophen	Anti-microbial	✓	✓	–	✓	–
Streptokinase	Thrombolytic	✓	✓	✓	–	✓
Streptomycin	Anti-microbial	✓	✓	✓	✓	✓
Sulfasalazine	Anti-inflammatory	✓	✓	–	✓	–
Sulfisoxazole	Anti-bacterial	✓	✓	✓	✓	–
Sulindac	Anti-inflammatory	✓	✓	✓	✓	✓^
Suprofen	NSAID	✓	✓	–	✓	✓^
Tartrazine	Colorant	✓	✓	✓	✓	–
Teicoplanin	Anti-microbial	✓	✓	–	✓	✓
Temafloxacin§	Anti-microbial	✓	✓	–	✓	–
Teniposide	Anti-neoplastic	✓	✓	–	✓	✓
Tetracycline	Anti-microbial	✓	✓	✓	–	–
Thiopental sodium	Anesthetic	✓	–	–	✓	–
Ticarcillin	Anti-microbial	✓	✓	✓	–	✓
Tolbutamide	Anti-diabetic	✓	✓	✓	–	–
Tolmetin	NSAID	✓	✓	–	✓	✓^
Triamterene	Diuretic	✓	✓	✓	✓	–
Trimellitic anhydride	Used in preparation of resins, dyes, adhesives, etc.	✓	–	✓	–	–
Trimethoprim and sulfamethoxazole	Anti-bacterial	✓	✓	✓	✓	✓

(*Continued*)

(Continued)

Drug (alternative name)	Therapeutic category	HA	Positive DAT	Method of detecting serum antibody		Reactive without drug added *in vitro*
				Drug-coated RBCs	Serum + drug + RBCs	
Vancomycin	Anti-bacterial	✓	✓	–	✓	–
Zomepirac	NSAID	✓	✓	–	✓	✓

HA = Hemolytic anemia; NSAID = Nonsteroidal anti-inflammatory drug.
[†]Positive or gives the strongest reactions when the drug metabolite is present.
[§]No longer manufactured.
[¶]Associated with nonimmunologic protein adsorption.
[^]Positive, possibly due to the presence of circulating drug or drug–antibody immune complexes.

Drugs associated with cases of immune hemolytic anemia and/or positive DAT caused by drug-independent antibodies (autoantibodies)

Drug (alternative name)	Therapeutic category	HA	Positive DAT	More evidence needed
Captopril	Anti-hypertensive	✓	✓	✓
Chaparral	Herbal	–	✓	✓
Cladribine (2-chlorode-oxyadenosine)	Anti-neoplastic	✓	✓	–
Fenfluramine	Anorexic	✓	✓	✓
Fludarabine	Anti-neoplastic	✓	✓	–
Interferon	Anti-neoplastic, anti-viral	✓	✓	✓
Interleukin-2	Anti-neoplastic	✓	✓	✓
Ketoconazole	Anti-fungal	✓	✓	✓

(*Continued*)

(Continued)

Drug (alternative name)	Therapeutic category	HA	Positive DAT	More evidence needed
Lenalidomide	Immunomodulatory	✓	✓	✓
Levodopa (L-dopa)	Anti-parkinsonian	✓	✓	–
Mefenamic acid	NSAID	✓	✓	–
Mesantoin (Mephenytoin)	Anti-convulsant	✓	✓	✓
Methyldopa	Anti-hypertensive	✓	✓	–
Nalidixic acid	Anti-bacterial	✓	✓	✓
Procainamide	Anti-arrhythmic	✓	✓	–
Tacrolimus	Immunosuppressant	✓	✓	✓

HA = Hemolytic anemia.

Drugs associated with the detection of non-immunologic protein adsorption onto RBCs

Drug (alternative name)	Therapeutic category	HA	DAT	Drug-dependent antibody(ies) also detected
Cefotetan	Anti-microbial	✓	✓	✓
Cephalothin	Anti-microbial	✓	✓	✓
Cisplatin	Anti-neoplastic	✓	✓	✓
Clavulanate potassium (Clavulanic acid)	β-Lactamase inhibitor	–	✓	–
Diglycoaldehyde (INOX)	Anti-neoplastic	–	✓	–
Oxaliplatin	Anti-neoplastic	✓	✓	✓
Sulbactam	β-Lactamase inhibitor	✓	✓	–
Suramin	Anti-helminthic, anti-protozoal	–	–	–
Tazobactam	β-Lactamase inhibitor	✓	✓	–

HA = Hemolytic anemia.

Blood group systems and their gene products

Carbohydrate based blood group systems

System	Gene product
ABO	*N*-acetylgalactosaminyltransferase (A glycosyltransferase) or galactosyltransferase (B glycosyltransferase)
P1PK	Galactosyltransferase
LE	Fucosyltransferase
H	Fucosyltransferase
I	*N*-acetylglucosaminyltransferase
GLOB	*N*-acetylgalactosaminyltransferase
FORS	*N*-acetylgalactosaminyltransferase

Blood group systems located on single pass membrane proteins

System	Gene product	Number of amino acids	N-terminus	Function in RBCs
MNS	Glycophorin A Glycophorin B	131 72	Exofacial Exofacial	Carrier of sialic acid, which contributes to the negatively charged barrier. Complement regulation. Facilitates membrane assembly of band 3.
GE	Glycophorin C Glycophorin D	128 107	Exofacial Exofacial	Carrier of sialic acid (see MNS). Interacts with band 4.1 and p55 in RBC membrane to maintain RBC shape.
KEL	Kell glycoprotein	732	Cytoplasmic	Zinc endopeptidase that cleaves big endothelin.
LU	Lutheran glycoprotein	597	Exofacial	Binds laminin.
XG	Xg^a glycoprotein	180	Exofacial	Unknown.
	CD99	163	Exofacial	Adhesion molecule.
LW	LW glycoprotein	241	Exofacial	Ligand for integrins.
IN	CD44	341	Exofacial	Adhesion molecule that binds to hyaluronic acid.
KN	CD35 (CR1)	1998	Exofacial	Complement regulation.
Ok^a	CD147	248	Exofacial	Possible cell–cell adhesion.
SC	ERMAP	446	Exofacial	Possible adhesion.

Blood group systems located on multipass membrane proteins

System	Gene product	Number of amino acids	Predicted number of spans	Function in RBCs
RH	RhD protein RhCE protein	417 417	12 12	The Rh/RhAG/band 3/ complex contributes to the RBC membrane structure and transports gases.
RHAG	RhAG	409	12	
FY	Fy glycoprotein (DARC)	336 (major product)	7^	Cytokine receptor for pro-inflammatory cytokines.
DI	Band 3 (AE1)	911	14	Anion transport (HCO_3^-/ Cl^-) essential for respiration).
CO	AQP1 (CHIP-1)	269	6	Water/CO_2 transport.
JK	Urea transporter	389	10	Urea transport.
XK	Kx glycoprotein	444	10	Possible neurotransmitter. Possible amino acid transporter.
RAPH	Tetraspanin	253	4	Cell adhesion, proliferation, differentiation.
GIL	AQP3	342	6	Glycerol/water/urea transport.
JR	ABCG2; breast cancer resistance protein	655	6	ATP-dependent transporter of a diverse range of substrates.
LAN	ABCB6	842	11	Mitochondrial transporter essential for heme biosythesis.

^=N-terminus oriented to exofacial surface, C-terminus to cytoplamic surface. All others are predicted to be oriented with both their N- and C-termini to the cytoplasmic aspect of the RBC membrane.

Blood group systems carried on glycosylphosphatidylinositol-linked proteins

System	Gene product	Number of amino acids	Function in RBCs
YT	Acetylcholinesterase	557	Enzymatic
CROM	CD55 (DAF)	347	Complement regulation
DO	Do glycoprotein	314	Possibly enzymatic
JMH	CD108	646	Adhesion molecule involved in cell migration

Blood group systems located on proteins adsorbed from the plasma

System name	Component	Antigen location	Function in RBCs
CH/RG	C′ component 4 (C4)	C4d fragment	Complement regulation

Proteins altered on Rh_{null} RBCs

Protein	Gene location	M_r	Copies per RBC	Comments
RhD/RhCE	1p36.11	30,000–32,000	100,000–200,000 for RhD/RhCE combined	Absent
RhAG	6p21.3	45,000–100,000	100,000–200,000	Absent
CD47	3q13	47,000–52,000	10,000–50,000	Reduced (~25% of normal)
LW	19p13.2	37,000–43,000	3,000–5,000	Absent
GPB	4q31.22	20,000–25,000	200,000	Reduced (30% of normal)
Duffy (Fy5)	1q23.2	35,000–45,000	6,000–13,000	Fy5 antigen absent

Blood Group Proteins, M_r, abundance, and selected reactive monoclonal antibodies (MAbs)

Blood group system	M_r (SDS-PAGE)	Approximate copy number/RBC	Conditions for immunoblotting	MAbs active by immunoblotting	Serology/flow cytometry MAbs
MNS (GPA)	43,000	800,000	R or NR	Sigma E3, many clones	BRIC256
MNS (GPB)	20–25,000	200,000	R or NR	Sigma E3, R1.3, Anti-N	
RH	30–32,000	100,000– 200,000, RhD and RhCE combined	NR Use 8 M Urea Do not boil	LOR15C9 (anti-D)	
LU	85,000	1,500–4,000	NR	BRIC224 (D1), BRIC221 (D4)	BRIC224 (D1), BRIC221 (D4)
KEL	93,000	3,500–18,000	NR	C-10 (R or NR), 195031 (NR only)	BRICs 18, 68, and 203, 4B10
FY	35–45,000	6,000–13,000	R or NR	MIMA107, MIMA29	Polyclonal antibodies available
JK	43,000	14,000	R or NR	MIMA128	
DI	95–105,000	1,000,000	R or NR	N-terminal: BIII-136, 2D5, BRIC170 C-terminal: BRIC155	BRIC6, BRAC18
YT	160,000	7,000–10,000		None available	Many clones
XG (Xg^a)	22–29,000	9,000	R or NR	NBL-1	
XG (CD99)	32,500	200–2,000	R or NR	12E7, MEM-131, BANRS1, MSG-B1	Many clones
SC	60–68,000	Not determined	NR	IgSF: 6F8, YS-6, C8. Intracellular: 10C132	C8
DO	47–58,000	Not determined	NR	MIMA52	MIMA52
CO	28,000 Glycosylated 40–60,000	120,000–160,000	R	Loop E: 7D11. C-terminal: 1A5F6, MIMA136	7D11

(*Continued*)

(Continued)

Blood group system	M_r (SDS-PAGE)	Approximate copy number/RBC	Conditions for immunoblotting	MAbs active by immunoblotting	Serology/flow cytometry MAbs
LW	37–43,000	D + 4,400 D– 2,800	NR	BS56, BS86	BS46, BS56, BS87
XK	37,000	1,000		None available	None available
GE (GPC)	40,000	135,000	R	BRIC4, BRIC10, E5, 1H3, 3H2007, BGRL-100, MIMA81	Many clones
GE (GPD)	30,000	50,000	R	BRAC11, (Human anti-Ge2)	BRAC11
CROM (CD55)	60–70,000	20,000	NR	CCP1: BRICs 128, 220, 230. CCP2: BRIC110. CCP3: BRIC216. CCP4: MEM-118. CCP2/3: MIMA28, MIMA69	CCP1: BRICs 128, 220, 230. CCP2: BRIC110. CCP3: BRIC216. CCP4: MEM-118.
KN (CR1)	220,000	20–1,500		LHR A-C: To5	Many clones
IN (CD44)	80,000	2,000–5,000	NR or R	NR: all clones R: only KZ-1, Hermes-3	Many clones
OK (CD147)	35–69,000	3,000	NR	D1: MEM-M6/1, HIM6 D2: MEM6/6, MIMA144	Many clones
RAPH (CD151)	40,000	Not determined	NR	IIG5a	IIG5a, TS151
JMH (CD108)	68–76,000	Not determined	NR	Sema7A: MEM-150 IgSF: 3D3, 1G1 Others: 9L98, 9G441.	MEM-150, KS-2, 310829
RHAG	45–100,000	100,000–200,000	NR Use 8 M Urea Do not boil	LA18.18, 2D10, MIMA77	

R = Reducing conditions; NR = Non-reducing conditions; D# = Domain number.

Changes in numbering of nucleotides and amino acids

For all alleles, the numbering for nucleotides and amino acids follows the ISBT system, i.e., nucleotides are counted as #1 being the "A" of the initiating "AUG," and amino acids are counted as #1 from the initiating methionine. This ISBT consistency policy means that the numbers for some nucleotides and amino acids may differ from those published.

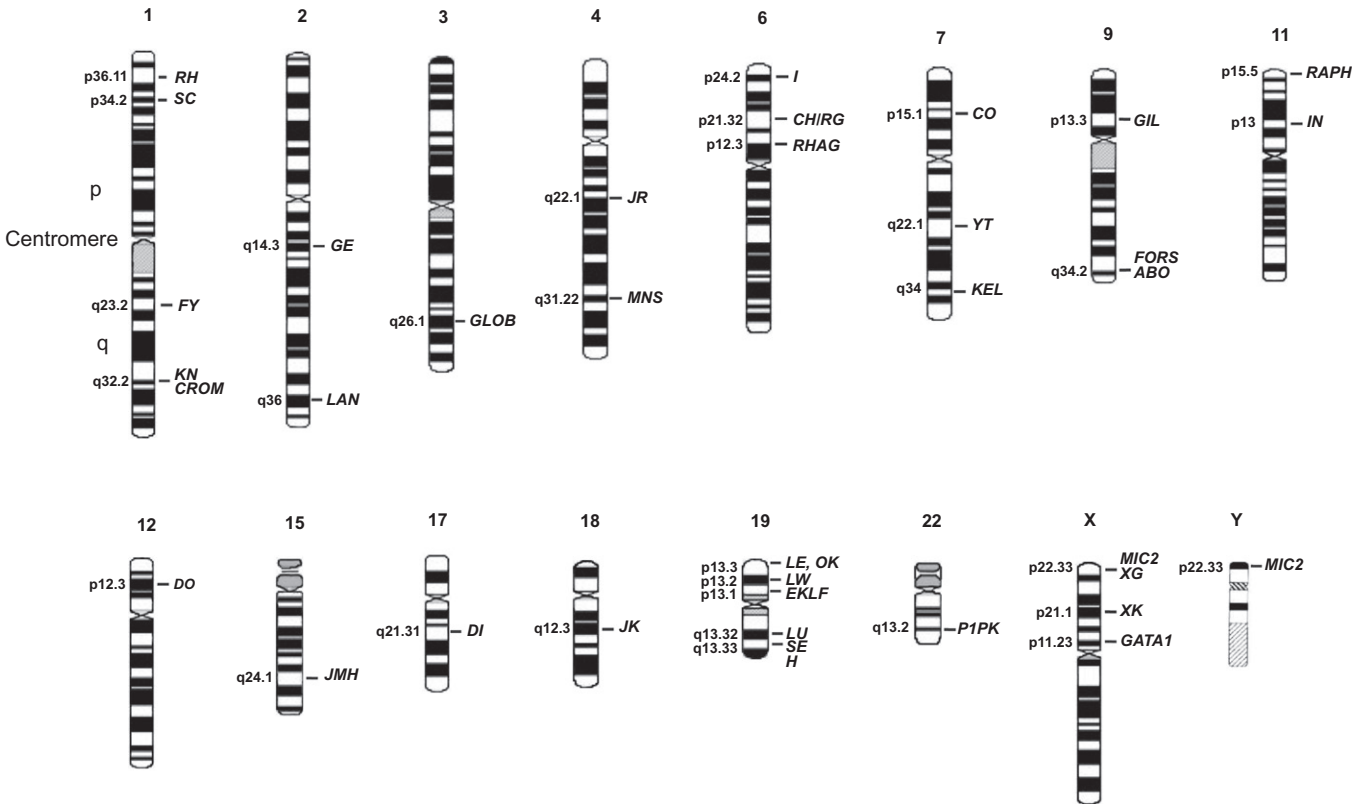
System	Nucleotide change	System	Amino acid change
KEL	−120	MNS	+19
KN	−27	FY	+2 from minor product
Kx	−82	LW CROM	+30 +34

Some causes of pseudo-discrepancies between genotype and phenotype

The gene is present, but the expected product is not detectable in the RBC membrane.

Event	Mechanism	Blood group phenotype
Transcription	Nt change in GATA box	Fy(b–) or Fy(a–)
Alternative splicing due to nt change in splice site	Partial/complete skipping of exon	S–s–; Gy(a–)
	Deletion of nts	Dr(a–)
Premature stop codon	Deletion of nt(s)→frame-shift	Fy(a–b–); D–; Rh_{null}
	Insertion of nt(s)→frame-shift	Ge: –2,–3,–4; Gy(a–)
	Nt change	D–; Co(a–b–); Fy(a–b–); r′; Gy(a–); K_0; McLeod
Amino acid change	Missense mutation	D–; Rh_{null}; K_0; McLeod
Reduced amount of protein	Missense mutation	Fy^X; Co(a–b–)
Hybrid genes	Cross-over	GP.Vw; GP.Hil; GP.TSEN
	Gene conversion	GP.Mur; GP.Hop; D− −; R_0^{Har}
Interacting protein	Absence of RhAG	Rh_{null}
	Absence of Kx	Kell antigens are weak
	Absence of aas 59 to 76 of GPA	Wr(b–)
	Absence of protein 4.1	Ge antigens are weak
Modifying gene	*EKLF* [*In*(*Lu*)]	Lu(a–b–)
	In(*Jk*)	Jk(a–b–)

Chromosomal location of genes encoding or influencing the expression of blood groups



Useful definitions

Absorbed	From; away
Adsorbed	Onto Thus, an antibody is *absorbed* from serum, but *adsorbed* onto RBCs. Another definition is that *absorbed* is a nonspecific term (as in "absorbed" by a sponge), while *adsorbed* is a specific reaction.
Allele	Alternative form(s) of a *gene* at a given locus (antigens cannot be allelic).
Antithetical	Refers to *antigens* produced by alleles (alleles cannot be antithetical).
Haplotype	A set of alleles of a group of closely linked genes, which are usually inherited together. People have haplotypes, RBCs do not.
Propositus	Singular male or index case (singular) regardless of sex.
Propositi	Plural male or index cases (plural) regardless of sex.
Proposita	Singular female.
Propositae	Plural female.
Proband	Index case regardless of sex.
Probands	Plural for index cases regardless of sex.
Transition	Change of purine (A, G) to purine or pyrimidine (C, T) to pyrimidine.
Transversion	Change between purine and pyrimidine (A or G to C or T).
Missense mutation	Nucleotide change leading to a change of amino acid (nonsynonymous).
Nonsense mutation	Nucleotide change leading to a stop codon.
Silent mutation	Nucleotide change that, due to redundancy in the genetic code, does not change the amino acid (synonymous).
Frameshift mutation	A change in DNA that occurs when the number of nucleotides inserted or deleted is not a multiple of three, so that every codon beyond the point of insertion or deletion is shifted during translation. This results in a novel sequence of amino acids and sooner or later a stop codon.
Northern blot	Analysis of RNA.
Southern blot	Analysis of DNA.
Western blot	Analysis of proteins.

Some lectins and their simple specificities[17,18]

Lectin	Common name	Carbohydrate-binding specificity
Arachis hypogaea	Peanut	D-Galβ(1–3)GalNAc > α-D-Gal
Dolichos biflorus	Horsegram	α-D-GalNAc >> α-D-Gal
Glycine max	Soybean	α-D-GalNAc > β-D-GalNAc > α-D-Gal
Griffonia simplicifolia I^	GS1	α-D-Gal > α-D-GalNAc
Griffonia simplicifolia II^	GS2	α-D-GlcNAc = β-D-GlcNAc
Helix pomatia	Edible snail	α-D-GalNAc > α-D-GlcNAc > α-D-Gal
Leonurus cardiaca	Motherwort	α/β-D-GalNAc
Phaseolus lunatus	Lima bean	α-D-GalNAc
Salvia horminum	Clary^^	α-D-GalNAc > β-D-GalNAc
Salvia sclarea	Clary^^	α-D-GalNAc
Ulex europaeus I	Gorse/furze	α-L-Fuc
Vicia cretica		D-Gal

^Previously known as *Bandeiraea simplicifolia* (BS) lectins.
^^Both *Salvia horminum* and *Salvia sclarea* are commonly known as clary, but they are botanically different.

Polyagglutination types, and the expected reactions with group O^ RBCs and lectins[18,19]

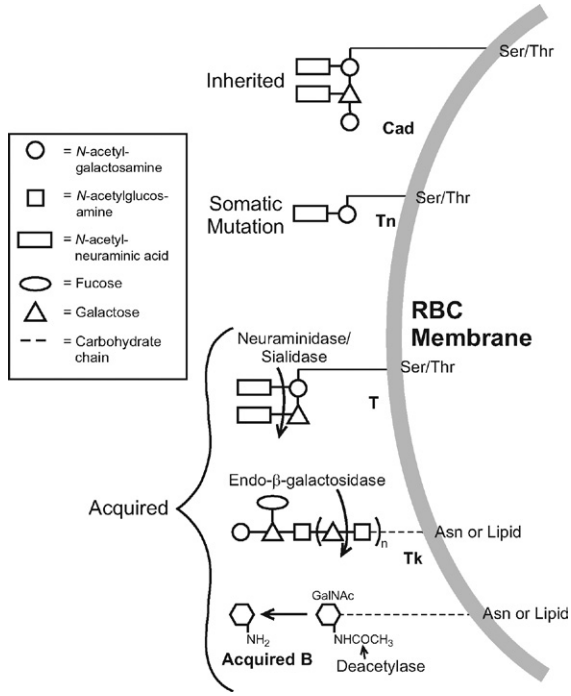
	Type of polyagglutination											
Lectin	**Acquired B**	**T**	**Tk**	**Th**	**Tx**	**Tn**	**Tr**	**Cad**	**NOR**	**VA**	**HEMPAS**	**HbM^^**
Arachis hypogaea	0	+	+	+	+	0	↓	0	0	0	0	↓↓
Dolichos biflorus	↓	0	0	0	0	+	0	+	0	0	0	0
Glycine max	0	+	0	0	0	+	+	0	0	0	0	+
GSI	+	0	0	0	0	+	+	0	0	0	0	0
GSII	0	0	+	0	0	0	+	0	0	0	0	+
Helix pomatia	+	+	0	NT	NT	+	↓	+	0	+	+	+
Leonurus cardiaca	0	0	0	0	0	0	NT	+	0	0	0	0
Phaseolus lunatus	+	0	0	0	0	0	0	0	0	0	0	NT
Salvia horminum	0	0	0	0	0	+	↓	+	0	0	0	↓↓
Salvia sclarea	0	0	0	0	0	+	↓	0	0	0	0	0
Ulex europaeus	+	↑↑	↓	+	+	↑	+	↓	+	↓	↓	↑↑
Vicia cretica	0	+	0	+	0	0	NT	0	0	0	0	↓↓

↓ (↓↓) Weaker than normal RBCs; ↑ (↑↑) Stronger than normal RBCs.
+Agglutination; 0 No agglutination.
^ Except acquired B, which occurs on group A; ^^HbM-Hyde Park.

Types of Polyagglutination

T	Neuraminidase made by some organisms (*Vibrio cholerae*, *Clostridium perfringens*, pneumococci, influenza virus) cleaves sialic acid (NeuAc) from RBC-bound disialylated alkali-labile tetrasaccharides, leaving Galβ1-3GalNAc-Ser/Thr, which is recognized by anti-T.
Tk	β-Galactosidases produced by some organisms (*Bacteroides fragilis*, *Aspergillus niger*, *Serratia marcescens*, *Candida albicans*) cleaves Gal from GlcNAc in complex carbohydrate structures including A, B, H, P1, I, active carbohydrate chains.

Diagrammatic representation of Cad, Tn, T, Tk, and acquired B



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