PREVENTION OF RhD ALLOIMMUNIZATION: HOW DO WE ENSURE THIS HAPPENS?

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I have no financial interests to disclose

LEARNING OBJECTIVES

Review causes of RhD alloimmunization

Talk about ways to prevent RhD disease

Review recommended OB prevention protocols

Examine prevention in the setting of mismatched blood transfusions

WHAT IS ALLOIMMUNIZATION?

- *Alloimmunization refers to an immunologic reaction against foreign antigens that are distinct from antigens on an individual's cells.*

WHAT IS RhD ALLOIMMUNIZATION?

- Human blood groups discovered 1900
- 1980 International Society Blood Transfusion working committee
- Genetically-based numerical terminology for red cell surface antigens
- Challenges in everyday communications led to variety of alternative names used for some blood groups

WHAT IS RhD ALLOIMMUNIZATION?

- **Rh D** to signify the **erythrocyte antigen**
- Women who **carry Rh D antigen** are identified as **Rh D positive**
- Those **not carrying Rh D antigen** are identified as **Rh D negative**
- ***RhD alloimmunization refers to the maternal formation of antibodies against fetal Rh D***

POTENTIAL DISEASE PREVALENCE

- Wide variation in prevalence rates of Rh D-negative individuals
- 5% in India
- 15% in North America
- Most common European / North American descent (15–17%)
- Comparatively decreased Africa / India (3–8%)
- Rarest in Asia (0.1–0.3%)
- 40% of infants of Rh D-negative women will be Rh D negative

BURDEN OF Rh D ALLOIMMUNIZATION DISEASE

- Advances in the prevention and treatment of Rh D alloimmunization have been one of the great success stories of modern obstetrics
- 1970s – 1st introduced postpartum administration of Rh D immune globulin
- Reduction of alloimmunization rate from **13 – 16% to ~ 0.5 – 1.8%**
  - *(deHaas M, Finning K, Massey E, Roberts DJ. Anti-D prophylaxis: past, present and future. Transfus Med 2014;24:1–7*
  - *Bowman J. Thirty-five years of Rh prophylaxis. Transfusion 2003;43:1661–6*

FURTHER DECREASE BURDEN OF DISEASE

This risk was further reduced to 0.14–0.2% with the addition of routine antepartum administration (28 weeks)

(deHaas M, 2014;Bowman J. 2003)

WITHOUT PREVENTATIVE PROGRAMS

- High birth rates in low prevalence areas means Rh hemolytic disease of the newborn is still an important cause of morbidity and mortality in countries without prophylaxis programs

- 14% of affected fetuses are stillborn and one half of live born infants suffer neonatal death or brain injury (Zipursky A, et al 2011)

HOW DOES THIS HAPPEN?

- Exposure of RhD negative individuals to the RhD antigen on red blood cells (RBCs) is highly immunogenic

- Causes alloimmunization and future production of RhD antibodies (Anti-D)

WHEN DOES THIS OCCUR?

- Sensitizing events in RhD negative women can occur:
  - *#1 through transfusion of RhD positive blood*
  - **or**
  - ***#2 transplacental hemorrhage from an RhD positive fetus***

POTENTIAL SENSITIZING EVENTS IN RH D-NEGATIVE WOMEN IN PREGNANCY

- Chorionic villus sampling(14%), amniocentesis(2-6%), cordocentesis
- Threatened miscarriage or miscarriage
- Ectopic pregnancy
- Evacuation of molar pregnancy
- Therapeutic termination of pregnancy
- Antepartum hemorrhage
- Abdominal trauma
- Intrauterine fetal death
- External cephalic version(2 – 6%)
- Delivery

FETAL-MATERNAL HEMORRHAGE

- 3% -11% with threatened abortion 1st trimester
- ~ 45% giving birth in the third trimester
- may take place in the first and second trimesters in association with spontaneous pregnancy loss or uterine instrumentation (e.g. dilation and curettage or evacuation)

RISK OF RhD ALLOIMMUNIZATION

- 1.5–2% in susceptible women after spontaneous miscarriage
- 4–5% after dilation and curettage
- Volume needed **small as 0.1 mL or as large as 30 mL**
- Until further evidence is available, expert advice continues to recommend administration of anti-D immune globulin within 72 hours of suspected breach of the choriodecidual space

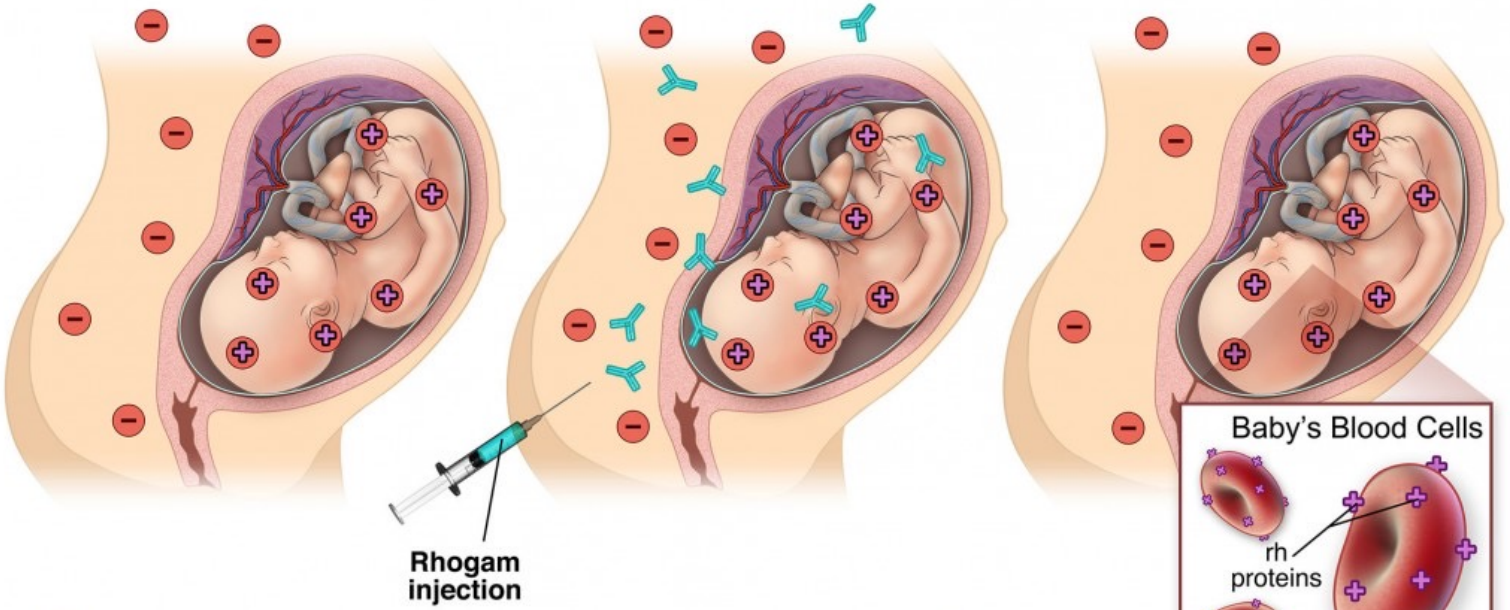
Rh Factor Sensitization Prevention

Mother's and Baby's Blood
Mother is Rh negative and has not previously been sensitized to Rh factor.

Rhesus Immune Globulin Injection is Administered To Prevent Antibody Formation

An intramuscular injection is given:
1. At 28 weeks into a pregnancy.
2. Within 72 hours of birth of a confirmed Rh positive baby.

Titers are Checked
Sensitivity to blood titers is checked:
1. 6 months after last Rhogam injection following birth of Rh positive baby to rule out false positive.
2. If titers remain positive after 6 months, sensitization has occured.



Rh blood titer = 0

Rh titer = 1:1 a week following injection.
Rh titer = 0 six months following shot if no sensitization has occured.

RhD IMMUNE GLOBULIN/ ANTI-D IMMUNE GLOBULIN

- Immune globulin used specifically to bind the Rh D antigen
- 5 commercial formulations available in U.S.:
  - HyperRho S/d Full Dose
  - MICRhoGAM Ultra-Filtered PLUS
  - RhoGAM Ultra-Filtered PLUS
  - Rhophylac
  - WinRho SDF
- *A prophylactic dose of 300 micrograms prevents Rh D alloimmunization after exposure to up to 30 mL of Rh D-positive fetal whole blood or 15 mL of fetal red blood cells*

UNITED STATES

- Recommendation for the administration of anti-D immune globulin was introduced in the 1970s
- Current practice:
  - single antenatal dose of 300 micrograms at 28 weeks of gestation
  - second dose after birth infant identified as Rh positive
  - Recommendations McMaster University Conference 1977
  - Associated with <0.2% rate of Rh alloimmunization

UNITED KINGDOM

- antenatal Rh D immune globulin using different doses may be given as two injections at 28 weeks of gestation and at 34 weeks of gestation
- or
- single administration at 28 weeks of gestation

FETAL MATERNAL HEMORRHAGE

- 2–3 per 1,000 deliveries > 30 mL
- Rh D-negative /Rh D-positive infant need additional testing
- Assess the volume of fetal–maternal hemorrhage
- Guide the amount of Rh D immune globulin required to prevent alloimmunization

SCREENING FOR FETAL–MATERNAL HEMORRHAGE

- Rosette fetal red blood cell assay
- Kleihauer–Betke test
- Flow cytometry

ROSETTE FETAL RED BLOOD CELL ASSAY

- Sensitive/qualitative test
- Detects >2 mL of fetal whole blood in the maternal circulation
- incubation maternal blood sample with Rh immunoglobulin that will bind fetal Rh D-positive red blood cells, followed by the addition of enzyme-treated reagent indicator red blood cells
- Rh D-positive fetal red blood cells present in maternal circulation result in forming aggregates (rosettes) seen with light microscopy
- A positive rosette test should be followed with a quantitative test

KLEIHAUER–BETKE TEST

- Acid elution test
- Relies on the principle fetal RBCs contain mostly fetal hemoglobin F, which is resistant to acid elution
- Adult hemoglobin is acid sensitive
- Inexpensive / requires no special equipment
- Lacks standardization / precision
- Not as accurate in maternal conditions RBCs inc hemoglobin F – sickle cell disease / thalassemia

FLOW CYTOMETRY

- Monoclonal antibodies to hemoglobin F or the Rh D antigen with quantification of fluorescence,
- Highly sensitive and accurate in identifying fetal red blood cells in maternal blood
- Limited by equipment and staffing costs

PREVENTION IN FETAL-MATERNAL HEMORRHAGE

- Additional vials of Rh immune globulin can be administered at one time (up to eight full vials)
- Can be administered IM at separate sites every 12 hours until the desired dosage has been reached
- An intravenous Rh immune globulin is available that also may be used in these cases and provides more comfort for the patient
- In the United States, Rhophylac and WinRho are available for IV administration

HOW DOES THIS HAPPEN?

- Sensitizing events in RhD negative women can occur:
  - ***#1 through transfusion of RhD positive blood***
  - *or*
  - *#2 transplacental hemorrhage from an RhD positive fetus*

TRANSFUSION OF RhD POSITIVE BLOOD

- RhD-mismatched transfusions in emergent /traumatic settings
- Probability of the development of anti-D antibodies in RhD negative patients who receive RhD positive blood products has been evaluated in multiple populations
- Alloimmunization risks:
  - 80% in healthy individuals
  - 15% in immunosuppressed transplant patients
  - 0% in those with Acquired Immunodeficiency Syndrome (AIDS)

TRANSFUSION OF RhD POSITIVE BLOOD

- 3 retrospective studies
- Average rate of RhD alloimmunization after receiving RhD positive packed red blood cells (pRBCs) is **20.5%** (72/351, 95% confidence interval (CI) 16.6%–25.1%)
- An additional study has reported an RhD alloimmunization rate of 1.4% following RhD positive platelet transfusions

TRAUMA TRANSFUSIONS

- Significant variation 8% - 44% reported
- Combine study results rate of alloimmunization in trauma patient receiving RhD-mismatched transfusions is around 27% (Ji et al 2022)

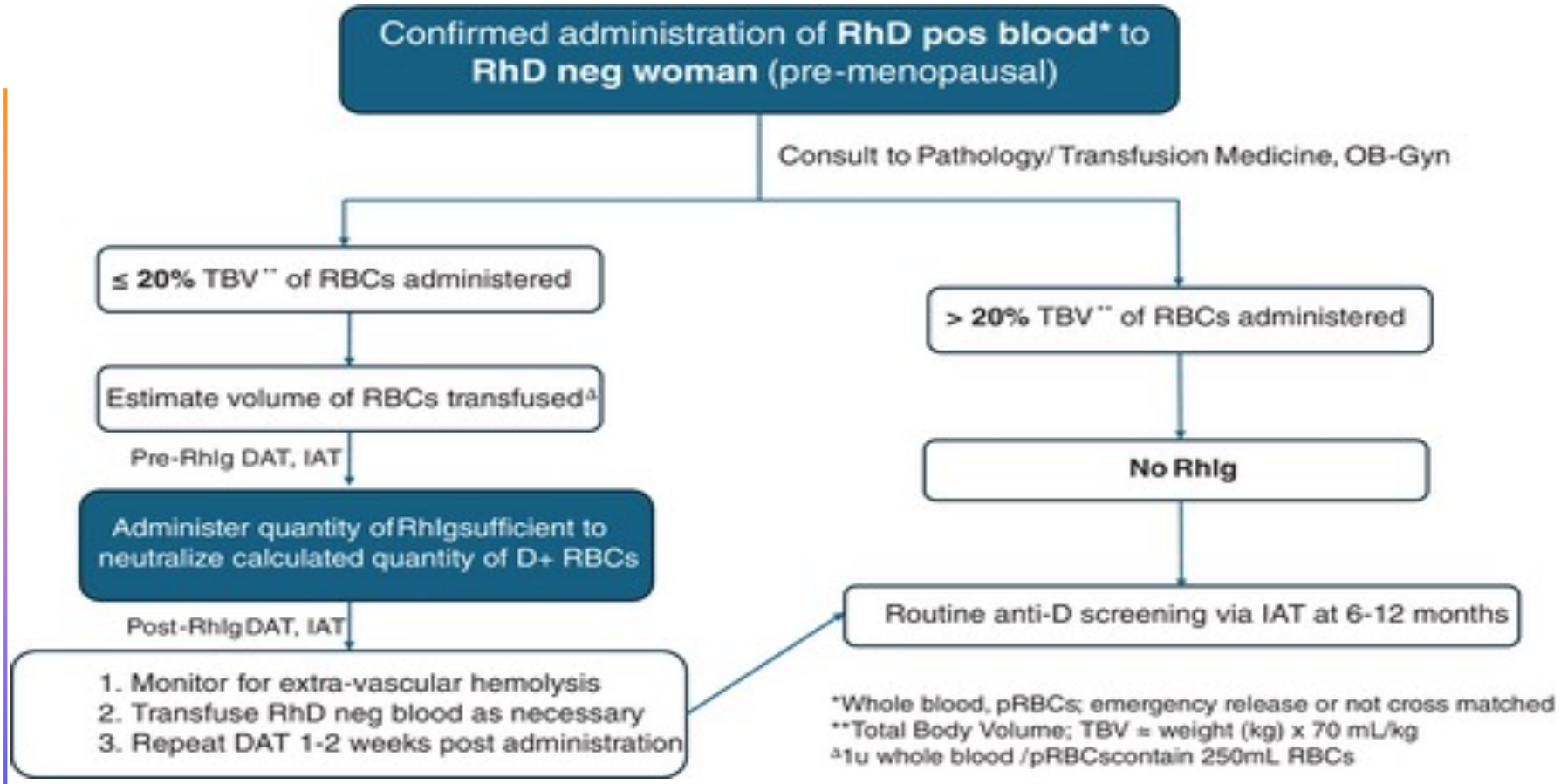
RhD IMMUNE GLOBULIN/

ANTI-D IMMUNE GLOBULIN IN SETTING OF MISMATCHED BLOOD

- Massive transfusion makes decision-making regarding the use of post-transfusion RhIG challenging
- Administration could lead to massive hemolysis of transfused RBCs and subsequent hemodynamic collapse
- Due to the inability to predict which patients will become alloimmunized, a management strategy for RhD negative women of childbearing potential who receive RhD-mismatched transfusions is needed to prevent potentially avoidable alloimmunization

RhD IMMUNE GLOBULIN/ ANTI-D IMMUNE GLOBULIN IN SETTING OF MISMATCHED BLOOD

- Rahaf Alkhateb et al University Hospital designed protocol
- March 2019 trauma protocol change
- Allow transfusion of LTO + WB in select trauma patients ≥10 years old, including females of childbearing age and children, both male and female
- Patients tx RBCv ≤20% of their TBV are eligible to receive RhIG
- RBCv >20% makes ineligible for prophylaxis with RhIG


Confirmed administration of RhD pos blood* to RhD neg woman (pre-menopausal)
Consult to Pathology/ Transfusion Medicine, OB-Gyn
≤ 20% TBV** of RBCs administered
> 20% TBV** of RBCs administered
Estimate volume of RBCs transfused△
Pre-RhIg DAT, IAT
No RhIg
Administer quantity of RhIg sufficient to neutralize calculated quantity of D+ RBCs
Routine anti-D screening via IAT at 6-12 months
Post-RhIg DAT, IAT
1. Monitor for extra-vascular hemolysis
2. Transfuse RhD neg blood as necessary
3. Repeat DAT 1-2 weeks post administration
*Whole blood, pRBCs; emergency release or not cross matched
**Total Body Volume; TBV = weight (kg) x 70 mL/kg
△1u whole blood /pRBCs contain 250mL RBCs

EXAMPLE CALCULATION

50-kg woman transfused 1 unit of LTO + WB with planned administration of IV Rhophylac:

1. 1 unit of LTO + WB contains approximately 250 mL of RBCs
2. 250 mL of RhD positive RBCs × 20 μg (100 IU) per mL = 5000 μg (25,000 IU)
3. 5000 μg RhIG/(300 μg RhIG per vial) ≈17 vials of RhIG

FAILURE TO PREVENT RH D ALLOIMMUNIZATION

- Rh alloimmunization Rh D-negative women may still occur
- Reasons:
  - failure of administering antenatal prophylaxis in the third trimester
  - insufficient dosage or timely administration (within 72 hours) of anti-D immune globulin given after a known sensitizing event during pregnancy (or after birth)
  - unrecognized fetal–maternal hemorrhage at some point in pregnancy
- In spite of recommendations for immunoprophylaxis, approximately 0.1–0.4% of women at risk become sensitized during pregnancy

SUMMARY

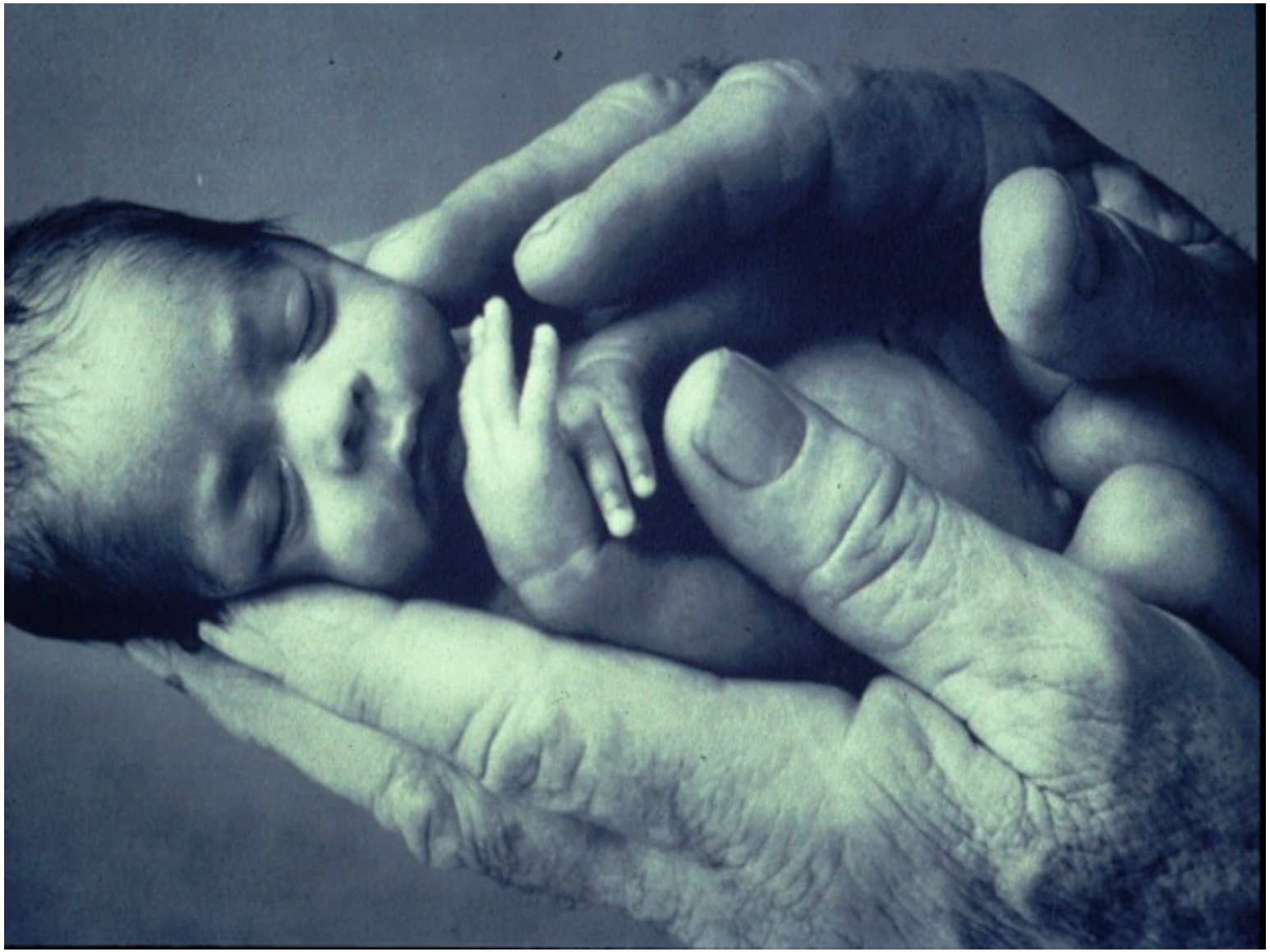
- Wide variation in prevalence rates of Rh D-negative individuals
- ***RhD alloimmunization refers to the maternal formation of antibodies against fetal Rh D***
- Advances in the prevention and treatment of Rh D alloimmunization have been one of the great success stories of modern obstetrics
- Reduction of alloimmunization rate from **13 – 16% to ~ 0.5 – 1.8%**
- Risk is further reduced to 0.14–0.2% with the addition of routine antepartum administration (28 weeks)

SUMMARY

- Sensitizing events in RhD negative women can occur:
  - *#1 through transfusion of RhD positive blood*
  - **Or *#2 transplacental hemorrhage from an RhD positive fetus***
  - ***Variety of inciting events including CVS, amnio procedures, miscarriage, D & C, version***
  - single antenatal dose of 300 micrograms at procedure, event, or 28 weeks of gestation second dose after birth infant identified as Rh positive
  - In mismatched blood tx setting Patients tx RBCv ≤20% of their TBV are eligible to receive RhIG

NATIONAL GUIDELINES

- United States, United Kingdom, and Canada
- Recommend routine administration of anti-D immune globulin to all Rh D-negative nonsensitized women in the third trimester, within 72 hours of delivery in women giving birth to a Rh-positive infant
- or when a sensitizing event occurs (eg, ectopic pregnancy, external cephalic version, or invasive obstetric procedures such as chorionic villus sampling or amniocentesis)



THANK YOU

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