Chapter 7

OTHER BLOOD GROUP SYSTEMS, HUMAN LEUKOCYTE ANTIGENS, AND PLATELET ANTIGENS

Objectives (1 of 2)

- Identify the major antigens within the other blood group systems
- List the frequencies of the observed phenotypes and the association of phenotypes with ethnic group diversity
- Describe the biochemical characteristics of antigens within each blood group system

Objectives (2 of 2)

Describe the genetic mechanisms for antigens within each blood group system

Compare and contrast the serologic characteristics and clinical relevance of the antibodies associated with each blood group system

Identify unique characteristics of selected blood group systems regarding disease association and biologic functions

Kell Blood Group System

- Similar to the Rh system
- 2 major antigens
  - K (K1): less than 9% of the population
  - k (K2/cellano): more than 90% of the population
- The K and k antigens are antithetical
- Well developed at birth
- The K (K1) antigen is very immunogenic (second to the D antigen) in stimulating antibody production

Other Kell Antigens

Other antithetical antigens also exist in the Kell system

Analogous to the Rh system: *C/c* and *E/e*

Kp antigens

- Kp^a is a low-frequency antigen (only 2%)
- Kp^b is a high-frequency antigen (99.9%)

Js antigens

- Js^a (20% in blacks, 0.1% in Caucasians)
- Js^b is a high-frequency antigen (80% to 100%)

Kell Antigens

Kell antigens have disulfide-bonded regions on the glycoproteins

This makes them sensitive to **sulfhydryl reagents**

- 2-mercaptoethanol (2-ME)
- Dithiothreitol (DTT)
- 2-aminoethylisothiouronium bromide (AET)

K_0 or $Kell_{null}$ Phenotype

Lacks all Kell system antigens (K_0K_0)

Expresses related Kx antigen

As a result of red blood cell (RBC) immune stimulation, K_0 individuals can develop anti-Ku (Ku is on RBCs that have Kell antigens)

Kell Genetics

- Sets of alleles include
  - *K* and *k*
  - *Kp*^{a} and *Kp*^{b}
  - *Js*^{a} and *Js*^{b}
  - *KEL11* and *KEL17 (Wk*^{a}*)*
- High-incidence alleles
  - *k, Kp*^{b}*, Js*^{b}, and *KEL11*
- Low-incidence alleles
  - *K, Kp*^{a}*, Js*^{a}*, KEL17*

Table 7.3 Common Phenotypes and Frequencies in the Kell Blood Group System

	Frequency (%)	
Phenotype	White	Black
K–k+	91	98
K+k–	0.2	Rare
K+k+	8.8	2
Kp(a+b–)	Rare	0
Kp(a–b+)	97.7	100
Kp(a+b+)	2.3	Rare
Js(a+b–)	0	1
Js(a–b+)	100	80
Js(a+b+)	Rare	19

From Reid ME, Lomas-Francis C, Olsson MI: The blood group antigen facts book, ed 3, San Diego, CA, 2012, Academic Press.

Kell Antibodies

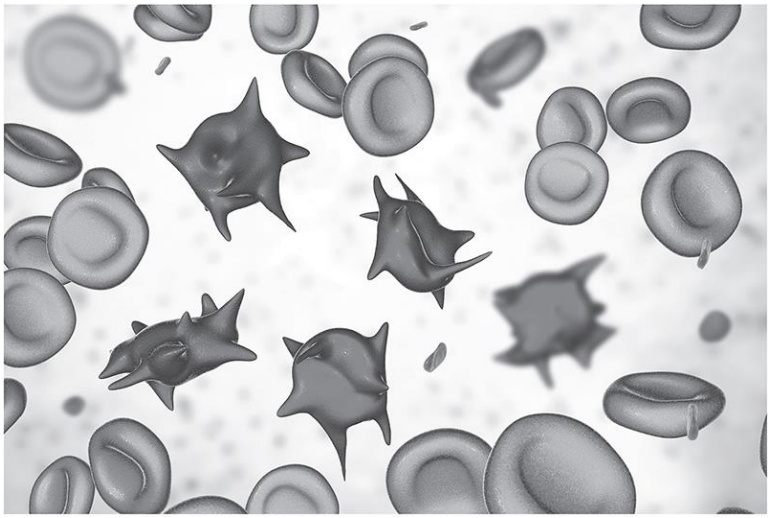
- Immunoglobulin G (IgG)
- RBC stimulated (transfusion or pregnancy)
- Agglutinate best in the indirect antiglobulin test (IAT)
- Usually do not bind complement
- Associated with hemolytic transfusion reactions (HTRs) and hemolytic disease of the fetus and newborn (HDFN)
- No effect when treated with enzymes
- Anti-K (K1) is the most common

Kx Blood Group System

Kx antigen is phenotypically related to the Kell system but is not genetically similar

Individuals who lack Kx antigen may demonstrate RBC abnormalities (**McLeod phenotype**)

Seen in males because it is inherited on the X chromosome



McLeod Syndrome

McLeod phenotype is attributed to McLeod syndrome

McLeod syndrome symptoms

- RBC abnormalities
- Muscular and neurologic defects
- Increased creatine kinase

Associated with **chronic granulomatous disease**

- Impaired phagocytosis (white blood cells [WBCs] engulf but cannot kill)

Duffy Blood Group System

Antigens are well developed at birth

Destroyed by enzymes

Fy^a and Fy^b

- Codominant alleles
- Most important for transfusion purposes

Duffy Antibodies

Anti-Fy^a and anti-Fy^b antibodies

- IgG
- Do not bind complement
- Stimulated by transfusion or pregnancy (not a common cause of HDFN)
- Do not react with enzyme-treated RBCs

Duffy System and Malaria

Most African Americans are Fy(a–b–)

Certain malarial parasites *(Plasmodium knowlesi* and *Plasmodium vivax)* will not invade Fy(a**–**) and Fy(b**–**) negative cells

Fy^a or Fy^b acts as a receptor for the merozoite to attach to the RBC

The Fy(a–b–) phenotype is found frequently in people from west and central Africa, supporting the theory of selective evolution

Kidd Blood Group System (1 of 2)

3 antigens: Jk^a, Jk^b, and Jk3

- In the United States, most blacks (51.1%) are Jk(a+b–)
- In the United States, most Caucasians (50.3%) are Jk(a+b+)

Jk3 is present whenever Jk^a and Jk^b are present

Kidd null phenotypes: Jk(a–b–)

- Usually seen in individuals from the Far East or Pacific Islands (rare)
- May produce anti-Jk3 antibody
- RBCs are resistant to 2M urea

Kidd Blood Group System (2 of 2)

Show dosage

Enhanced by enzymes

Kidd Antibodies

- Anti-Jk^{a} and anti-Jk^{b} antibodies
  - IgG
  - Clinically significant
  - May bind complement
  - Implicated in HTRs and HDFN
  - Common cause of delayed HTRs
  - Usually appear with other antibodies when detected
  - Detection is aided by enzymes, low-ionic-strength solution (LISS), and polyethylene glycol (PEG)

Lutheran Blood Group System

- 19 antigens exist (chromosome 19)
- Weakly expressed on cord blood cells
- Most are high-incidence antigens; antibodies are rare
- Primary antigens include Lu^a and Lu^b
  - 92.4% Lu(a–b+)
  - 7.4% Lu(a+b+)
  - 0.2% Lu(a+b–)
  - Lu_{null} phenotype is rare, inherited recessively
- Not affected by enzymes

Lutheran Antibodies

Anti-Lu^a

- May occur without RBC stimulation
- Immunoglobulin M (IgM) and IgG
- Reacts best at room temperature
- Shows mixed-field pattern
- Not clinically significant

Anti-Lu^b

- Rare due to high incidence of antigen
- IgG
- Reacts best at antihuman globulin (AHG)
- Shows mixed-field pattern
- Associated with transfusion reactions (clinically significant)

Lewis Blood Group System

Lewis antigens are found in secretions (glycoproteins) and plasma (glycolipids)

The glycolipids adsorb onto the RBC membrane

Table 7.10 Lewis System Phenotypes and Frequencies				
Phenotype			Frequency (%)	
Reactions with anti-Le^{a}	Reactions with anti-Le^{b}	Interpretation	Whites	Blacks
+	0	Le(a+b–)	22	23
0	+	Le(a–b+)	72	55
0	0	Le(a–b–)	6	22
+	+	Le(a+b+)	Rare	Rare
From Fung MK: *Technical manual*, ed 18, Bethesda, MD, 2014, AABB.				

Lewis Antigens

Le^a and Le^b are not alleles

Lewis system depends on *Hh*, *Se*, and *Le* genes

le, *h*, and *se do not* produce products

If the *Le* gene is inherited, Le^a substance is produced

Le, *H*, and *Se* genes must *all* be inherited to convert Le^a to Le^b

Le(a+b+) RBCs are rare

Lewis Genes

Table 7.12 Lewis Genes and Red Cell Phenotypes		
Genes present	Antigens in secretions	Red cell phenotype
Le sese H	Le^a	Le(a+b–)
Le Se H	Le^a Le^b H	Le(a–b+)
lele sese H	None	Le(a–b–)
lele Se H	H	Le(a–b–)
Le sese hh	Le^a	Le(a+b–)
Le Se hh	Le^a	Le(a+b–)
lele sese hh	None	Le(a–b–)
lele Se hh	None	Le(a–b–)

Lewis Antibodies

- Lewis antibodies are produced by Le(a–b–)
- IgM
- Not clinically significant
- Agglutination can occur at immediate-spin (IS), 37°C, and AHG
- Enzymes enhance anti-Le^b reactivity
- Anti-Le^a binds complement; may cause hemolysis in vitro
- Neutralization can confirm the presence or eliminate reactions with Lewis antibody

I Blood Group System i Antigen

I and i antigens are not antithetical antigens

They form on the precursor A, B, and H chains of RBCs

Newborns have i antigen

Adults have I antigen

i antigen (linear) converts to I antigen (branched) as a child matures (about 2 years of age)

I Antibodies

- Cold-reacting, IgM, bind complement
- Not clinically significant
- Usually autoantibody (autoanti-I)
- Alloanti-I is rare
- Reactions are avoided by **prewarming**
- Reacts as compound antibody
  - Often found as an anti-IH
  - Stronger agglutination with RBCs having many H sites (O and A_2)

Disease Association

Autoanti-I

Mycoplasma pneumoniae

Cold hemagglutinin disease

Anti-i

Infectious mononucleosis

Lymphoproliferative disease

Cold hemagglutinin disease (occasionally)

P1PK Blood Group System

P1PK blood group system: P1 and P^k

- P1 antigen is detected in plasma and **hydatid cyst fluid**

Globoside blood group system: P antigen

Globoside blood group collection: Luke (LKE) and PX2 antigens

P, P^k, and LKE antigens are high-frequency antigens

P Antigens and Antibodies

Table 7.14 P1PK and GLOB Blood Group Systems Antigen and Antibody Characteristics			
Phenotype	Antigen characteristics	Possible antibodies	Alloantibody Characteristics
P_1	Red cells express P, P1, and P^k antigens P1 antigen is not well developed at birth Most common phenotype	None	Not applicable
P_2	Lacks P1 antigen but expresses P and P^k antigens	Anti-P1	IgM; room temperature; not clinically significant
	Second most common phenotype		Variable reactions with adult cells
P_1^k	Red cells express P1 and P^k antigens	Anti-P	Clinically significant; associated with spontaneous abortions (rare)
	Very rare phenotype		
P_2^k	Red cells express only P^k antigens Very rare phenotype	Anti-P and anti-P1	Anti-P and anti-P1 characteristics
p	Null phenotype of system Negative for P, P1, and P^k antigens Very rare phenotype	Anti-$PP1P^k$ (Tj^a)	Hemolytic; clinically significant; can be separated into three specificities

P Antibodies

Anti-P1

- Found in P_2 individuals
- IgM; enhanced by enzymes
- Non-RBC stimulated
- Can be neutralized by P1 substance

Autoanti-P

- Associated with **cold paroxysmal hemoglobinuria**
- IgG (**Donath-Landsteiner antibody**); a biphasic hemolysin that binds with P_1 or P_2 cells at low temperatures before the complement is activated
- May appear in children after viral infection

Anti-PP1P^k

- Occurs in individuals with the null phenotype
- Causes hemolysis in vitro
- Clinically significant

MNS Blood Group System

M and N

Coded by glycophorin A

- Membrane structure is called sialoglycophorin A
- Consists of 131 amino acids

M and N differ at positions 1 and 5 on glycophorin A (GPA)

Show dosage

- Homozygous inheritance enhances agglutination [(M+N–) or (M–N+)]

S, s, and U

Coded by glycophorin B

- Membrane structure is called sialoglycophorin B
- Consists of 72 amino acids

S and s differ at position 29

- S has methionine; s has threonine

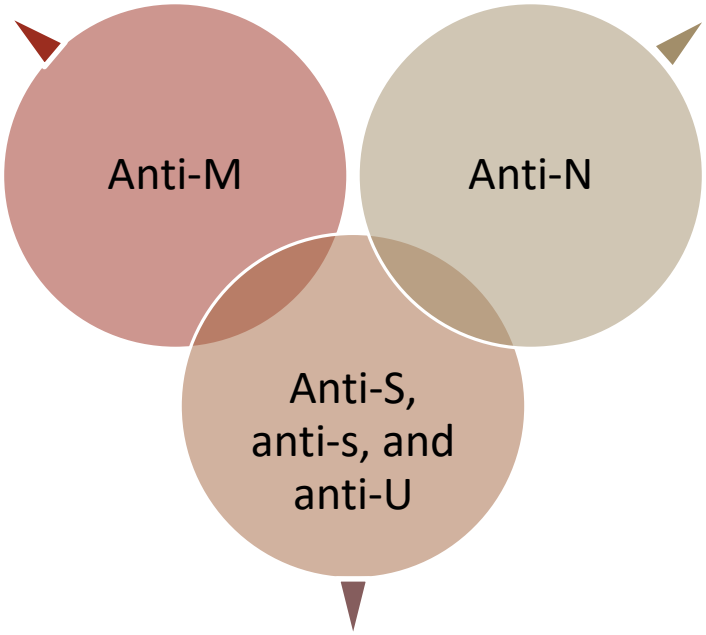
U antigen

- Located near membrane
- Present when S or s is inherited

Absence of glycophorin B (GPB) would result in S–s–U–

MNS Antibodies

- IgM and IgG
- Rarely encountered in HDFN
- Variable reactions depend on reagent pH



- Rare IgM
- N-like antibodies found in dialysis patients from formaldehyde-sterilized instruments

- Clinically significant IgG
- Anti-U is rare but can be found in S–s– persons (black population)

MNS System Frequencies

Table 7.16 Phenotype Frequencies in MNS System		
Antigen	Phenotype frequencies (%)	
	Whites	Blacks
M+	78	74
N+	72	75
S+	55	31
s+	89	93
U+	99.9	99
From Reid ME, Lomas-Francis C, Olsson ML: The blood group antigen facts book, ed 3, San Diego, CA, 2012, Academic Press.		

Miscellaneous Systems

Table 7.17 Miscellaneous Blood Groups and Antigens				
Name	Antigen Symbol	ISBT NO.	Antigens	Characteristics
Diego	Di. Wr	010	Di^{a} **Di^{b}** Wr^{a} **Wr^{b}**	Di^{a} is more common in South American Indians anti-Wr^{a} is commonly found with other antibodies
Cartwright	Yt	011	**Yt^{a}** Yt^{b}	Variably sensitive to enzymes; sensitive to DTT
Xg	Xg	012	Xg^{a}	Inherited on X chromosome; frequency varies with sex
Scianna	SC	013	**SC:1** SC:2 **SC:3**	
Dombrock	Do	014	Do^{a} Do^{b} **Gy^{a} Hy Jo^{a}**	Hy phenotype is found only in blacks; anti-Do^{a} and anti-Do^{b} antibodies are rarely found as a single specificity
Colton	Co	015	Co^{a} Co^{b} Co3	Anti-Co^{b} is rarely found as a single specificity
Chido/Rodgers	Ch/Rg	017	**Ch Rg**	Antigens are sensitive to enzymes and found in plasma; antibodies have HTLA characteristics
Gerbich	Ge	020	**Ge2 Ge3 Ge4** Wb Ls^{a} An^{a} Dah	All antigens except for Ge4 are sensitive to enzymes
Cromer	Cr	021	**Cr^{a} Tc^{a}** Tc^{b} Tc^{c} **Dr^{a} Es^{a} IFC WES^{a}** WES^{b} **UMC**	Antigen is also found in plasma; located on decay-accelerating factor
Knops	Kn	022	**Kn^{a}** Kn^{b} **McC^{a} Sl^{a} Yk^{a}**	Antigen depression in SLE, PNH, and AIDS; antigens are weakened by ficin treatment; antibodies have HTLA characteristics
Cost	Cs	205	**Cs^{a}** Cs^{b}	Part of a blood group collection rather than a system
VEL	Vel	034	**Vel**	Variable antigen expression on red cells both IgG and IgM antibodies are associated with hemolytic reactions; antibodies react best with enzyme-treated red cells
JMH	JMH	026	**JMH**	Autoanti-JMH is often found in elderly patients with absent or weak antigen expression; antibodies have HTLA characteristics; antigens are sensitive to enzymes and DTT
Sd^{a}	Sd^{a}	901.012	**Sd^{a}**	Antigen found in guinea pig and human urine; antibodies are typically weak and agglutination is mixed field; reduction of Sd^{a} expression during pregnancy
Note: Items in boldface indicate antigens of high incidence. AIDS, Acquired immunodeficiency syndrome; DTT, dithiothreitol; HTLA, high-titer, low-avidity; PNH, paroxysmal nocturnal hemoglobinuria; SLE, systemic lupus erythematosus.				

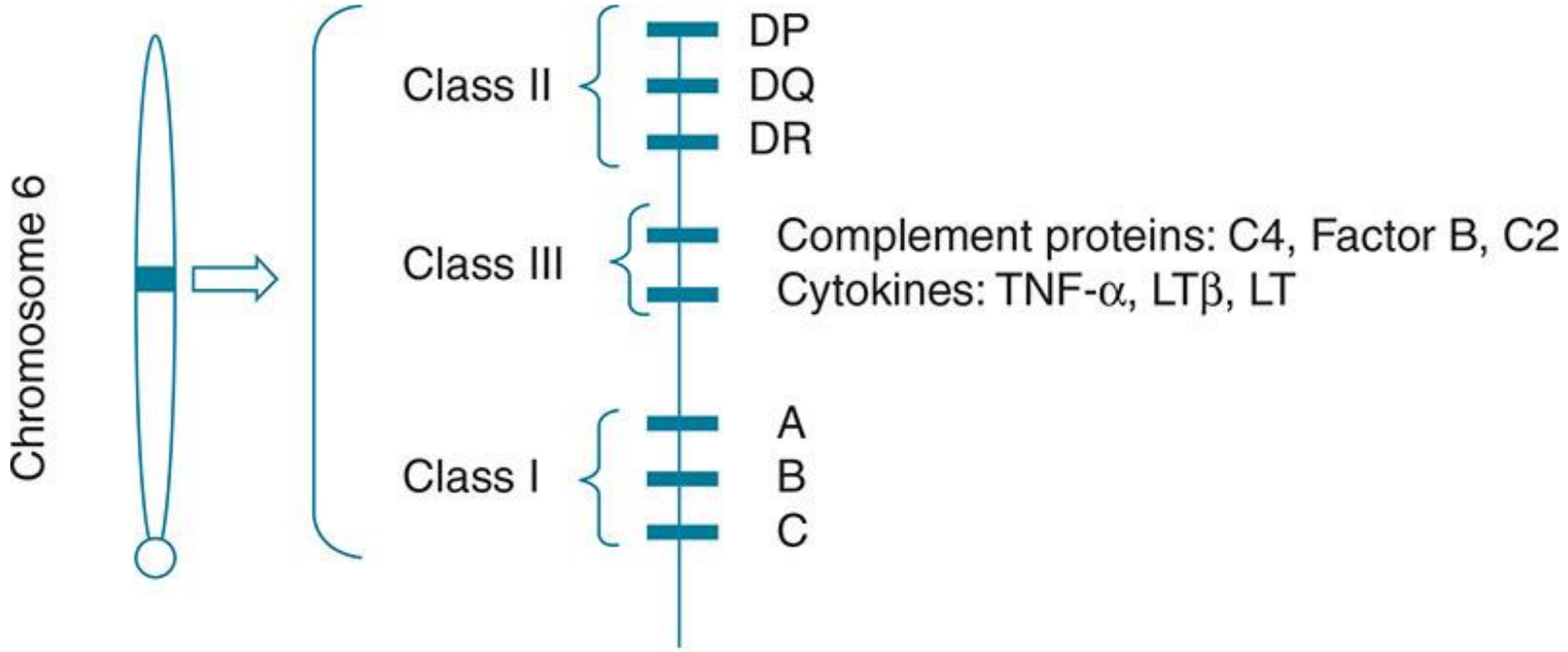
HLAs

- HLAs are found on leukocytes and tissue cells
- HLA antibodies are produced as a result of transfusion and/or pregnancy
- Antibodies have been associated with **refractoriness** and transfusion reactions
- HLA testing is used to assess risk factors for disease susceptibility
- Matching for organ and HPC transplants

Inheritance of HLAs

- Genes that code for HLA are part of the MHC
- MHC genes are divided into 3 classes
  - Class I: platelets, leukocytes, nucleated cells
  - Class II: macrophages, dendritic cells, B cells
  - Class III: code for complement and **cytokines**
- Individuals inherit one **haplotype** (closely linked genes) from each parent
- Antigens are named with a letter followed by a number (e.g., A2)

MHCs



Class I and II HLA Inheritance: Example

MOTHER	
A2	A11
B7	B44
Cw7	Cw12
DR17	DR13
DQ2	DQ8

FATHER	
A1	A3
B8	B35
Cw3	Cw5
DR4	DR8
DQ5	DQ7

Potential offspring:

Child 1	
A2	A1
B7	B8
Cw7	Cw3
DR17	DR4
DQ2	DQ5

Child 2	
A2	A3
B7	B35
Cw7	Cw5
DR17	DR8
DQ2	DQ7

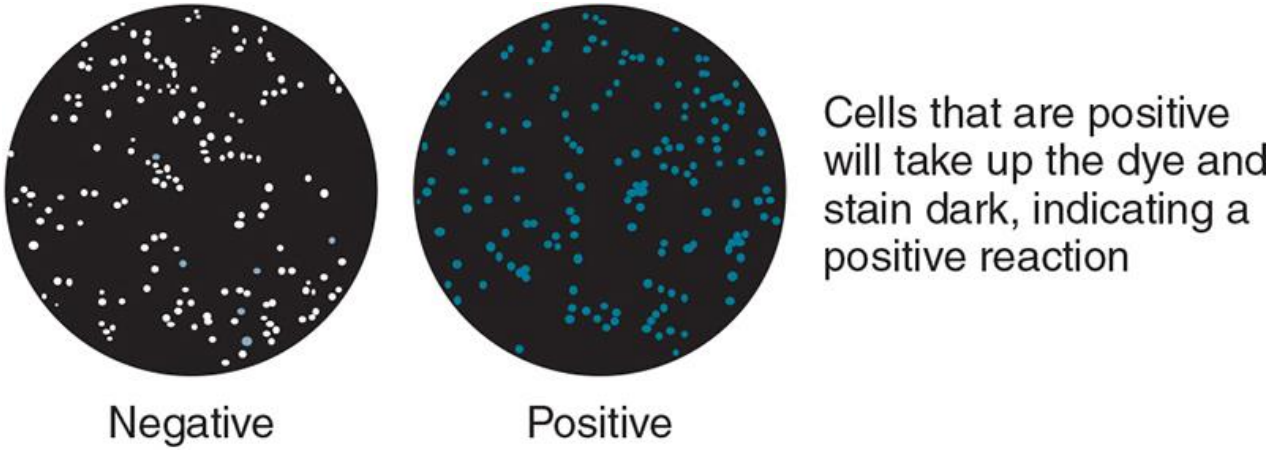
Child 3	
A11	A1
B44	B8
Cw12	Cw3
DR13	DR4
DQ8	DQ5

Child 4	
A11	A3
B44	B35
Cw12	Cw5
DR13	DR8
DQ8	DQ7

HLA Testing

Serologic identification requires the lymphocytotoxicity test method

- Complement and dye are used to determine whether there is antigen–antibody recognition



Antibody Detection and Identification

Matching HLAs in patients with existing antibodies is important for graft survival

Patients may become sensitized to HLAs by the following exposures:

- Pregnancy
- Blood transfusions
- Previous transplant

HPC Transplants

- HPCs can be obtained from bone marrow, peripheral blood, and cord blood
- HPCs can be used to treat diseases such as aplastic anemia, leukemia, lymphoma, and Hodgkin's disease
- HLA matching at the allelic level is important to avoid rejection and GVH disease

Platelet Antigens

- Platelet proteins can elicit immune responses
- Antibodies to platelets may cause
  - *Neonatal alloimmune thrombocytopenia (NAIT)*: destruction of newborn platelets by maternal antibody
  - *Posttransfusion purpura (PTP)*: destruction of platelets after transfusion
- The most common platelet antibody is directed against HPA-1a (or $P1^{A1}$)