Transfusion Medicine
Part 1

Marisa Saint Martin, MD
Assistant Professor, Anatomic and Clinical Pathology
Medical Director Pathology and Laboratory Services Gottlieb Memorial Hospital
Associate Director Blood Bank and Transfusion Services

Objectives

• Describe how blood groups are determined and their clinical significance
• Summarize key steps of pre-transfusion testing
• Compare Direct Antiglobulin Test (DAT) to Indirect Antiglobulin Test (IAT)
• Compare “type and screen” to “type and cross match” ordering
• Summarize key differences between Isoagglutinins, Alloantibodies, and Autoantibodies
• Summarize why blood is screened for unexpected clinically significant antibodies

Blood Donors and Medicine

• 15 million donations collected in the US annually
• 9.2 million donors
• 38% of US population eligible to donate
• Less than 6% of eligible population do so
• Next American Red Cross blood drive will be held on 11/21/18 from 9am-3pm in the Stritch gym
Blood Types
Do you know yours?

<table>
<thead>
<tr>
<th>Type</th>
<th>This Class</th>
<th>US Population</th>
</tr>
</thead>
<tbody>
<tr>
<td>A+</td>
<td>30%</td>
<td></td>
</tr>
<tr>
<td>A–</td>
<td>6%</td>
<td></td>
</tr>
<tr>
<td>B+</td>
<td>9%</td>
<td></td>
</tr>
<tr>
<td>B–</td>
<td>2%</td>
<td></td>
</tr>
<tr>
<td>AB+</td>
<td>4%</td>
<td></td>
</tr>
<tr>
<td>AB–</td>
<td>1%</td>
<td></td>
</tr>
<tr>
<td>O+</td>
<td>33%</td>
<td></td>
</tr>
<tr>
<td>O–</td>
<td>9%</td>
<td></td>
</tr>
</tbody>
</table>

Blood Types
What does it mean?

<table>
<thead>
<tr>
<th>ABO Type</th>
<th>Expected (pre-formed)</th>
<th>Antigens in Plasma</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>Anti-B</td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>Anti-A</td>
<td></td>
</tr>
<tr>
<td>O</td>
<td>Anti-A, anti-B</td>
<td>&amp; anti-A,B</td>
</tr>
<tr>
<td>AB</td>
<td>Neither anti-A or anti-B</td>
<td></td>
</tr>
</tbody>
</table>
Why?

- Genetics – Over 600 antigens in RBC’s
- Response to Genetics
  - Isoagglutinins
  - Alloantibodies
  - Autoantibodies

RBC Isoagglutinins

- Antibody against dominant polysaccharide
- Naturally occurring: made against bacteria which share polysaccharide epitopes
- IgM
- Activate complement
- Intravascular hemolysis (fast)

ABO Group System

Alloantibodies

- Antibodies against antigens we lack
- Requires exposure to antigen on RBC’s
  - Pregnancy
  - Transfusion
  - Transplantation
- IgG
- Most do not bind Complement
- Extravascular hemolysis (slow)

Autoantibodies

- Antibodies against generic (non-specific) RBC membrane antigens
- Common – increase with each decade of life
- Usually “silent”
- Can be detected in plasma of normal individuals
- Some can cause hemolysis
Blood Transfusion

• Blood
  – Biological product
  – Pharmaceutical product
• FDA regulated
• “Liquid transplant”
• Requires a doctor’s order

Transfusion Therapy

Pre-Transfusion Testing

• Patient identification
• Specimen identification
• Type and Screen
• Type and Crossmatch
Pre-Transfusion Testing

Blood Type
• ABO and Rh typing
  -- Forward: Typing of patient’s RBC's with:
    • Anti A
    • Anti B
    • Anti D
  -- Reverse: Typing of patient’s isoagglutinins. Patient serum with:
    • A cells
    • B cells

Antibody Screen
• Patient’s serum with set of three reagent RBC’s with known antigens
• RBC’s have about 600 serologically defined surface antigens
• 33 blood group systems
• D antigen of the Rh blood group is the most immunogenic after A and B
• Other Rh antigens: C, c, E, e
• Kell: K, k
• Duffy: Fy^a, Fy^b
• Kidd: Jk^a, Jk^b
• MNS: M, N, S, s
• Lutheran: Lu^a, Lu^b
• Others

Unexpected Alloantibodies
• Found in patients who lack the corresponding antigen
• Stimulated by exposure to another person’s RBC’s
• Most IgG
• Most do not bind complement
• Extravascular hemolysis (slow), destruction of RBC’s in 10-14 days

Antibody Screen/Indirect Coombs Test

1. Patient’s serum with Ab
2. Testing O RBC’s with known Ag
3. Agglutination
Agglutination

Direct Coombs Test/DAT

Antiglobulin

Patient's RBC's coated with Ab

Agglutination
Direct Coombs Test/ DAT

- Delayed hemolytic transfusion reactions
- Autoimmune hemolytic anemia
- Hemolytic diseases of the newborn
- Acute hemolytic transfusion reactions

Pre-Transfusion Compatibility Testing

- Type: ABO and Rh
- Screen: Tests for unexpected antibodies
- Crossmatch: Donor and recipient compatibility
  - ID and allocates units for the patient
  - Major X-match: recipient plasma with donor cells
  - Minor X-match: donor plasma with recipient cells
  - Only order it when transfusion is required
  - 72 hour hold for the unit
- History check

Type and Screen

- Focuses only on your patient's blood
- Typing determines the patient's blood type
- Screening screens the patients for pre-formed or unexpected antibodies in serum
- Type and screen is the first thing to order

Type and Crossmatch

- Crossmatch involves mixing a small amount of the patient's blood and a small amount of blood taken out of the donor blood
- Crossmatching is physically mixing two people's bloods in a container to see if agglutination happens
- If agglutination happens, it tells us that there are some unknown antibody in the patient's serum that is reacting badly with the donor's red cells (major)
- Or donor plasma agglutinating patient's red blood cell (minor)
Summary

- How blood types are determined and their importance
- Pre-transfusion testing
- DAT vs IAT
- Differences between isoagglutinins, alloantibodies, and autoantibodies
- Why we screen for unexpected antibodies
- Compared TAS to TXM

Questions?

Transfusion Medicine
Part 2

Marisa Saint Martin, MD
Assistant Professor, Anatomic and Clinical Pathology
Medical Director Pathology and Laboratory Services Gottlieb Memorial Hospital
Associate Director Blood Bank and Transfusion Services
Associate Director Pathology Residency Program
Objectives

- Classify Transfusion Reactions
- Outline steps to follow when a transfusion reaction occurs
- Explain the pathophysiology of Hemolytic Disease of the fetus and the Newborn (HDN)
- Summarize options for antenatal management and prophylaxis of HDN
- Define clinical characteristics of AutoImmune Hemolytic Anemias (AIHA)

Transfusion Reactions

- ANY unfavorable consequence is considered an adverse effect of blood transfusion. It is also referred to as a TRANSFUSION REACTION
- Risks vs. Benefits

- Acute (<24 hours)
  - Immunologic: Hemolytic; Febrile-non hemolytic; Allergic; Anaphylactic; TRALI (Transfusion Related Acute Lung Injury)
  - Acute (<24 hours)
    - Non Immunologic: Circulatory Overload; Hemolytic (physical or chemical destruction of RBCs); Air Embolus; Hypocalcemia; Hypothermia
Transfusion Reactions

- Delayed (>24 hours)
  -- Immunologic: Hemolytic (anamnestic response); Graft vs. Host Disease; Post-Transfusion Purpura
- Delayed (>24 hours)
  -- Non Immunologic: Iron Overload
- Infectious Complications of Blood Transfusion
- Other Consequences of Blood Transfusion (immunomodulation, refractoriness)

Acute Transfusion Reactions

Immunologic

Acute Hemolytic Transfusion Reaction

- Incidence: 1 in 76,000
- Fatal: 1 in 1.8 million
- Cause: Red Cell Incompatibility Intravascular hemolysis that starts after pre-formed antibodies activate complement to completion in the vasculature

Acute Transfusion Reactions

Immunologic

Acute Hemolytic Transfusion Reaction

- Signs and Symptoms:
- Onset within minutes to hours
- Release of cytokines leading to fever, hypotension, shock
- Pain along infusion site and patient anxiety
- Coagulopathy that may progress to DIC (disseminated intravascular coagulopathy)
- Renal failure with oliguria and anuria due to Hgb and RBC stroma
- Free Hemoglobin in serum and urine
- Prevention: Give ABO Compatible Blood
Acute Transfusion Reactions
  Immunologic
Febrile Non-Hemolytic Transfusion Reactions
  • Incidence: 1:17 to 1:200 RBCs and 1:3 to 1:100 platelets. Down to about 1-2% after universal leukoreduction, (from ~30%)
  • Cause: Increase in temperature of 1°C (2°F) due to recipient antibodies against donor leukocyte antigens, and/or accumulated cytokines
  • Signs and Symptoms: Usually mild, fever, chills, rigors, headaches, vomiting

Leukoreduction of Blood Products
  • Leukocyte-reduced RBCs
    — At least 85% of original RBCs
    — < 5 x 10^6 WBCs in 95% of units tested
  • Leukocyte-reduced Platelet Concentrates:
    — At least 5.5 x 10^10 platelets in 75% of units tested
    — < 8.3 x 10^6 WBCs in 95% of units tested
  • Leukocyte-reduced Apheresis Platelets:
    — At least 3.0 x 10^11 platelets in 90% of units tested
    — < 5.0 x 10^6 WBCs in 95% of units tested

Acute Transfusion Reactions
  Immunologic
Febrile Non-Hemolytic Transfusion Reactions
  • Seen in multiply transfused patients, multiple pregnancies
  • Must rule out:
    — Hemolytic transfusion reaction
    — Bacterial contamination
  • Prevention: Leukoreduction of cellular blood products (antipyretics?)
Acute Transfusion Reactions
Immunologic

Allergic (Urticarial) Transfusion Reaction

- Incidence: 1:33 to 1:100
- Cause: Recipient antibodies against donor plasma proteins or other allergens (food, medications)
- Signs and Symptoms: Begins within minutes of infusion, release of histamine leading to rash, itching and hives
- Must be sure that the only reaction is the urticaria
- Prevention: Pre-treatment with anti-histamines

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Acute Transfusion Reactions
Immunologic

Allergic (Urticarial) Transfusion Reaction

- May re-start the unit slowly after treatment, if symptoms resolve

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Acute Transfusion Reactions
Immunologic

Anaphylactic Transfusion Reaction

- Incidence: 1:20,000 to 1:50,000
- Cause: Recipient is IgA deficient and has anti-IgA in serum that reacts to even small amounts of donor IgA in the plasma of any blood component
- Signs and Symptoms:
  - Sudden onset of symptoms. Life threatening!!!
  - Hypotension leading to shock, loss of consciousness and death
  - Difficulty breathing and bronchospasms leading to cyanosis
  - N/V, severe abdominal cramps, diarrhea
- Prevention: STOP transfusion immediately
- Wash cellular blood products or give IgA deficient products

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Acute Transfusion Reactions
Immunologic
Transfusion Related Acute Lung Injury (TRALI)
• Acute onset of respiratory distress
• During or within 6 hours of transfusion
• Clinical Features
• Hypoxemia
• Dyspnea
• Non-cardiogenic Pulmonary edema
• Hypotension
• Fever
• Bilateral lung infiltrates on CXR

Acute Transfusion Reactions
Immunologic
Transfusion Related Acute Lung Injury (TRALI)
• Incidence: 1:1,200 to 1:190,000
• Can contribute to mortality in 1 in 200,000 transfusion recipients (leading cause)

• Cause: Donor HLA antibodies against recipient WBCs: antibodies bind to antigen in PMN causing aggregation and adhesion to capillary endothelium in the lungs, causing complement activation and resulting in edema

Rafal Fudala, Agnieszka Krupa, Dorota Stankowska, Timothy C. Allen, Anna K. Kurdowska, Clinical Science Apr 01, 2010, 118(8)519–526; DOI: 10.1042/CS20090422
Acute Transfusion Reactions

Immunologic

Transfusion Related Acute Lung Injury (TRALI)

- 15-20% of female donors have HLA antibodies
  - 1 pregnancy 10% risk of HLA alloimmunization, 2 pregnancies 20%, etc.
- 75% fatalities linked to female donors with Ab
- <1% of male donors have HLA antibodies
- There is a 5-7 fold greater TRALI occurrence with high volume plasma products:
  - FFP/plasma frozen>Platelets>Red cells


Acute Transfusion Reactions

Non-Immunologic

Circulatory Overload (TACO)

- Incidence: ~1%. Elderly and pediatric patients at risk
- Cause: Rapid increase in blood volume to a patient with compromised cardiac or pulmonary function
- Signs and Symptoms:
  - Dyspnea, cyanosis, severe headaches, hypertension, congestive heart failure. Must r/o TRALI
- Prevention:
  - Stop infusion and place patient in a sitting position
  - Slow down future infusions

Acute Transfusion Reactions

Non-Immunologic

- Physically or Chemically induced hemolysis
- Hypothermia
- Hypocalcemia
- Air embolism
Infectious Complications of Blood Transfusion

**Bacterial Contamination**
- Most often seen from platelets transfusions (room temperature)
- Red cell units will look dark
- Symptoms:
  - Rapid onset
  - Fever, shaking chills, hypotension, muscle pain
  - NV, abdominal cramps, diarrhea, hemoglobinuria, shock, renal failure and DIC

Infectious Complications of Blood Transfusion

**Bacterial Contamination**
- Stop transfusion immediately
- Gram stain and blood cultures on unit, patient and infusion set samples
- IV broad spectrum antibiotics
- Prevention: Maintain standards of donor selection, blood collection and maintenance of collected blood components

Estimated Residual Risk After Currently Mandated Testing

<table>
<thead>
<tr>
<th></th>
<th>HIV</th>
<th>HTLV*</th>
<th>Hepatitis B*</th>
<th>Hepatitis C*</th>
<th>Bacterial contamination</th>
<th>WNV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Risk</td>
<td>1:2,000,000</td>
<td>1:2,900,000</td>
<td>1:1,000,000</td>
<td>1:1,150,000</td>
<td>1:2,000-1,400,000 (and &lt;1:10,000 culture)</td>
<td>11 cases reported since 2002</td>
</tr>
</tbody>
</table>

*Modified from AABB Technical Manual, 2018*
Transfusion Reaction Work up

• What to do FIRST if an Acute Transfusion reaction is suspected

• STOP THE TRANSFUSION!!!!!!!

• Keep the line open with saline solution

• Notify the Blood Bank and Physician immediately

Transfusion Reaction Work up

• Post-transfusion blood samples to be collected:
  – Clotted specimen: Repeat ABO, Rh, IAT and Crossmatch
  – Visual check for hemolysis comparing with pre-transfusion sample
  – EDTA specimen: Direct antiglobulin test
  – Bilirubin levels, LDH

Transfusion Reaction Work up

• Recipient diagnosis
• Prior history of pregnancy or transfusions
• Current medications
• Signs and symptoms during transfusion reaction
• How many mls of blood product were infused
• Was a blood warmer used? Infusion pump?
• Was the component manipulated in any way?
• Other solutions/medications given concomitantly?
• Pre and Post transfusion vital signs?
Suspected Transfusion Reaction

- Verify all patient identification and unit identification
- Misidentified patients is one of the leading causes of transfusion related deaths in the United States
- If misidentification is found, search the floor/unit for other patients receiving blood and verify their identification and unit
- Even today 1:10,000 units areTxed to the wrong Pt

Suspected Transfusion Reaction

- Always double check the patient identifiers with the unit tags before spiking the bag
- Recognize the symptoms
- Observation within 15 minutes of start is critical for detection of an acute reaction
- **STOP the transfusion when symptoms occur!**

Mindfulness Exercise

![Mindfulness Exercise Image]

**HDN Pre-requisites**
- Mother lacks the antigen and is exposed by pregnancy/transfusions
- Fetus possesses the antigen and it’s well developed in utero (most RBC antigens are, except for ABO, I, P, Lewis, Cartwright: ABO NOT FULLY EXPRESSED)
- Mother produces the IgG antibody

**HDN Pathogenesis**

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[Link to the full text is not provided in the image.]
HDN
Pathogenesis
• Fetomaternal hemorrhage (FMH) always occur: as early as 8 weeks-most at delivery
• Immunization risk increases with FMH volume
• Trauma, procedures, abortion
• Always avoid using father’s blood when the mom needs a transfusion

HDN
Rh System: D most immunogenic (severe HDN)
• Other blood groups: >K, >Fya, >S
• ABO: Group O mom with an A fetus (mild if any HDN)
• ABO HDN: 15% of pregnancies. Common!

HDN
Clinical Findings
• Hemolysis
  — Anemia  “anemia neonatorum”
  — Jaundice  “icterus gravis/icterus praecox”
• Accelerated red cell destruction-Compensation
  — Accelerated RBC production with increased extramedullary erythropoiesis and circulating nucleated RBCs “erythroblastosis fetalis”
  — Hepatomegaly
• Edema-Decompensation
  — Generalized edema/anasarca “hydrops fetalis”
  — CHF, hemorrhage, enlarged placenta
• Severe anemia: Fetal death
HDN Clinical Findings

• Bilirubinemia
  – Fetus in utero: Mom’s liver conjugates the bilirubin
  – Newborn: Liver is immature. Build up leads to kernicterus

Pre-Natal Testing

• ABO
• Rh
• Antibody screen: If + we do serial titration of the antibody throughout the pregnancy
• History

HDN Assessing Risk to Fetus

• Antibody titer comparison
• Amniocentesis: good indicator of intrauterine hemorrhage
• Liley’s Graph: checks the optical density in the amniotic fluid to correlate with levels of bilirubin
• PUBS, (percutaneous umbilical blood sampling) or cordocentesis: direct measure of fetal hemoglobin
HDN Intrauterine Transfusion

- To correct fetal anemia
- 24-26 weeks of gestation
- Done periodically (every 2 weeks) until delivery

HDN Prevention
Rh Immune Globulin (RhIg)

- What is it?
  - Concentrate of IgG anti D
  - Developed from pools of human plasma
  - Trade names: RhoGam, Rhophylac

- How does it work?
  - Antigen blocking-antibody mediated immunosuppression?

HDN Prevention
RhIg

- Dosage: 1ml = 300 ug
- Sufficient to prevent immunization from 15ml of RBCs or 30 ml of whole blood
- IM or IV
- At 28 weeks
- Within 72 hrs postpartum after determining volume of FMH
- Abortions
- Trauma
- Procedures
- Transfusion of Rh+ products to an Rh- woman of child bearing age
HDN Summary

<table>
<thead>
<tr>
<th>Clinical Finding</th>
<th>Pathogenesis</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anemia</td>
<td>Alloimmune hemolysis</td>
<td>Intravenous, exchange and or simple transfusions</td>
</tr>
<tr>
<td>Jaundice</td>
<td>Hemolysis</td>
<td>Phototherapy</td>
</tr>
<tr>
<td>Erythroblastosis</td>
<td>Extramedullary erythropoiesis in the liver</td>
<td>Transfusion</td>
</tr>
<tr>
<td>Anasarca</td>
<td>Hypalbuminemia-CHF</td>
<td>Exchange transfusion</td>
</tr>
<tr>
<td>CHF</td>
<td>Anemia</td>
<td>Transfusion</td>
</tr>
</tbody>
</table>

Prevention = RhIg at 28 weeks and 72 hrs post partum, others

Autoimmune Hemolytic Anemia (AIHA)

- AutoAntibodies against RBC antigens
- Most are not significant
- Some cause hemolysis
- Risk increases with each decade of life
- Idiopathic
- SLE, CLL, Lymphomas, malignancies
- Cold agglutinin disease usually transient in children
- Cause Xmatch incompatibility with all RBC units
AutoImmune Hemolytic Anemia
(AIHA)

- Decreased Hg/Htc
- Increased Bilirubin
- Increased LDH
- +DAT
- +Ab Screen (IAT)
- Low or absent haptoglobin
- Peripheral smear with spherocytes, polychromasia and circulating nucleated RBCs

<table>
<thead>
<tr>
<th>AIHA</th>
<th>Disease Associations</th>
<th>C'</th>
<th>Clearance of Hemolysis</th>
<th>DAT/IAT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Warm Autoimmune HA</td>
<td>Autoimmune diseases Lymphoid malignancies</td>
<td>IgG (HA-Ag reaction at 37°C)</td>
<td>Extravascular [Fc receptors in liver and spleen]</td>
<td>Both IgG and C3 +DAT +IAT (Ab Screen)</td>
</tr>
<tr>
<td>Cold Agglutinin Disease</td>
<td>Post-infection (mycoplasma, IM) Lymphoid malignancies</td>
<td>IgM (HA-Ag reaction at 25°C or colder)</td>
<td>Extravascular [C3 receptors in liver]</td>
<td>+ DAT (C' only) + IAT</td>
</tr>
<tr>
<td>Drug Associated HA</td>
<td>Drug associated</td>
<td>Usually IgG</td>
<td>Extravascular</td>
<td>IgG +IAT in the presence of the drug in plasma</td>
</tr>
</tbody>
</table>

AIHA Management

- Warm
  - Steroids
  - IVIG
  - Immunosuppression
  - Splenectomy
- Cold
  - Supportive care
  - Keep patient warm
  - Treat underlying disease
- Drug induced
  - Stop drug exposure