Cleveland Clinic

Hemolytic Disease of the Fetus and Newborn

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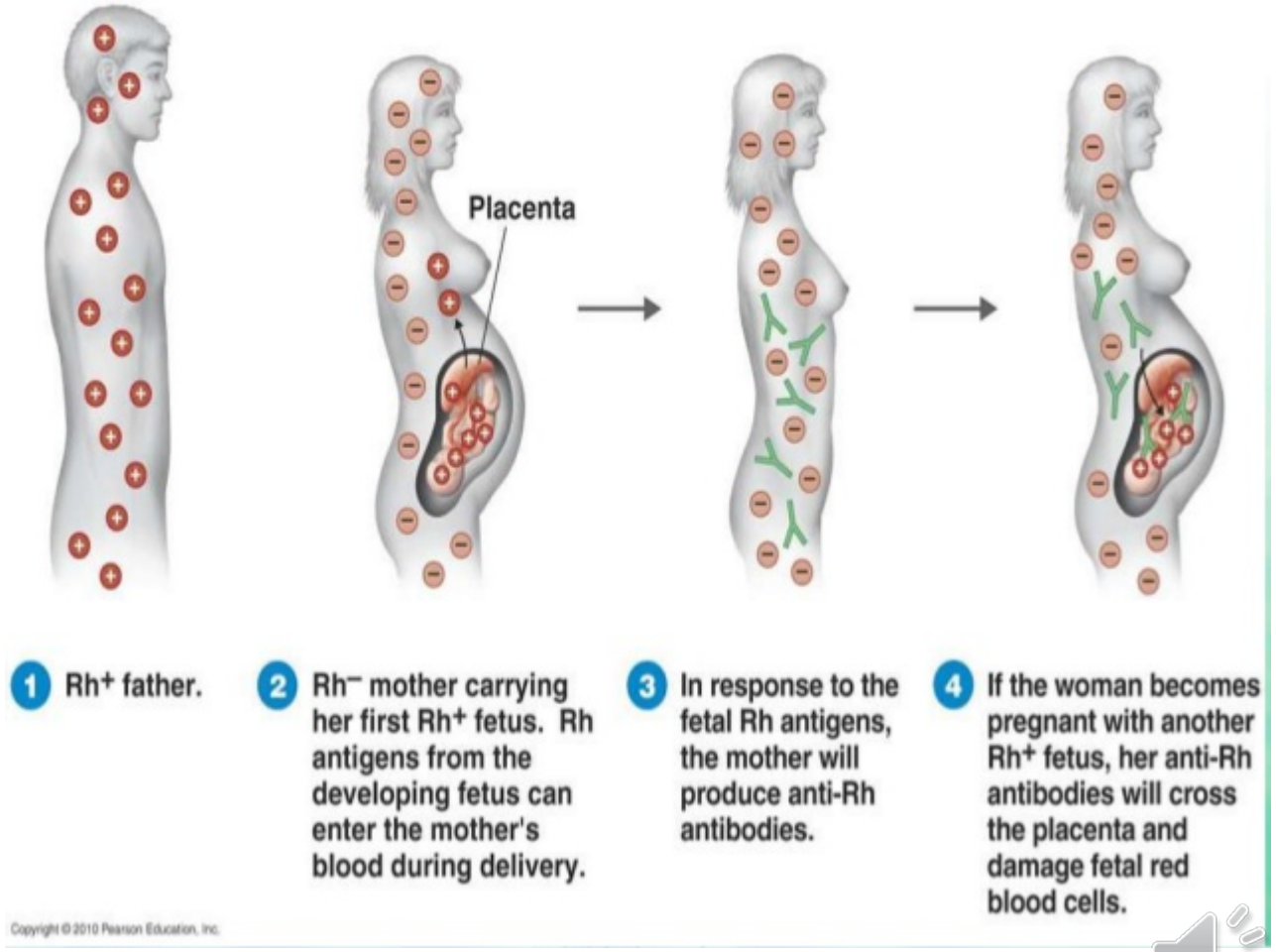
Hemolytic Disease of the Fetus and Newborn

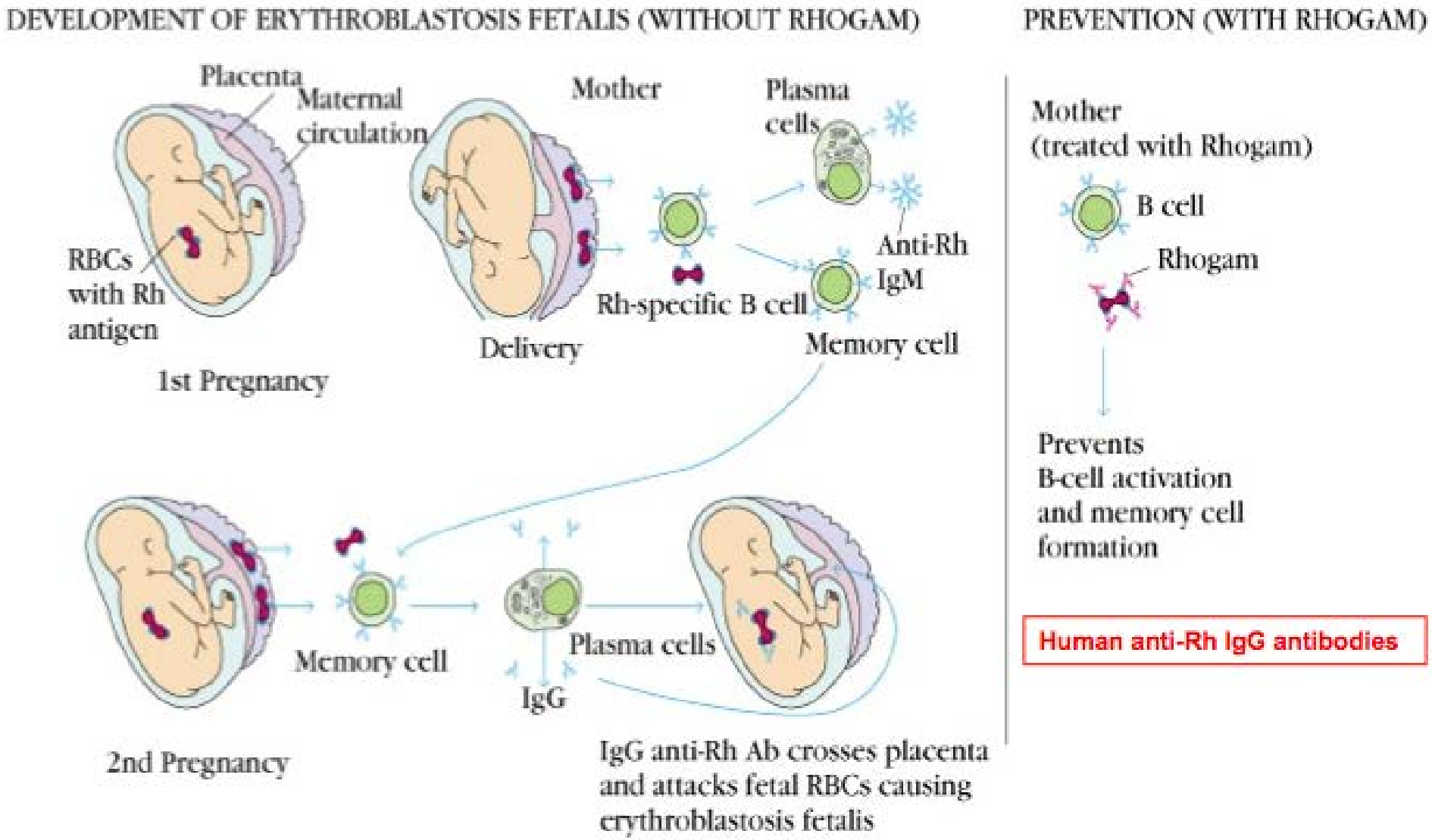
- Destruction of RBCs of a fetus and neonate by antibodies produced by the mother directed against paternal antigens inherited
- 95% used to be caused by anti-D
- Cases have decreased since Rhogam



What is Rhogam?

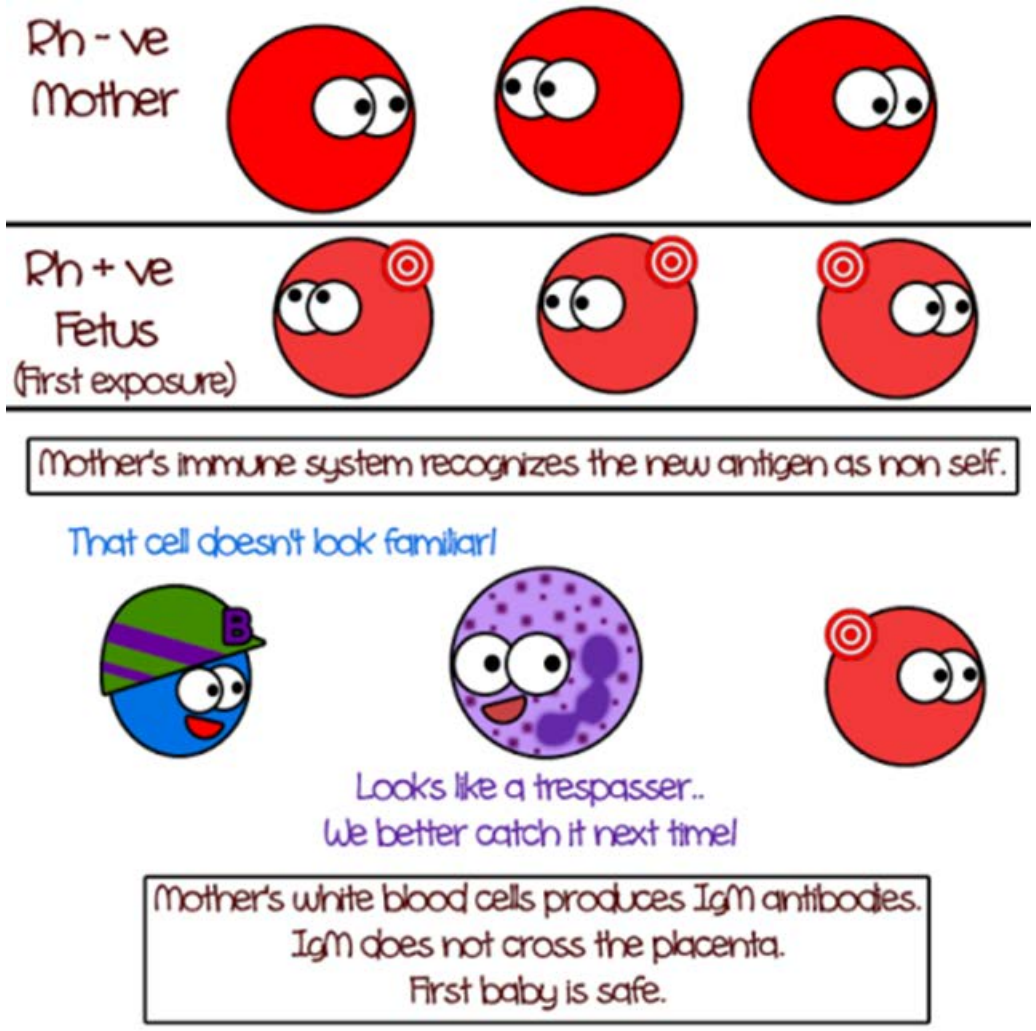
- Prevents immunization to D antigen
- Contains D antibody which attaches to fetal Rh-positive RBCs in maternal circulation
- Interferes with B-cell priming to make anti-D
- Fetal RBCs removed by spleen
- Administered intramuscularly or intravenously
- Only helps prevent HDFN due to anti-D

What is Rhogam?

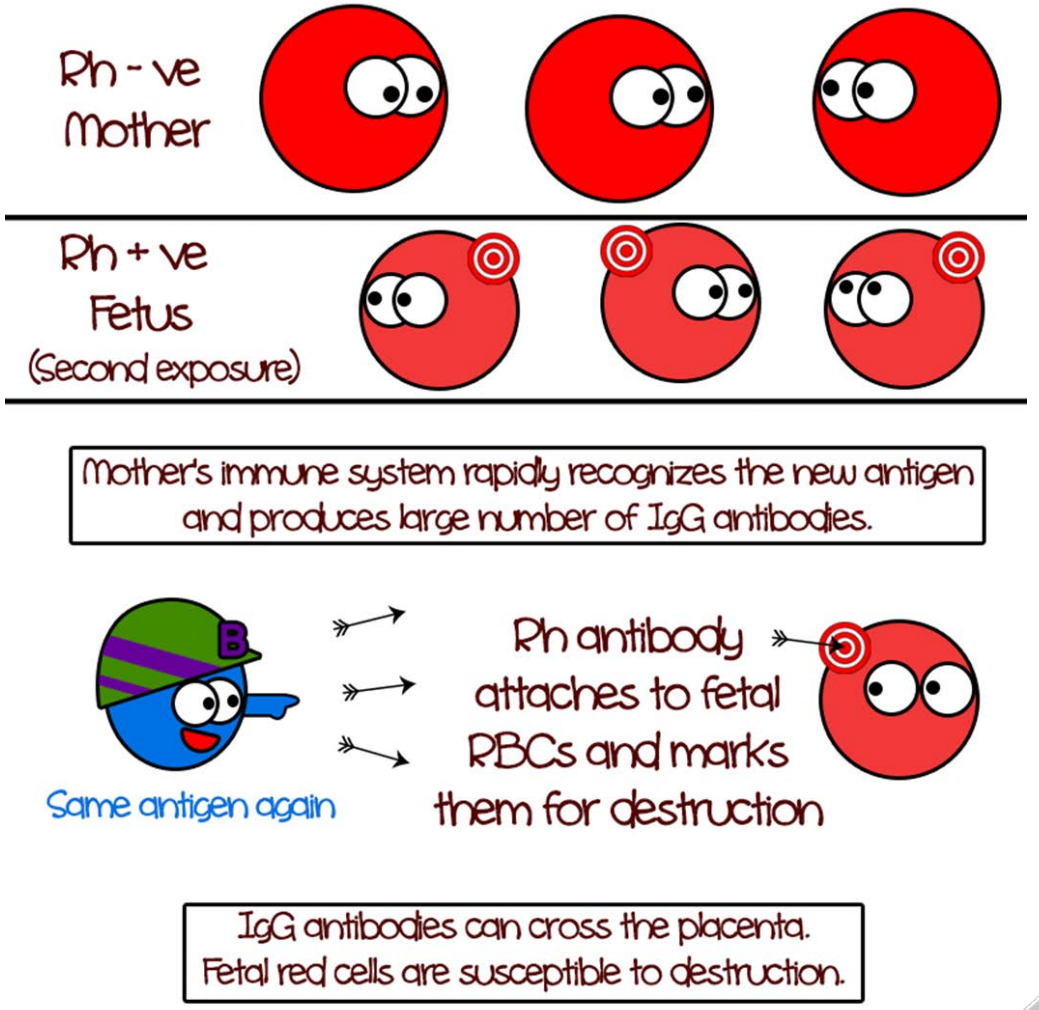


Pathogenesis

1st Pregnancy:



2nd Pregnancy:

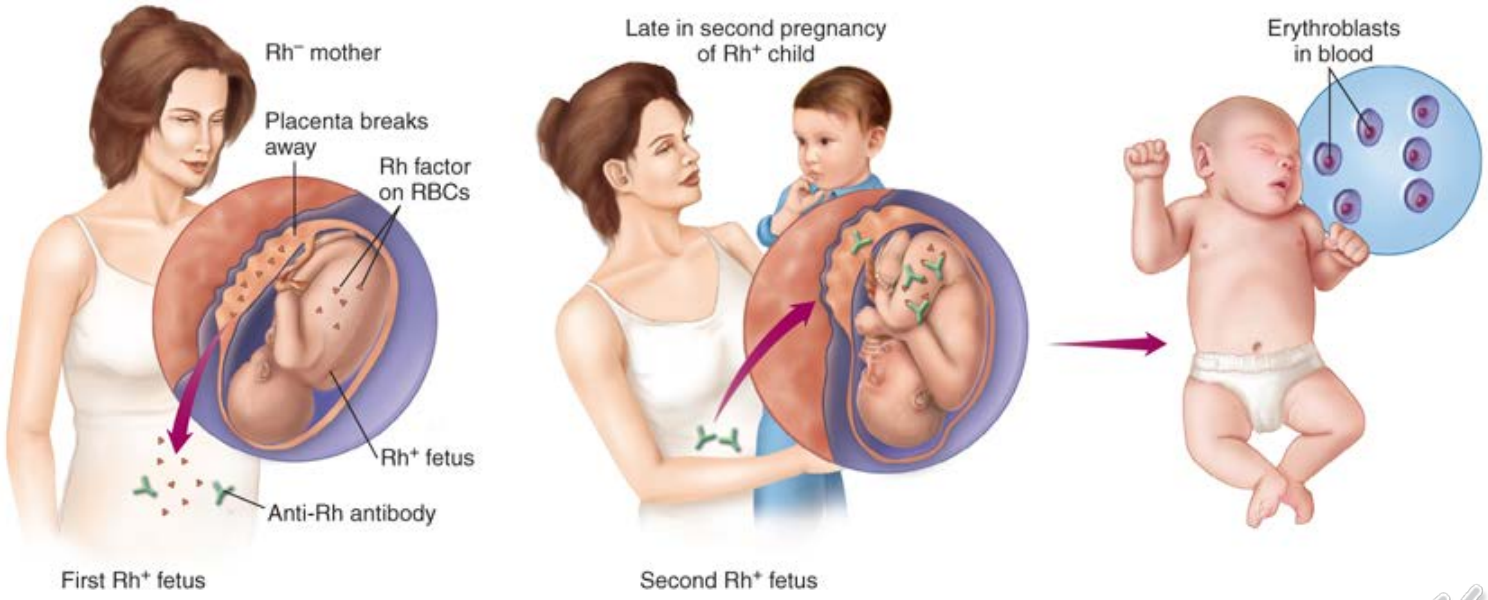


Pathogenesis

- Rate of destruction depends on antibody titer, specificity, and # of antigenic sites
- Destruction causes anemia
- Anemia stimulates fetal bone marrow to produce RBCs at accelerated rate
- Immature RBCs

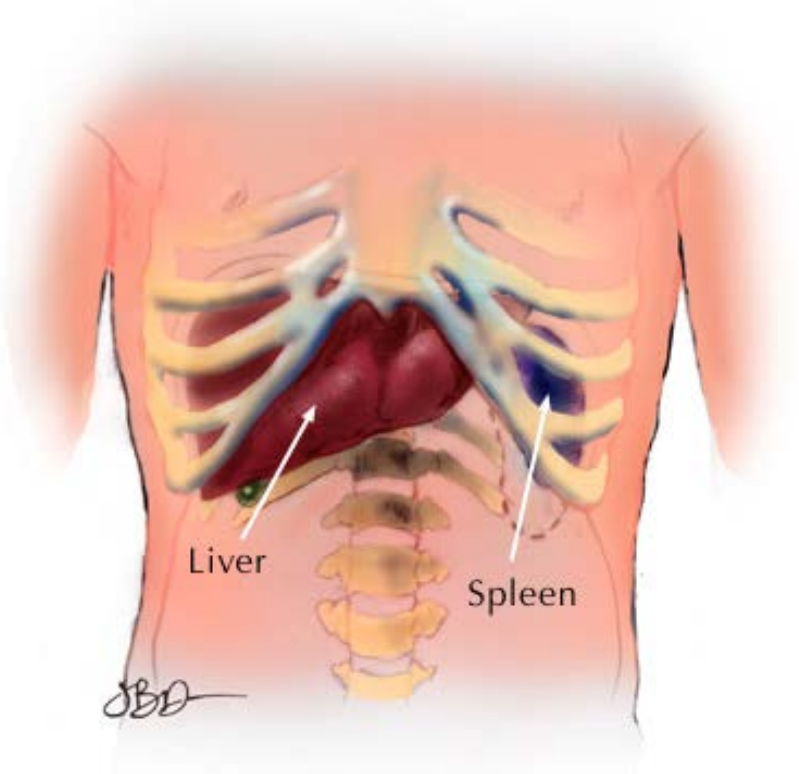
released into circulation

- Called *Erythroblastosis fetalis*



Pathogenesis

- Bone marrow can’t produce RBCs fast enough
- Increased RBC formation in hematopoietic tissues
  - Spleen
  - Liver
- Spleen and liver become enlarged
- Causes portal hypotension and liver damage



Pathogenesis

- **Liver damage leads to:**

Decreased production of plasma proteins

↓

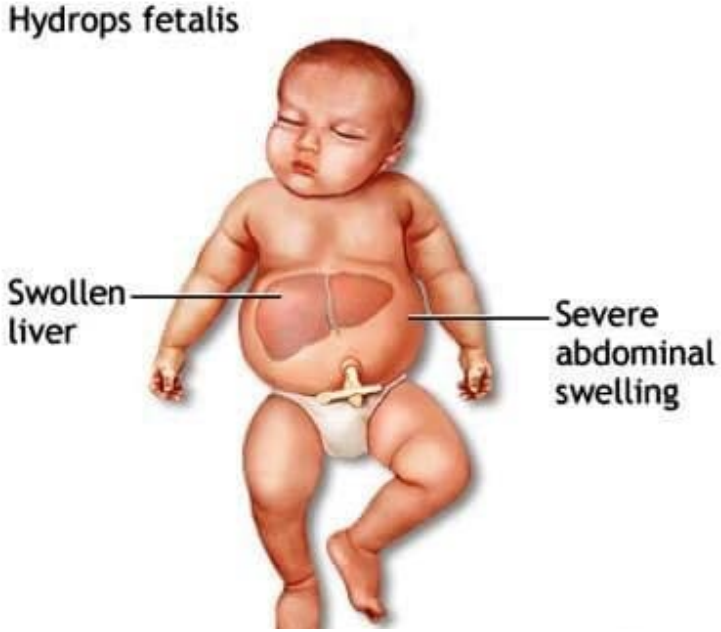
Hypoproteinemia

↓

Cardiac failure

Chain of reactions leads to *Hydrops fetalis (fetal hydrops)*

- Abnormal accumulation of fluid causing swelling including:
  - Edema (fluid beneath skin)
  - Ascites (fluid in abdomen)
  - Effusion (fluid around lungs)

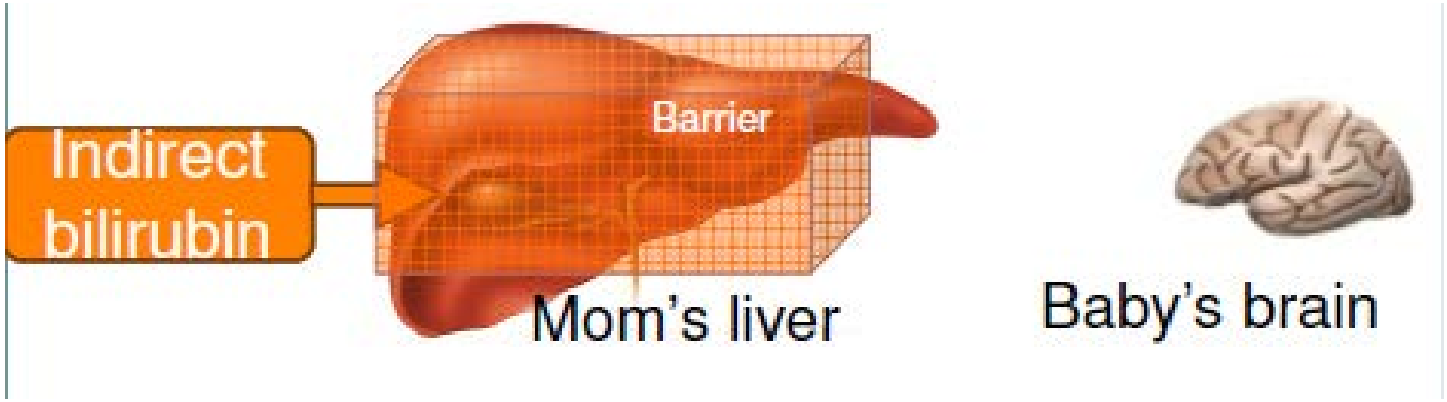


Pathogenesis

- RBC destruction continues while maternal antibody persists
- IgG has a half life of 25 days
- Destruction continues a couple weeks after birth

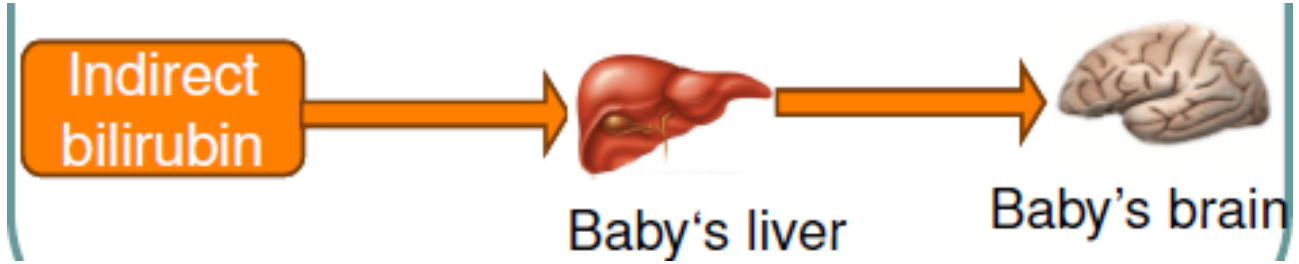
RBC Destruction- Before Birth

- Releases hemoglobin- metabolized to indirect bilirubin
- Crosses placenta- conjugated to direct bilirubin in mother's liver
- Direct bilirubin excreted by mother
- Bilirubin cannot accumulate or harm baby



RBC Destruction After Birth

- Mother cannot remove baby's bilirubin
- Newborn liver can not conjugate bilirubin effectively
- Bilirubin accumulates
- Can cause kernicterus: toxic levels of bilirubin accumulation in newborn's brain

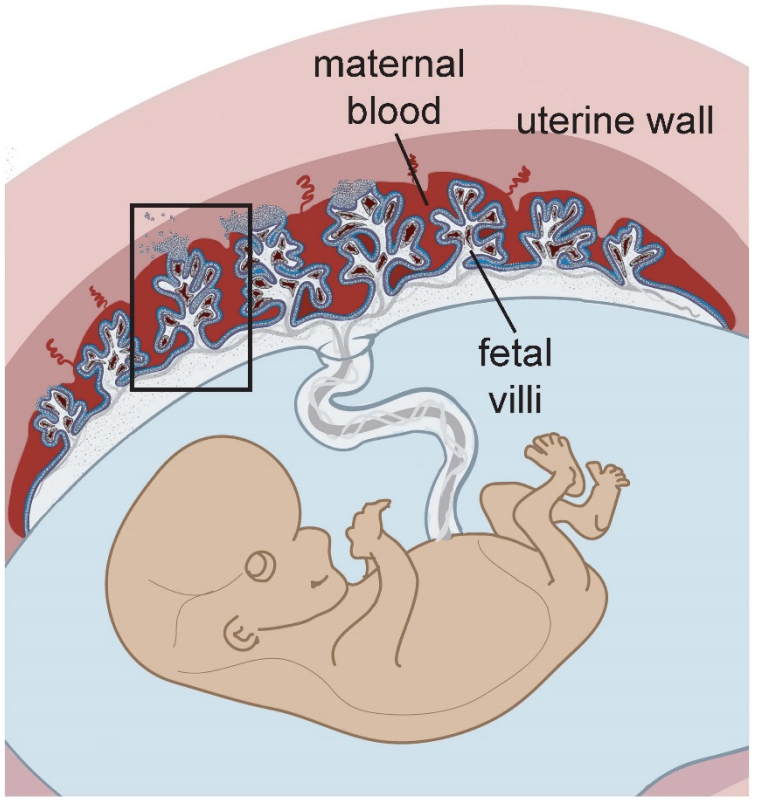


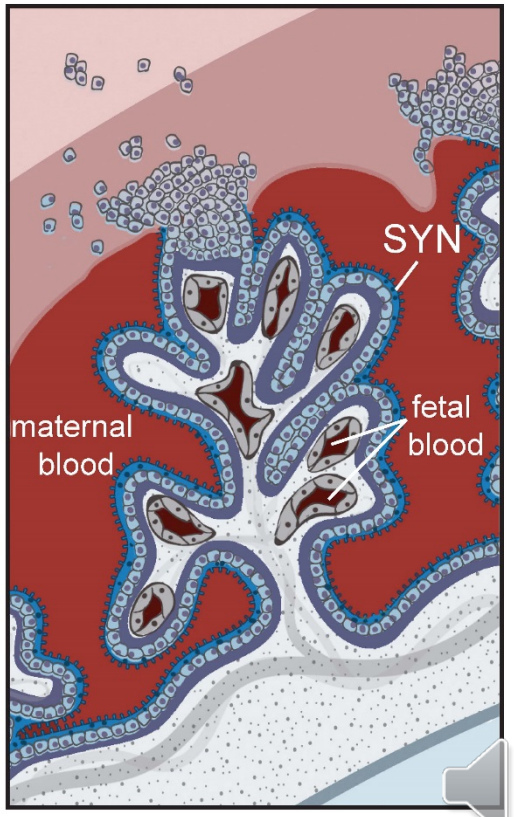
Factors Affecting Immunization and Severity of HDFN

- Antigenic Exposure
- Host Factors
- Immunoglobulin Class
- Antibody Specificity
- Influence of ABO Group

Antigenic Exposure

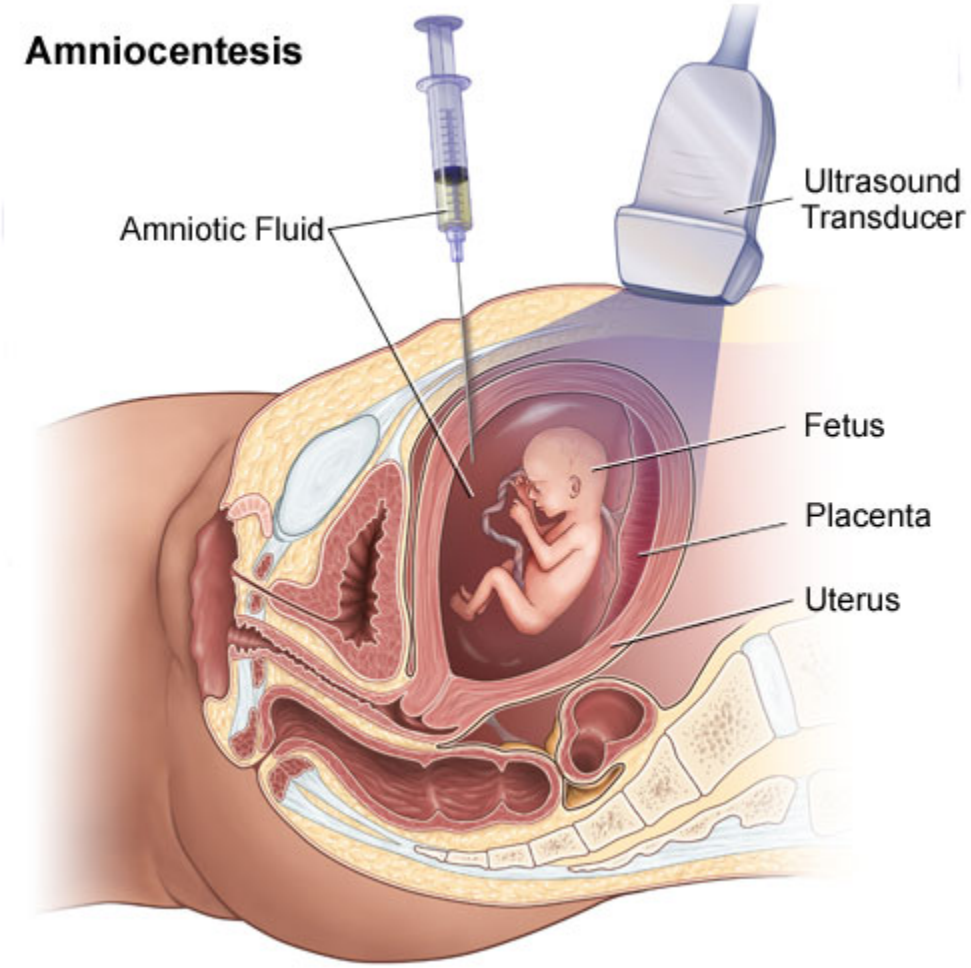
- Maternal and Fetal blood do not normally mix
- Exposed to fetal RBCs in previous pregnancies
- Fetomaternal hemorrhage causes increased antibody titers
  - Amniocentesis
  - Cordocentesis
  - Trauma to abdomen





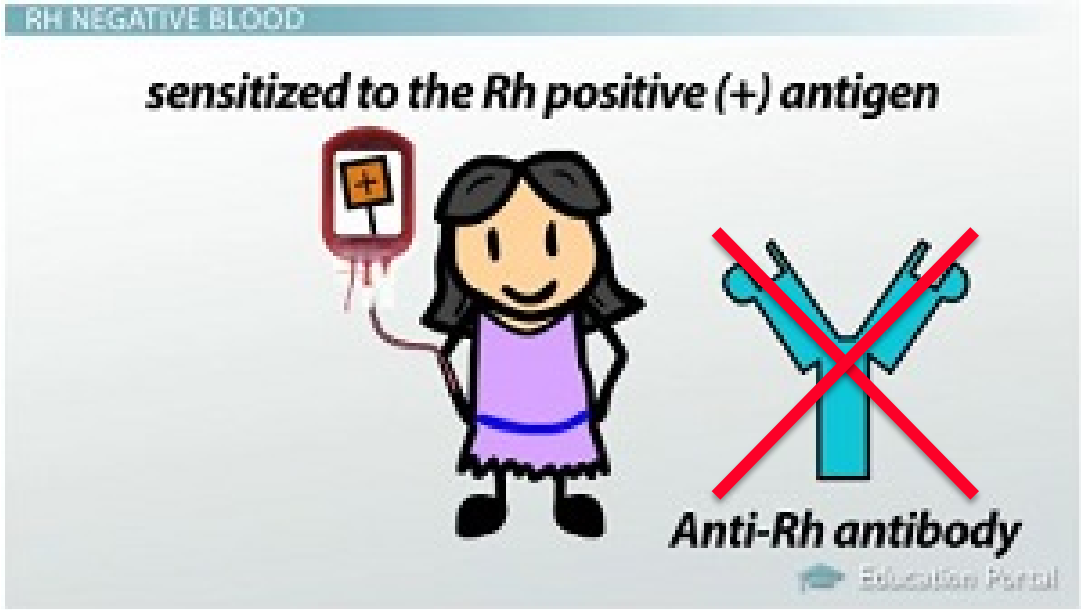
Antigenic Exposures

- At delivery: hemorrhage risk >50%
- Transplacental hemorrhage risk 7%
- 1 mL fetal RBCs can immunize mother



Host Factors

- Host ability to produce antibodies varies
- Depends mostly on genetic factors
- Ex. 15% of Rh negative individuals do not produce anti-D in response to exposure to Rh positive blood



Immunoglobulin Class

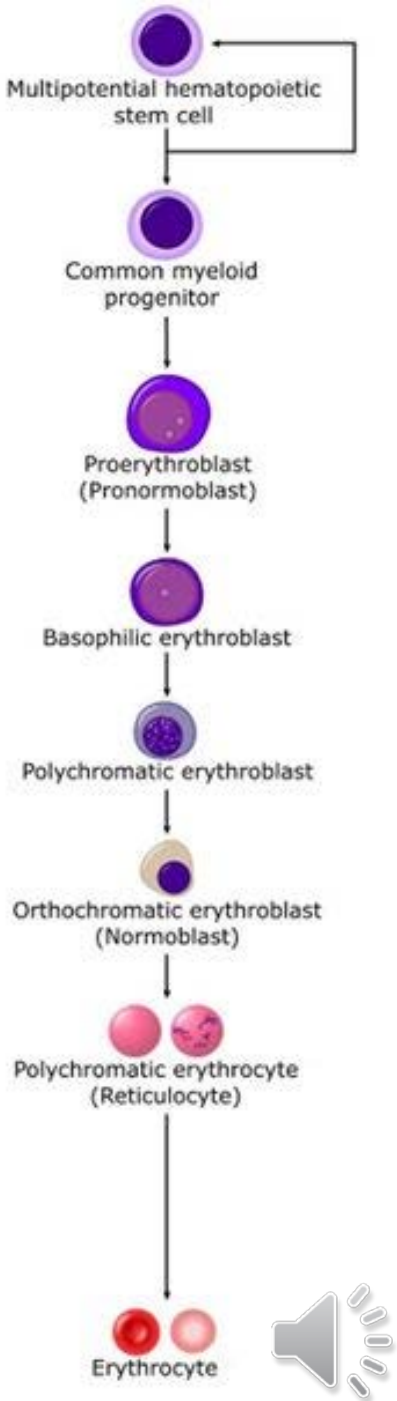
- **Only IgG can cross the placenta**
- **IgG1 and IgG3 are most efficient at hemolysis and cause most HDFN**

	IgG1	IgG2	IgG3	IgG4	IgA	IgM	IgE
Crosses Placenta	+	+	+	+	0	0	0
Fixes Complement	++	+	+++	0	0	+++	0
Binding to macrophage Fc receptors	+++	++	+++	+	0	0	0

Antibody Specificity

- 64 antigens have been associated with HDFN
- D is most immunogenic
- Other Rh's (C, E, c) also very immunogenic
- Anti-K most clinically significant in HDFN:
  - K is on erythroid precursors
  - Mature RBCs and erythroid precursors destroyed

Common	Rare	Never
Anti-D	Anti-Fy^a	Anti-Le^a
Anti-D+C	Anti-s	Anti-Le^b
Anti-D+E	Anti-M	Anti-I
Anti-C	Anti-N	Anti-IH
Anti-E	Anti-S	Anti-P_1
Anti-c	Anti-Jk^a	
Anti-e		
Anti-K		



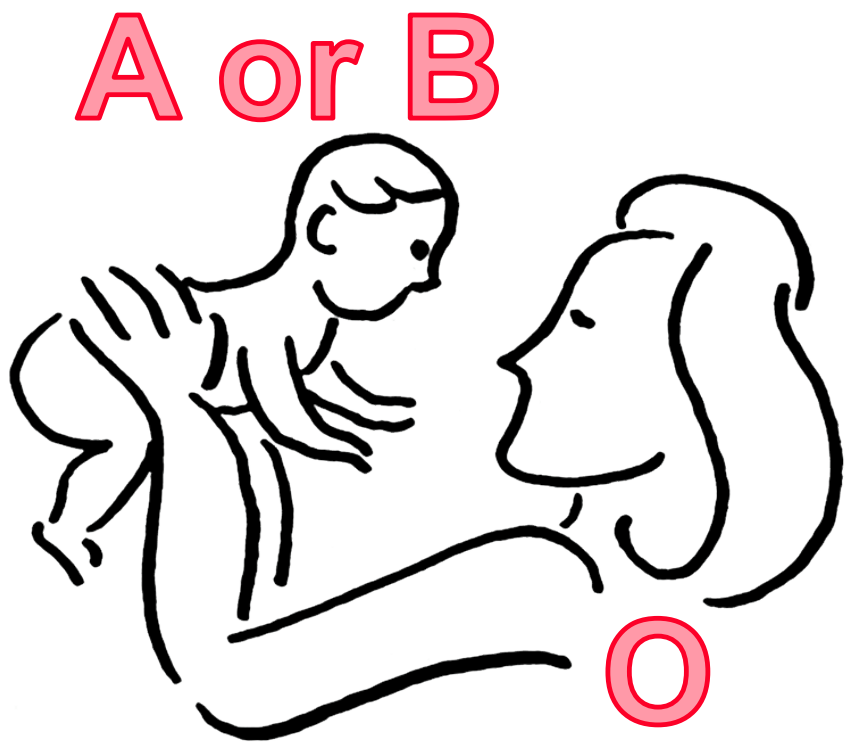
Influence of ABO Group

- If mother/fetus are ABO incompatible, D immunization is less
- Incompatibility protects against Rh immunization
- Incompatible ABO cells are destroyed by mother's circulation before the D antigen is recognized as foreign by the immune system
  - Not enough time to recognize and make anti-D

Risk of Alloiminization without RhIG
Rh incompatible ABO compatible: 16%
Rh and ABO incompatible: 2%

ABO HDFN

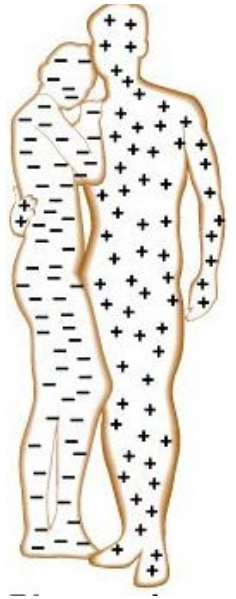
- ABO incompatibility between mother and infant
- IgG maternal ABO antibodies cross placenta
- Usually group O mother with A or B infant
  - Anti-A,B is IgG and usually made by O individuals
- Occur in first pregnancy- ABO antibodies naturally occurring
- A and B antigens not fully developed at birth- weaker HDFN



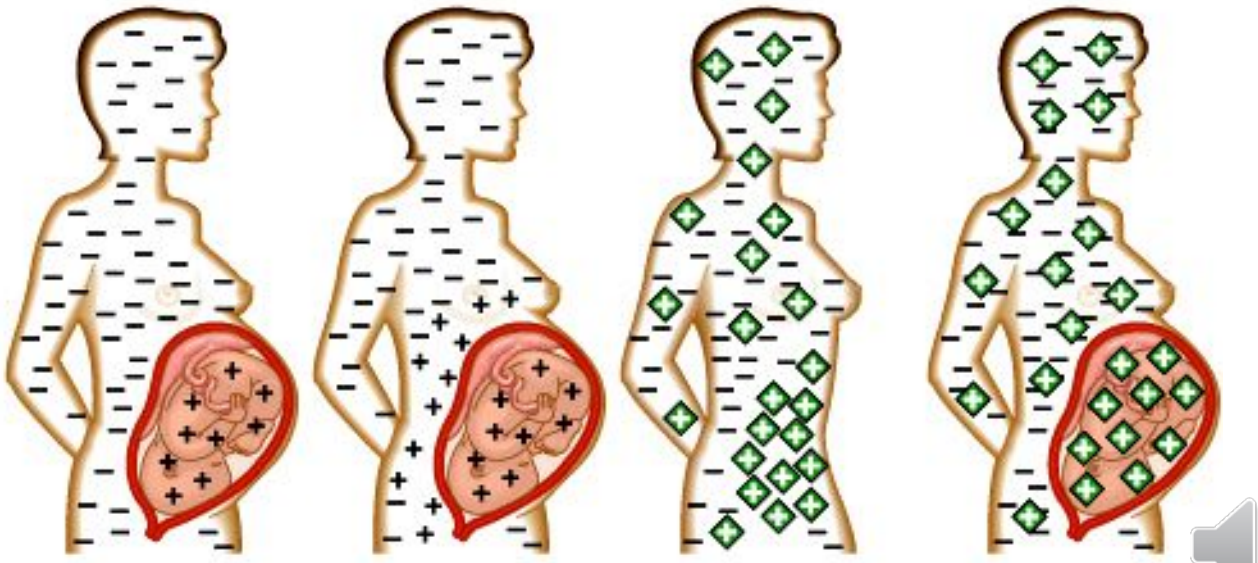
ABO HDFN

- Destruction of RBCs rarely leads to severe anemia
- Hyperbilirubinemia and jaundice 12-48 hours after birth
- Treatment: phototherapy
- Since Rhogam, ABO HDFN= most common cause of HDFN

Rh HDFN



- Delivery- many fetal RBCs can enter maternal circulation
- D antigen inherited from father, mother (D negative) becomes immunized and produces anti-D
- Anti-D will affect all subsequent children (can cross placenta and bind fetal Rh positive cells)
- First child therefore not affected
- Can cause severe anemia



Comparing ABO and Rh HDFN

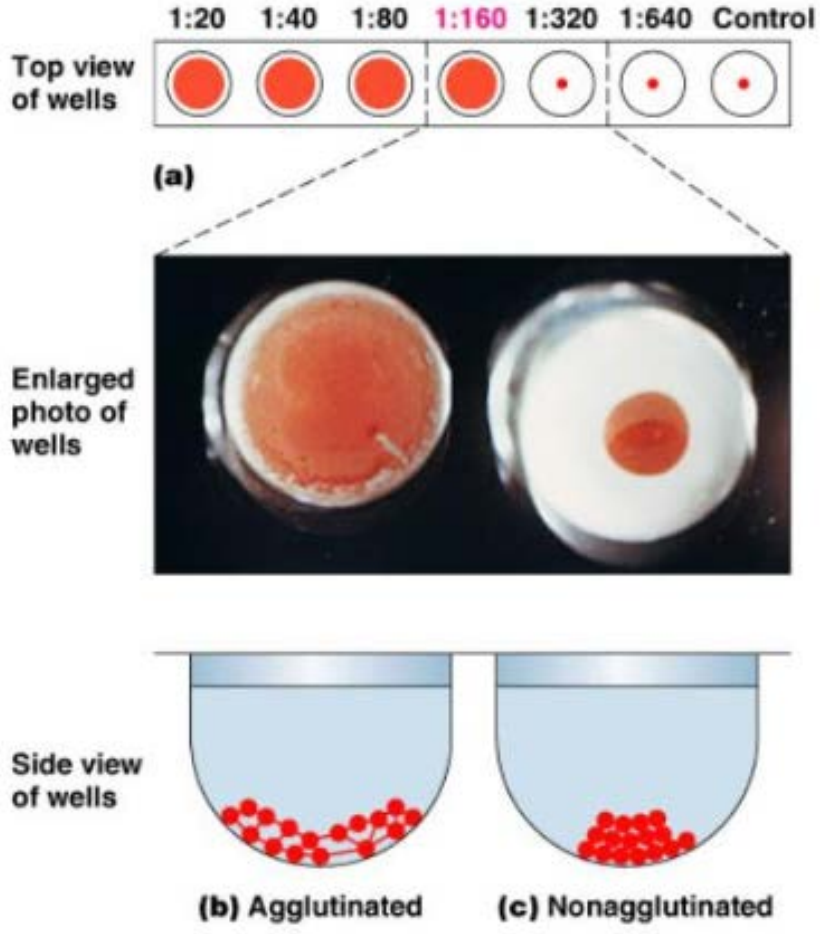
Characteristic	ABO	Rh
First Pregnancy	Yes	Rare
Disease predicted by titers	No	Yes
Antibody IgG	Yes	Yes
Bilirubin at birth	Normal Range	Elevated
Anemia at birth	No	Yes
Phototherapy	Yes	Yes
Exchange transfusion	Rare	Uncommon
Intrauterine transfusion	None	Sometimes

Serologic Testing of the Mother

- **ABO, Rh, Antibody Screen**
  - Preferably in first trimester
  - Weak D testing for Rh
  - Antibody screen negative- repeat at 28 weeks if Rh negative before Rhogam given
  - Antibody screen positive- ID antibody and perform titration

Serologic Testing of the Mother

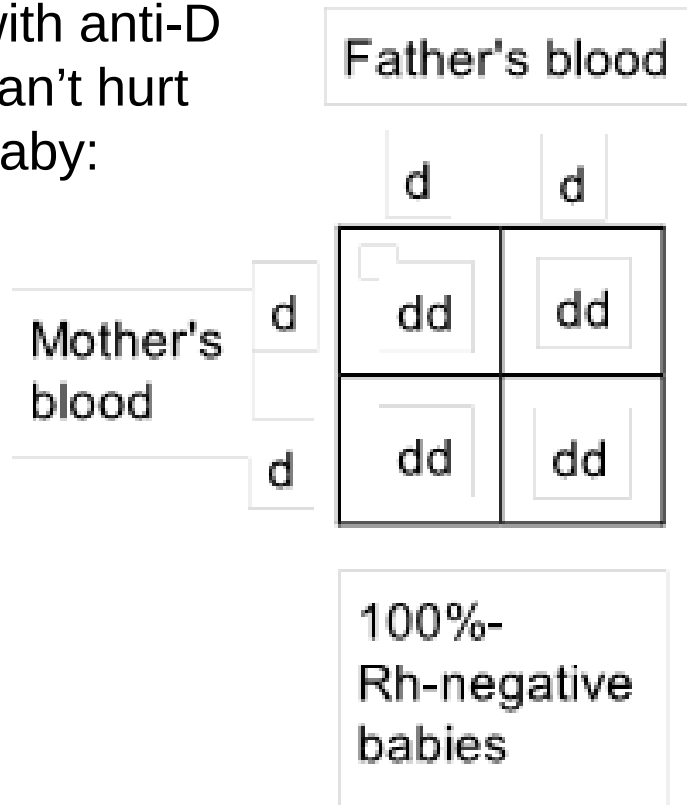
- **Antibody Titer**
  - Determine concentration of antibodies capable of crossing the placenta
  - Repeat each month to determine if concentration increases
  - Perform previous months sample at same time
  - Difference of 2 dilutions= significant
  - A titer of 16 is critical
  - After 16, patient is followed using MCA-PSV



Serologic Testing of the Father

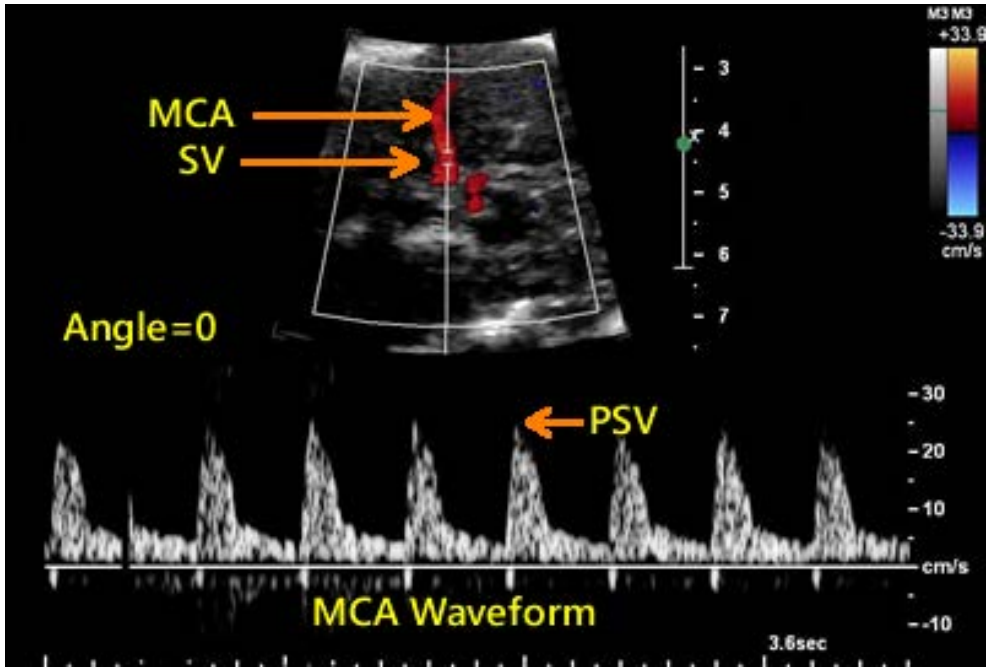
- **Paternal Phenotype**
  - Test father's red cells for antigen the mother has an antibody to
  - If father negative, baby must be negative as well and HDFN is not a concern

Mother with anti-D can't hurt baby:



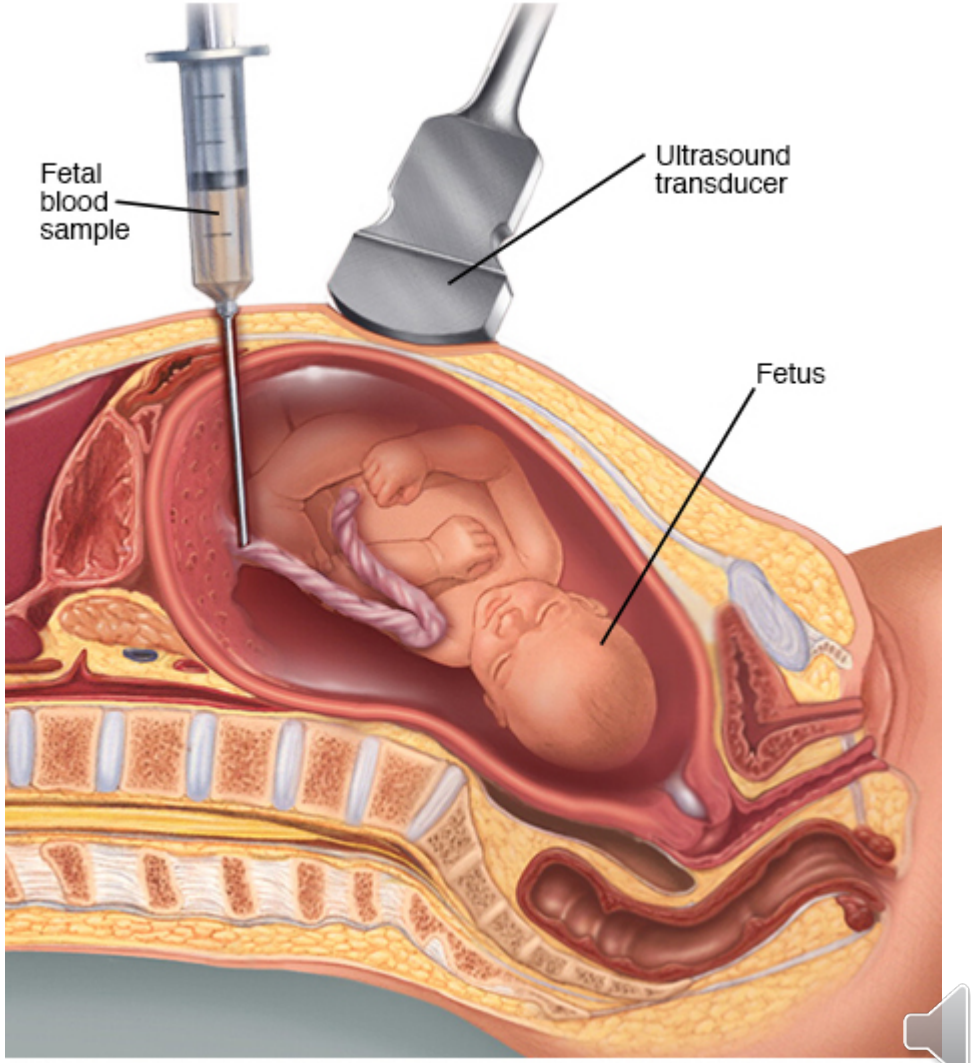
Other Testing

- **Color Doppler Middle Cerebral Artery Peak Systolic Velocity (MCA-PSV)**
  - Non-invasive
  - Done between 16-20 weeks if anemia is suspected
  - Reliably predicts anemia in fetus
  - Detects blood flow in brain
    - Will increase if anemic (vasodilation and increased cardiac output)



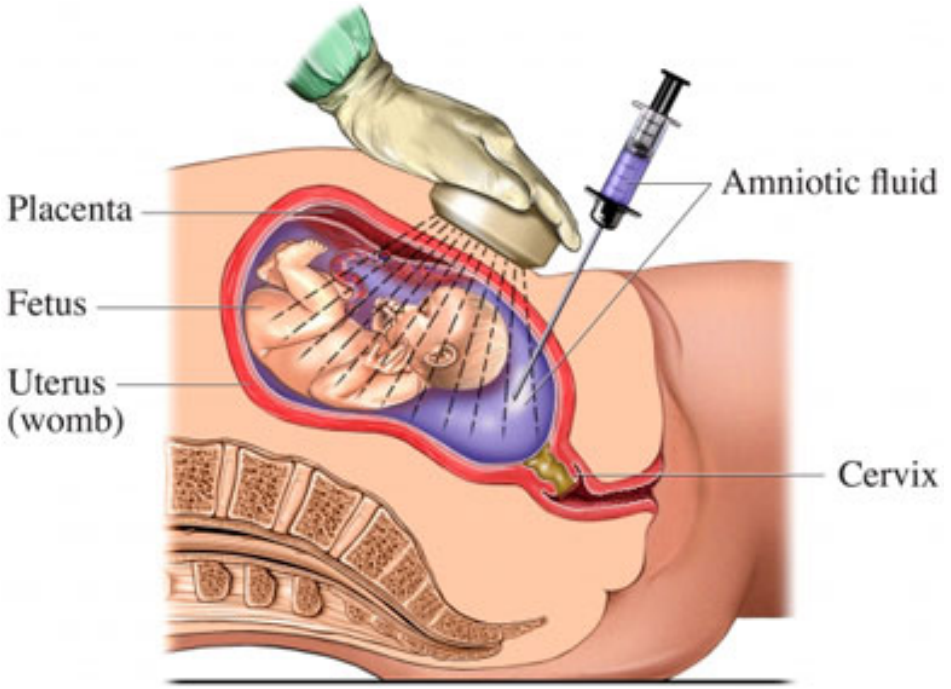
Other Testing

- **Cordocentesis**
  - Umbilical visualized with color Doppler to obtain fetal blood
  - Test blood for: hemoglobin, hematocrit, bilirubin, blood type, DAT, antigen phenotype/genotype
  - Invasive- more dangerous



Other Testing

- Amniocentesis
  - Obtain sample of amniotic fluid
  - Measure concentration of bilirubin pigment in amniotic fluid
    - Estimates extent of fetal hemolysis
  - Increase overtime indicates HDFN
  - Invasive
  - Not used very often- same information as MCA-PSV which is non-invasive

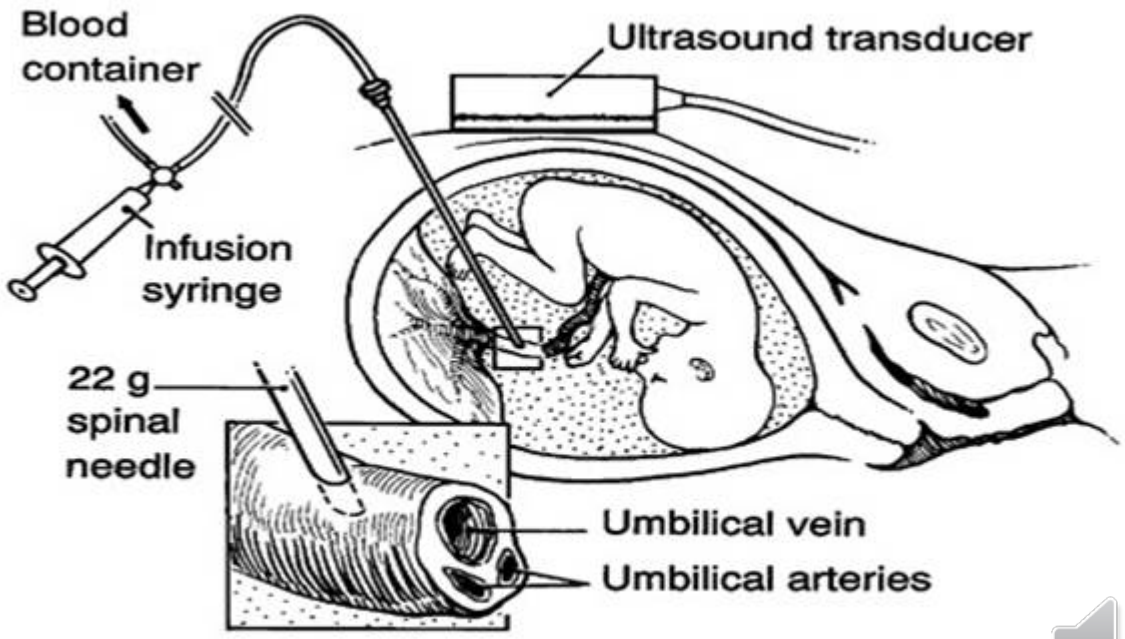


Treatment for Anemic Fetus

- **Intrauterine Transfusion**
  - Access fetal umbilical vein (cordocentesis)
  - Inject RBCs directly into vein
  - Repeat every 2-4 weeks until delivery

Indications:

- MCA-PSV indicates anemia
- Fetal hydrops noted on ultrasound
- Cordocentesis sample Hgb <10g/dL
- Amniotic fluid results high

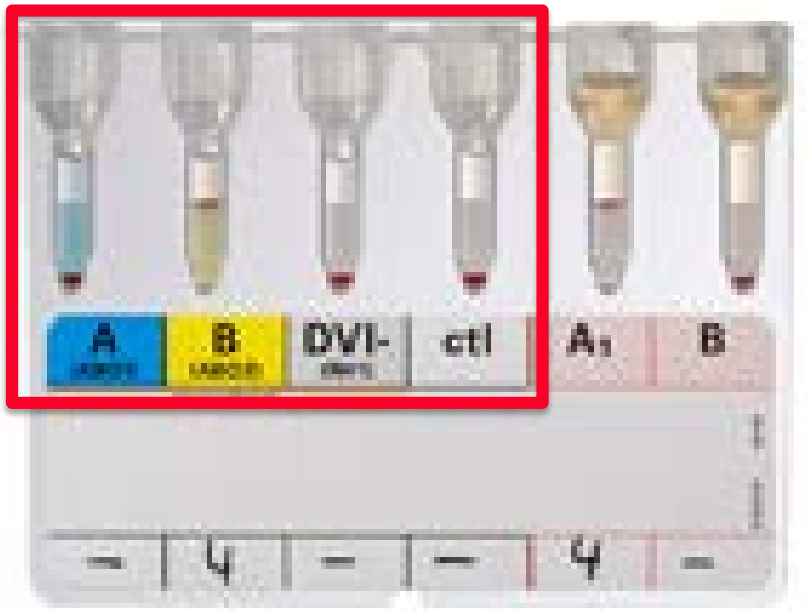


Products Used for Intrauterine Transfusion

- O Negative RBCs
- <7 days post phlebotomy
- CMV negative
- Hemoglobin S negative
- Antigen negative for maternal antibody if applicable
- Irradiated RBCs

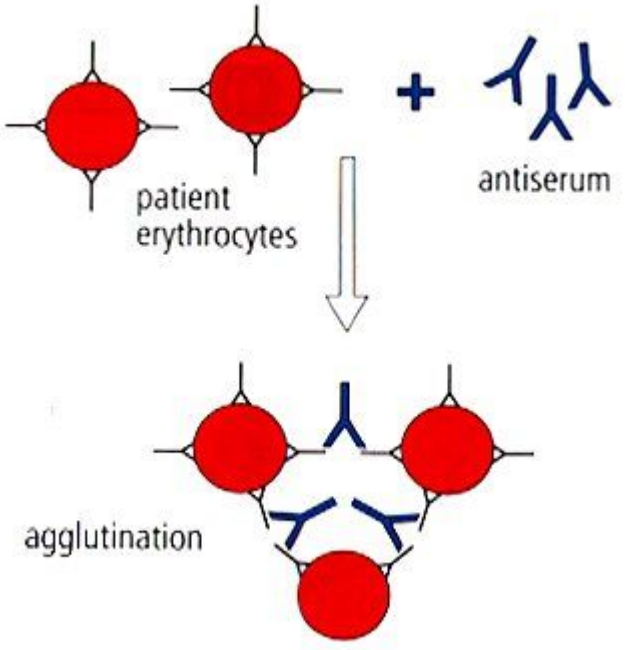
Serologic Testing of Neonate

- Performed on cord blood sample after birth
- ABO/Rh Grouping
  - Only forward type (can't make ABO antibodies yet)
  - Weak D if Rh negative (determine if mother needs Rhogam)



Serologic Testing of Neonate

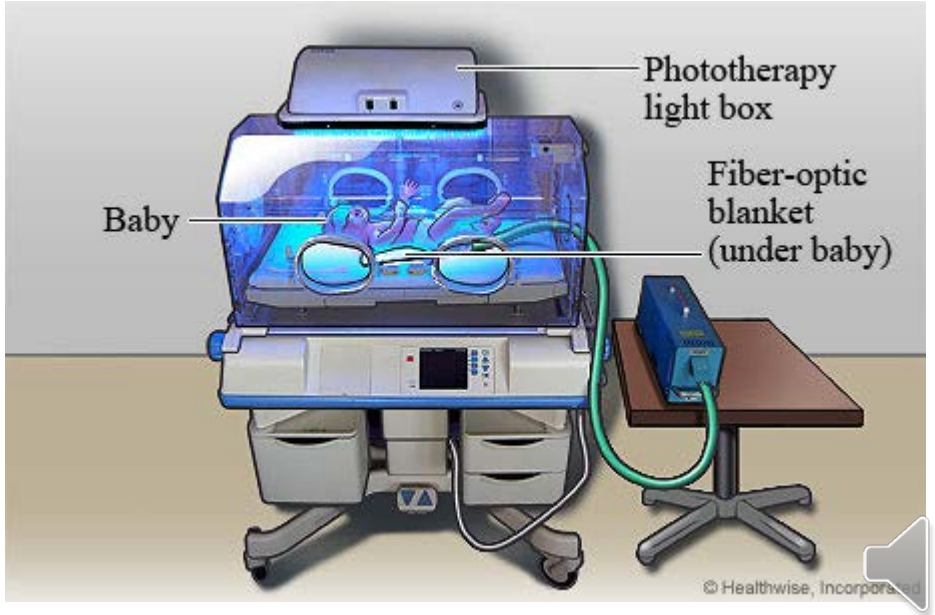
- DAT
  - If positive, may have HDFN
  - Strength of reaction does not = severity of HDFN
- Eluate
  - Not needed for ABO incompatibility- therapy won't change based on results
  - Helpful if cause of HDFN is in question



Positive DAT

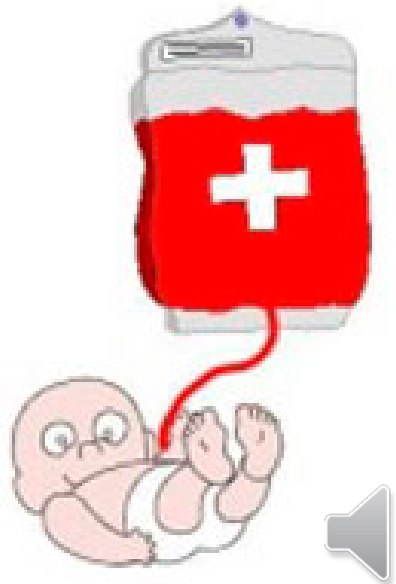
Treatment for Newborn

- **Phototherapy**
  - Fluorescent light absorbed by baby's skin
  - Bilirubin accumulates from RBC destruction (liver can't remove)
  - Changes unconjugated bilirubin to isomers which are less toxic to the brain and more easily excreted



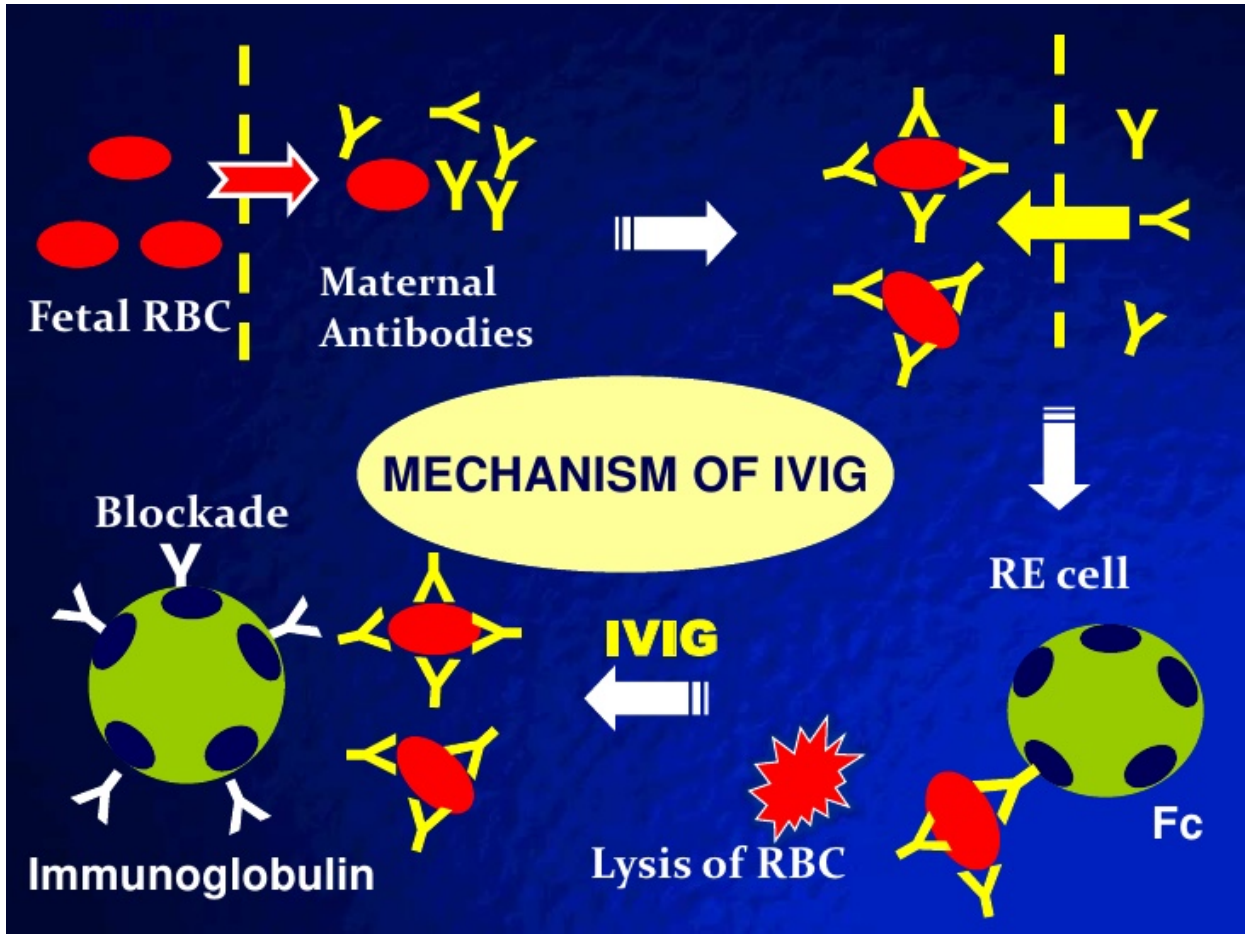
Treatment for Newborn

- **Simple Newborn Transfusion**
  - Small aliquots of RBCs to correct anemia
  - <10 g/dL Hgb requires transfusion
  - Normal for newborn = 14-20 g/dL Hgb
  - Same type of blood product as intrauterine transfusion



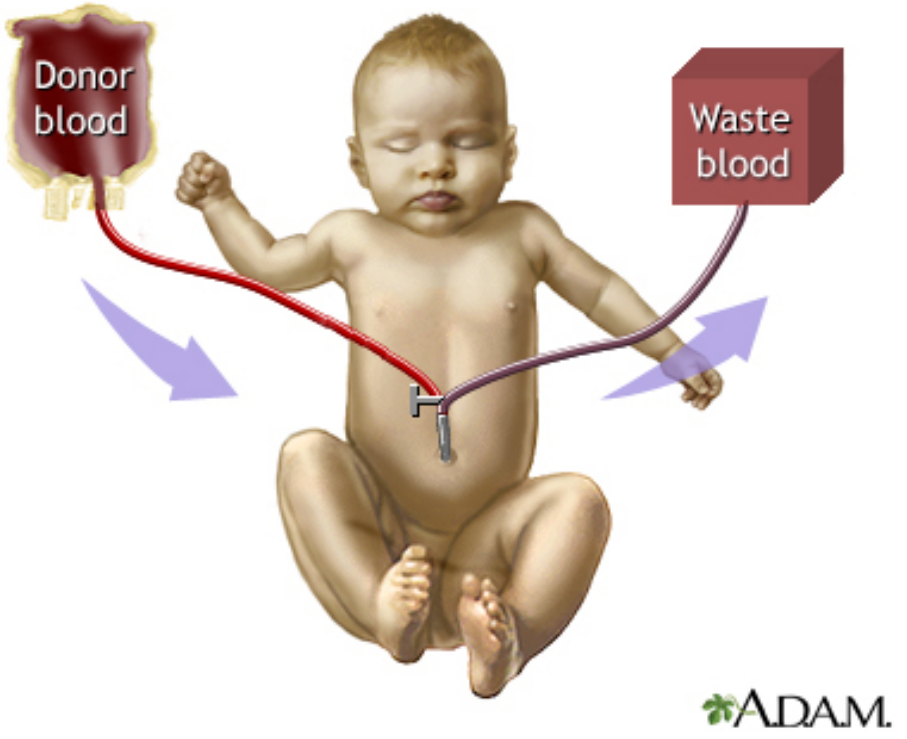
Treatment for Newborn

- **IVIG (intravenous immune globulin)**
  - IVIG blocks FC receptors on phagocytes
  - Phagocytes can’t bind antibody and do not destroy red cell
  - Treats hyperbilirubinemia of newborn



Treatment for Newborn

- **Exchange Transfusion**
  - Phototherapy and IVIG help prevent need for exchange transfusions
  - Use whole blood to replace circulating blood
  - Removes high levels of unconjugated bilirubin to prevent kernicterus
  - Removes maternal antibodies and sensitized cells
  - Replace incompatible RBCs with compatible ones



Rhogam Indication

- First dose given at 28 weeks to all Rh negative mothers
- Second dose given after birth (within 72 hours) if infant is Rh positive
- No benefit of Rhogam if mother already has anti-D
- Rh positive mothers do not need Rhogam

Rhogam Indication

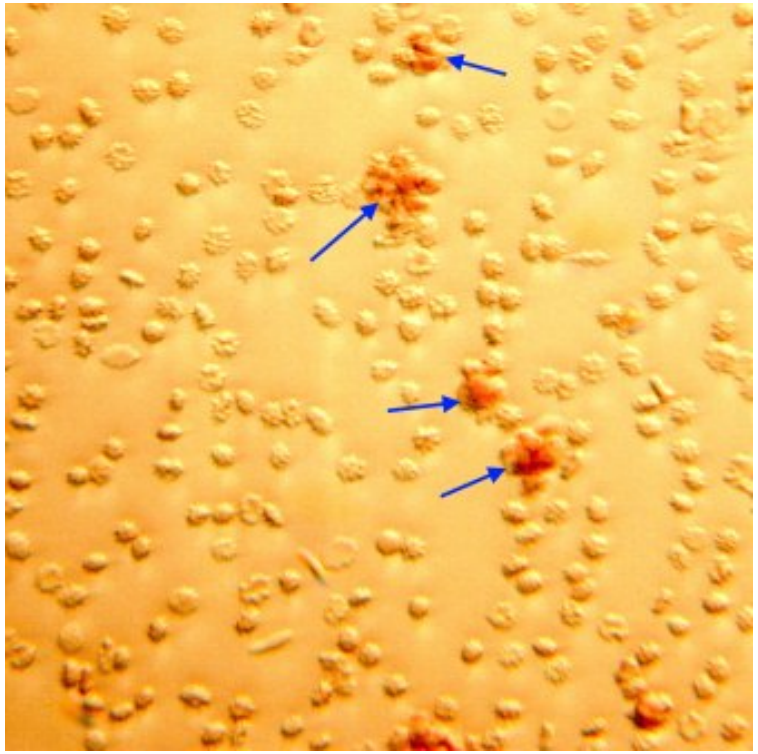
- Anything that could allow fetal RBCs into maternal circulation
- Amniocentesis
- Cordocentesis
- Abortion
- Ectopic pregnancy
- Abdominal trauma
- Delivery

Postpartum Indication for Rhogam

- One dose of RhIG contains enough anti-D (300µg) to protect against 15 mL of packed RBCs or 30 mL of whole blood
- Massive fetal maternal hemorrhage (more than 30 mL of whole blood) occur in <1% of deliveries
- Need to calculate how much Rhogam to give depending on the size of the fetal maternal hemorrhage

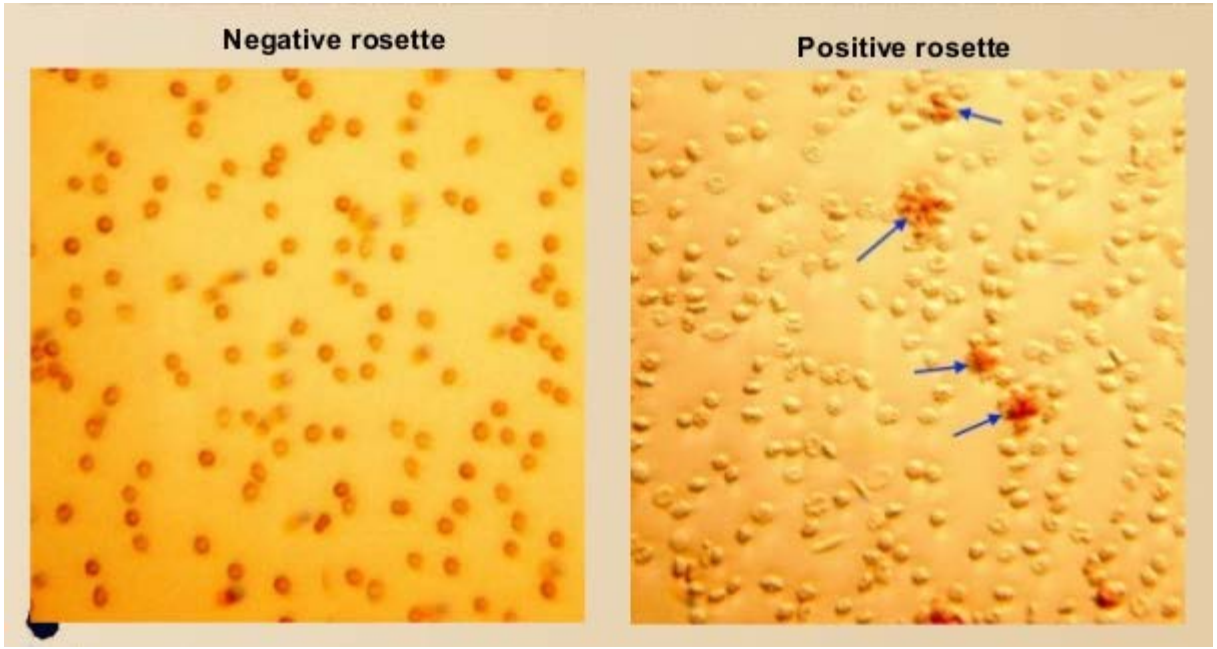
Determining Fetomaternal Hemorrhage

- Rosette Test
  - Qualitative test
  - Screen maternal sample after delivery for hemorrhage
  - Make suspension of mother's RBCs
  - Add anti-D reagent to bind to any D positive fetal cells (incubate)
  - Wash and add D positive indicator cells- bind anti-D bound Rh positive fetal cells forming Rosette



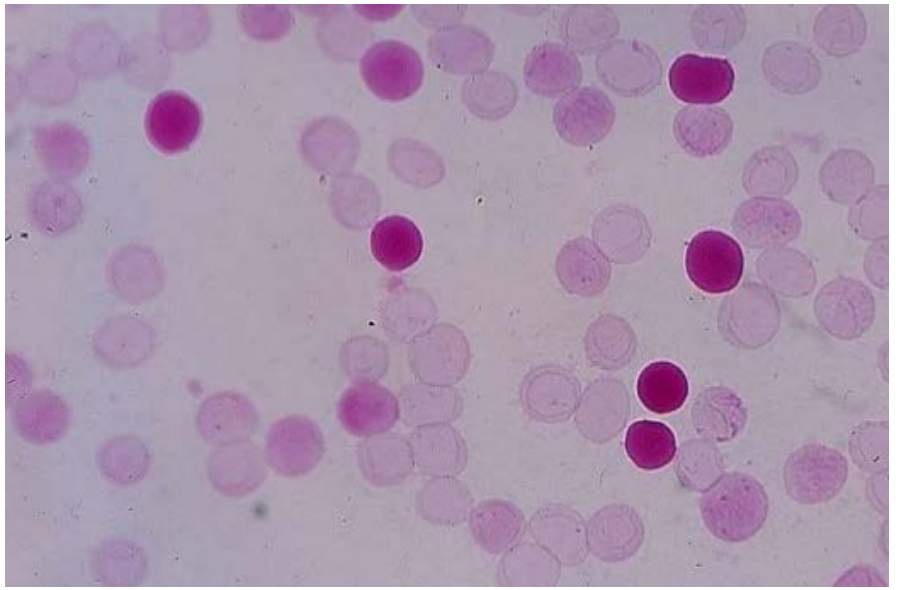
Determining Fetomaternal Hemorrhage

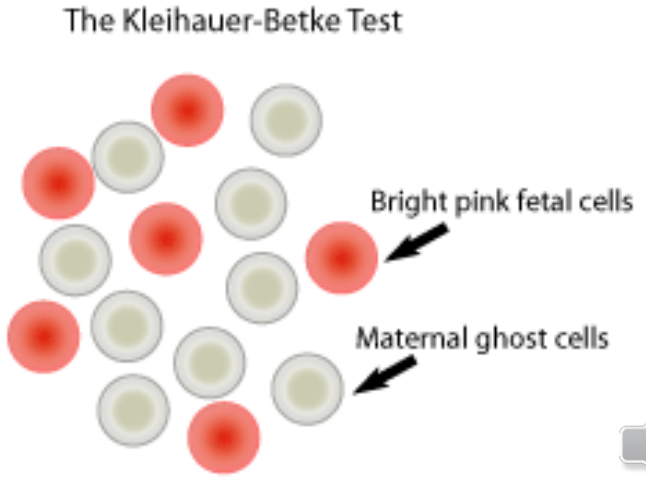
- Rosettes are detected microscopically
- If negative- give one dose of Rhogam just in case
- If positive- determine dose necessary for size of the bleed
  - Perform Kleihauer-Betke Test



Determining Fetomaternal Hemorrhage

- Kleihauer-Betke Acid Elution
  - Quantitative- determines how large the bleed is
  - Maternal blood smear treated with acid and stained and counterstained
  - Fetal Hemoglobin (Hgb F) = resistant to acid, remains pink
  - Maternal cells = not resistant, appear as ghost cells
  - 2,000 cells counted and percentage of fetal cells is determined





Kleihauer-Betke Calculations

- Step One: Calculate the % fetal cells seen:

$$\frac{\#\text{ of fetal cells}}{\text{Total \# of RBCs}} \text{ x } 100 = \%\text{ fetal cells}$$

Kleihauer-Betke Calculations

- Step Two: Calculate total mL of bleed:

$$\frac{\%\ fetal\ cells}{100}\ \text{x}\ 5{,}000 = \text{total mL bleed}$$

OR

$$\%\ \text{fetal cells x}\ 50 = \text{total mL bleed}$$

Note: 5,000 mL is the average blood volume of mom

Kleihauer-Betke Calculations

- Step Three: Calculate the number of vials of Rhogam to be given:

$$\frac{\text{Total mL of bleed}}{30\ \text{mL}} = \#\ \text{of vials to be given} + 1$$

Notes: 30 mL is the equivelant of 1 vial of Rhogam.

Always add one extra to the vial number to ensure enough Rhogam is given.

Cleveland Clinic

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