HEMOLYTIC ANEMIAS

HEMOLYTIC ANEMIA

• Anemia of increased destruction
  - Normochromic, normochromic anemia
  - Shortened RBC survival
  - Reticulocytosis - Response to increased RBC destruction
  - Increased indirect bilirubin
  - Increased LDH

HEMOLYTIC ANEMIA

Causes

• INTRACORPUSCULAR HEMOLYSIS
  - Membrane Abnormalities
  - Metabolic Abnormalities
  - Hemoglobinopathies
• EXTRACORPUSCULAR HEMOLYSIS
  - Nonimmune
  - Immune

HEMOLYTIC ANEMIA

Testing

• Absent haptoglobin
• Hemoglobinuria
• Hemoglobinemia

HEMOLYTIC ANEMIA

Membrane Defects

• Microskeletal defects
  - Hereditary spherocytosis
• Membrane permeability defects
  - Hereditary stomatocytosis
• Increased sensitivity to complement
  - Paroxysmal nocturnal hemoglobinuria
**RED CELL CYTOSKELETON**

**HEREDITARY SPHEROCYTOSIS**
- Defective or absent spectrin molecule
- Leads to loss of RBC membrane, leading to spherocytosis
- Decreased deformability of cell
- Increased osmotic fragility
- Extravascular hemolysis in spleen

**SPLENIC ARCHITECTURE**

**HEREDITARY SPHEROCYTOSIS**

*Osmotic Fragility*

<table>
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<tr>
<th>NaCl (% of normal saline)</th>
<th>% Hemolysis</th>
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<tr>
<td>0.3</td>
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<tr>
<td>0.5</td>
<td>60</td>
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**Paroxysmal Nocturnal Hemoglobinuria**
- Clonal cell disorder
- Ongoing Intra- & Extravascular hemolysis; classically at night
- Testing
  - Acid hemolysis (Ham test)
  - Sucrose hemolysis
  - CD-59 negative (Product of PIG-A gene)
- Acquired deficit of GPI-Associated proteins (including Decay Activating Factor)
GPI BRIDGE

- GPI links a series of proteins to outer leaf of cell membrane via phosphatidylinositol bridge, with membrane anchor via diacylglycerol bridge
- PIG-A gene, on X-chromosome, codes for synthesis of this bridge; multiple defects known to cause lack of this bridge
- Absence of decay accelerating factor leads to failure to inactivate complement & thereby to increased cell lysis

G6PD DEFICIENCY - Function of G6PD

- Regenerates NADPH, allowing regeneration of glutathione
- Protects against oxidative stress
- Lack of G6PD leads to hemolysis during oxidative stress
  - Infection
  - Medications
  - Fava beans
- Oxidative stress leads to Heinz body formation, extravascular hemolysis
Hemolytic Anemia

EXTRACORPUSCULAR HEMOLYSIS

Nonimmune

- Mechanical
- Infectious
- Chemical
- Thermal
- Osmotic

Microangiopathic Hemolytic Anemia

Causes

- Vascular abnormalities
  - Thrombotic thrombocytopenic purpura
  - Renal lesions
    - Malignant hypertension
    - Glomerulonephritis
    - Preeclampsia
    - Transplant rejection
  - Vasculitis
    - Polyarteritis nodosa
    - Rocky mountain spotted fever
    - Wegener's granulomatosis
    - Scleroderma renal crisis

Microangiopathic Hemolytic Anemia

Causes - #2

- Vascular abnormalities
  - AV Fistula
  - Cavernous hemangioma
- Intravascular coagulation predominant
  - Abruptio placentae
  - Disseminated intravascular coagulation

HEMOLYTIC ANEMIA

Causes

- INTRACORPUSCULAR HEMOLYSIS
  - Membrane Abnormalities
  - Metabolic Abnormalities
  - Hemoglobinopathies
- EXTRACORPUSCULAR HEMOLYSIS
  - Nonimmune
  - Immune

Glucose 6-Phosphate Dehydrogenase

Different Isozymes

Level needed for protection vs ordinary oxidative stress

- Normal (GdB)
- Black Variant (GdA-)
- Mediterranean (Gd Med)
IMMUNE HEMOLYTIC ANEMIA

General Principles
- All require antigen-antibody reactions
- Types of reactions dependent on:
  - Class of Antibody
  - Number & Spacing of antigenic sites on cell
  - Availability of complement
  - Environmental Temperature
  - Functional status of reticuloendothelial system
- Manifestations
  - Intravascular hemolysis
  - Extravascular hemolysis

IMMUNE HEMOLYTIC ANEMIA

Coombs Test - Direct
- Looks for immunoglobulin &/or complement of surface of red blood cell (normally neither found on RBC surface)
- Coombs reagent - combination of anti-human immunoglobulin & anti-human complement
- Mixed with patient's red cells; if immunoglobulin or complement are on surface, Coombs reagent will link cells together and cause agglutination of RBCs

IMMUNE HEMOLYTIC ANEMIA

Coombs Test - Indirect
- Looks for anti-red blood cell antibodies in the patient's serum, using a panel of red cells with known surface antigens
- Combine patient's serum with cells from a panel of RBC's with known antigens
- Add Coombs' reagent to this mixture
- If anti-RBC antigens are in serum, agglutination occurs

IMMUNE HEMOLYTIC ANEMIA

General Principles - 2
- Antibodies combine with RBC, & either
  1. Activate complement cascade, &/or
  2. Opsonize RBC for immune system
- If 1, if all of complement cascade is fixed to red cell, intravascular cell lysis occurs
- If 2, &/or if complement is only partially fixed, macrophages recognize Fc receptor of Ig &/or C3b of complement & phagocytize RBC, causing extravascular RBC destruction

HEMOLYTIC ANEMIA - IMMUNE
- Drug-Related Hemolysis
- Alloimmune Hemolysis
  - Hemolytic Transfusion Reaction
  - Hemolytic Disease of the Newborn
- Autoimmune Hemolysis
  - Warm autoimmune hemolysis
  - Cold autoimmune hemolysis
IMMUNE HEMOLYSIS
Drug-Related

- Immune Complex Mechanism
  - Quinidine, Quinine, Isoniazid
- “Haptenic” Immune Mechanism
  - Penicillins, Cephalosporins
- True Autoimmune Mechanism
  - Methyldopa, L-DOPA, Procaineamide, Ibuprofen

DRUG-INDUCED HEMOLYSIS
True Autoantibody Formation

- Certain drugs appear to cause antibodies that react with antigens normally found on RBC surface, and do so even in the absence of the drug

DRUG-INDUCED HEMOLYSIS
Immune Complex Mechanism

- Drug & antibody bind in the plasma
- Immune complexes either
  - Activate complement in the plasma, or
  - Sit on red blood cell
- Antigen-antibody complex recognized by RE system
- Red cells lysed as “innocent bystander” of destruction of immune complex
- REQUIRES DRUG IN SYSTEM

ALLOIMUNE HEMOLYSIS
Hemolytic Transfusion Reaction

- Caused by recognition of foreign antigens on transfused blood cells
- Several types
  - Immediate Intravascular Hemolysis (Minutes) - Due to preformed antibodies; life-threatening
  - Slow extravascular hemolysis (Days) - Usually due to repeat exposure to a foreign antigen to which there was a previous exposure; usually only mild symptoms
  - Delayed sensitization - (Weeks) - Usually due to 1st exposure to foreign antigen; asymptomatic

DRUG-INDUCED HEMOLYSIS - Haptenic Mechanism

- Drug binds to & reacts with red cell surface proteins
- Antibodies recognize altered protein, ± drug, as foreign
- Antibodies bind to altered protein & initiate process leading to hemolysis
ALLOIMMUNE HEMOLYSIS

Testing Pre-transfusion

- ABO & Rh Type of both donor & recipient
- Antibody Screen of Donor & Recipient, including indirect Coombs
- Major cross-match by same procedure (recipient serum & donor red cells)

ALLOIMMUNE HEMOLYSIS

Hemolytic Disease of the Newborn - #2

- Can cause severe anemia in fetus, with erythroblastosis and heart failure
- Hyperbilirubinemia can lead to severe brain damage (kernicterus) if not promptly treated
- HDN due to Rh incompatibility can be almost totally prevented by administration of anti-Rh D to Rh negative mothers after each pregnancy

AUTOIMMUNE HEMOLYSIS

Warm Type

- Usually IgG antibodies
- Fix complement only to level of C3, if at all
- Immunoglobulin binding occurs at all temps
- Fc receptors/C3b recognized by macrophages; therefore,
  - Hemolysis primarily extravascular
  - 70% associated with other illnesses
  - Responsive to steroids/splenectomy

ALLOIMMUNE HEMOLYSIS

Hemolytic Disease of the Newborn

- Due to incompatibility between mother negative for an antigen & fetus/father positive for that antigen. Rh incompatibility, ABO incompatibility most common causes
- Usually occurs with 2nd or later pregnancies
- Requires maternal IgG antibodies vs. RBC antigens in fetus
AUTOIMMUNE HEMOLYSIS

Cold Type

- Most commonly IgM mediated
- Antibodies bind best at 30º or lower
- Fix entire complement cascade
- Leads to formation of membrane attack complex, which leads to RBC lysis in vasculature
- Typically only complement found on cells
- 90% associated with other illnesses
- Poorly responsive to steroids, splenectomy; responsive to plasmapheresis

HEMOLYTIC ANEMIA

Summary

- Myriad causes of increased RBC destruction
- Marrow function usually normal
- Often requires extra folic acid to maintain hematopoiesis
- Anything that turns off the bone marrow can result in acute, life-threatening anemia