



ALLOIMMUNIZATION SECONDARY TO PREGNANCY AND TRANSFUSION IN WOMEN OF CHILD BEARING AGE

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Objectives

Overall objective

To appreciate the effect of red cell alloimmunization in women of child bearing age

Alloimmunization from paternal exposure

Know the indications for RhIG to prevent D alloimmunization

Alloimmunization from red cell transfusion

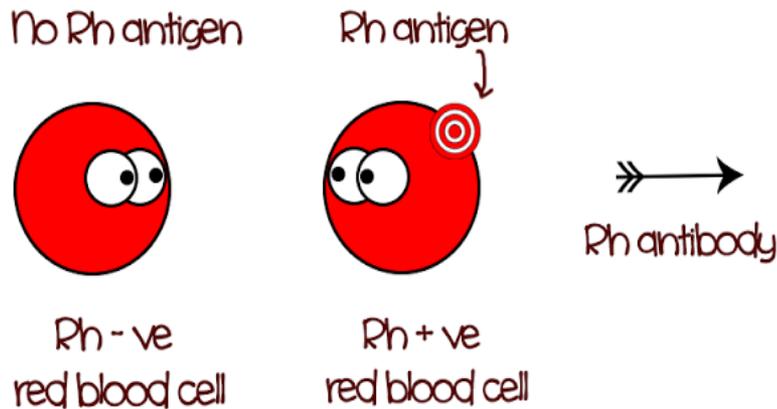
Appreciate the changes in hemoglobin levels during pregnancy, indications and alternatives for transfusion peripartum to prevent red cell alloimmunization

28 year old G1 at 22 weeks gestation, D negative has vaginal spotting.

You (assuming partner is D+)

1. Advise for RhIG
2. Advise for RhIG and testing for fetomaternal hemorrhage
3. Test for fetomaternal hemorrhage

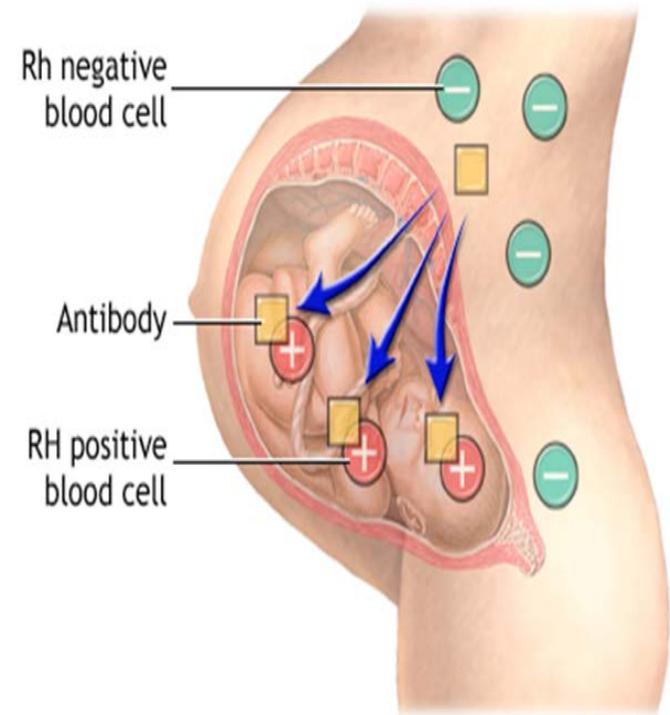
Alloimmunization (development of an alloantibody) occurs following



1. Exposure to paternal antigens by fetal maternal hemorrhage during pregnancy
2. Exposure by RBCs

IgG maternal alloantibodies traverse the placenta

Can bind to fetal antigens on red cells resulting in destruction of fetal/neonatal red cells=Hemolytic Disease of the Fetus and Newborn (HDFN)



ADAM.

The ability to develop an alloantibody depends on

The immunogenicity and embryonic development of the red cell antigen

The volume of red cell antigen exposure

The immune responsiveness of the individual

e.g. alloimmunization following exposure to paternal D antigens occurs in 16% of women

The ability to develop an alloantibody depends on the **red cell antigen**

Immunogenicity of the antigen

Polypeptide antigens give rise to IgG e.g. Rh vs. carbohydrate antigens tend to give rise to IgM-class antibodies (ABO), P, Lewis and I

Embryonic development of the antigen

D antigen appears as early as 38 days gestation vs. AB antigens postnatally

The ability to develop an alloantibody depends on the volume of **red cell antigen exposure**

Transplacental hemorrhage (transfer of fetal RBCs into the maternal circulation)

Is high in each of the trimesters

1st: 56% (0.07 ml)

2nd : 63% (0.08 ml)

Increases with invasive procedures, trauma, delivery

Transfusion

RhD: Primary immune response: 15% after 1 ml RBCs and 70–90% after 250 ml

Secondary immune response may occur after exposure to as little as 0.03 ml RBCs

The ability to develop an alloantibody depends on the **immune responsiveness of the individual**

Characteristics of the antibody

Ability to present the red cell antigen (e.g. HLA genotype)

Ability to lyse

Ability to suppress erythropoiesis

ABO incompatibility between mother and fetus, less lysis with ABO incompatibility

Spectrum of HDFN for those who develop an alloantibody



No disease

No fetal
destruction
of RBCs



Mild
disease

Hemolytic
disease of
the
newborn



Severe
disease

Hemolytic
disease of
the fetus
requiring
intrauterine
transfusion



Fetal
hydrops

Antigen	Severe Disease in Alloimmunized Pregnancies*
D	20%
K	11-50%
c	17-26%
Duffy	14-16% (Duffy a > b)
E	1%-18%

*Requiring intrauterine transfusion or neonatal exchange transfusion

Prevention of Alloimmunization

Paternal

Rh Immune globulin for exposure to D antigen in D negative mothers

Transfusion

Transfusion avoidance

Antigen negative transfusion e.g. D-, K-

Rh immune globulin can only be administered within

- A. 24 hours
- B. 72 hours
- C. 72 hours but up to 10 days

Rh immune globulin is a plasma derived IgG protective for exposure to D antigen

Pooled plasma product derived from donors with high anti-D antibodies

Risk of contracting viruses reduced by the manufacturing process

Mechanism

The antibody coated cells, are destroyed by the spleen before D is seen by antigen-presenting cells

The antibody alters D antigen processing reducing T cell and B cell activation

Prophylactic RhIG has reduced alloimmunization from 16% - ~ 0.2-1% in D negative women

300 micrograms suppresses the response of up to 15 ml of RBCs

300 micrograms to all D-negative women at 26-28 weeks' gestation, and within 72 hours of delivery (for fetuses that are D+ or D unknown)

Administer RhIG within 72 hours of a sensitizing event: may be given up to 10 days of the event

Prior to 12 weeks gestation (fetoplacental blood volume is 3ml)

120-300 μg

Between 12-20 weeks gestation

300 μg

More than 20 weeks

Minimum dose 300 μg

Rosette/Betke Kleihauer/Flow cytometry to determine presence and volume of a bleed

Postpartum

120-300 μg if the neonate is D+ and BK/flow cytometry to determine presence and volume of a bleed

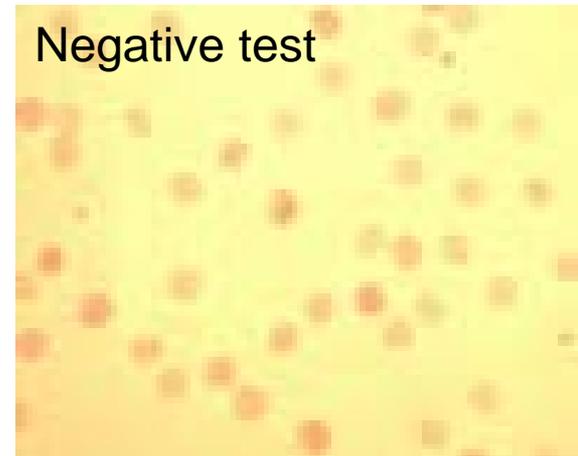
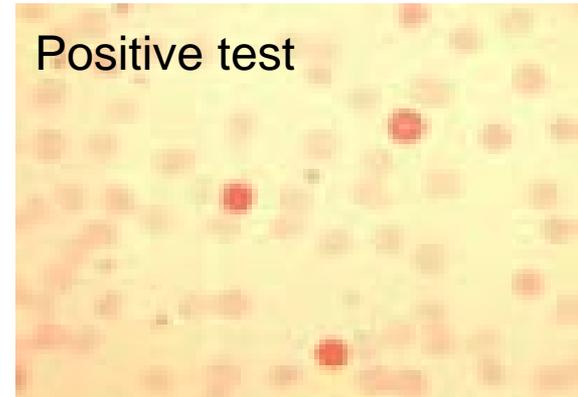
The Betke Kleihauer acid elution test detects Hemoglobin F

Fetus: Hemoglobin F is the predominant hemoglobin of the fetus, is more acid-resistant than other hemoglobins (fetal cells appear as dark reddish/pink cells)

Adult: Hemoglobin F usually < 1% in adults, concentrated in F cells

Predominant HbA is denatured by acid and eluted=ghost cells

$\text{ml bleed} = \% \text{ fetal cells } (\# \text{ fetal} / \# \text{ counted}) \times 5\text{L}$
(BV)



Flow cytometry has better accuracy: can differentiate fetal cells from adult F cells

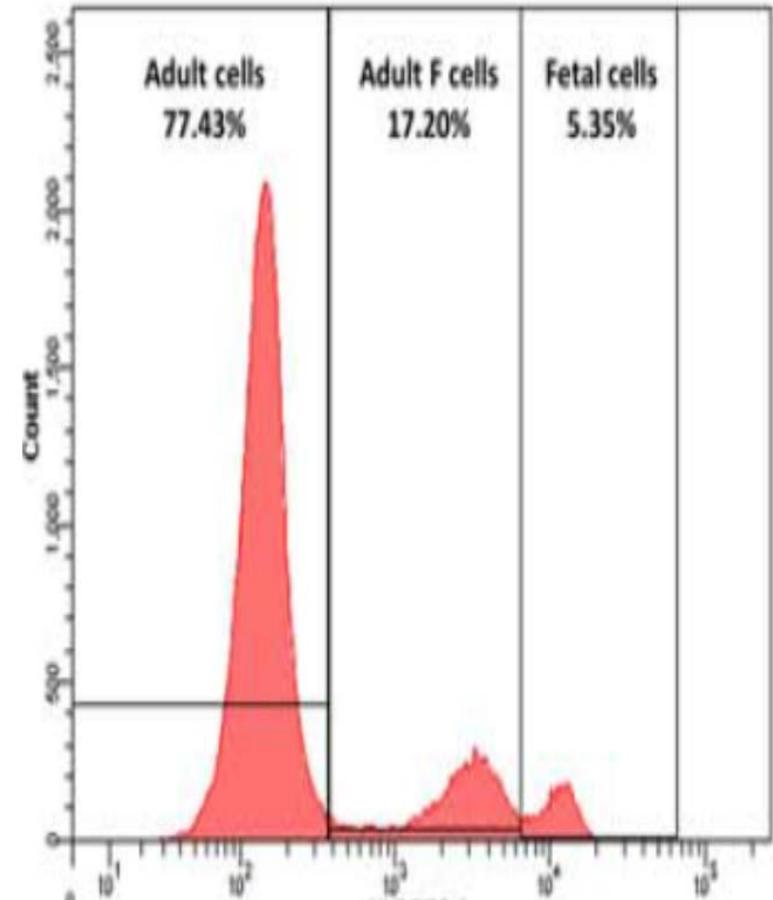
F cells contain 20-25% HbF in an adult

F cells increases during pregnancy

Can be as high as 7% by 32
wks

F cells increased with hemoglobin
disorders

Better accuracy in estimating volume of
FMH



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Appreciate the changes in hemoglobin levels during pregnancy, indications and alternatives for transfusion peripartum to prevent red cell alloimmunization

Would you transfuse RBCs?

25 year old female G1P0 at 27 weeks gestation with vaginal spotting, Hb 70 g/L

1. Yes
2. No

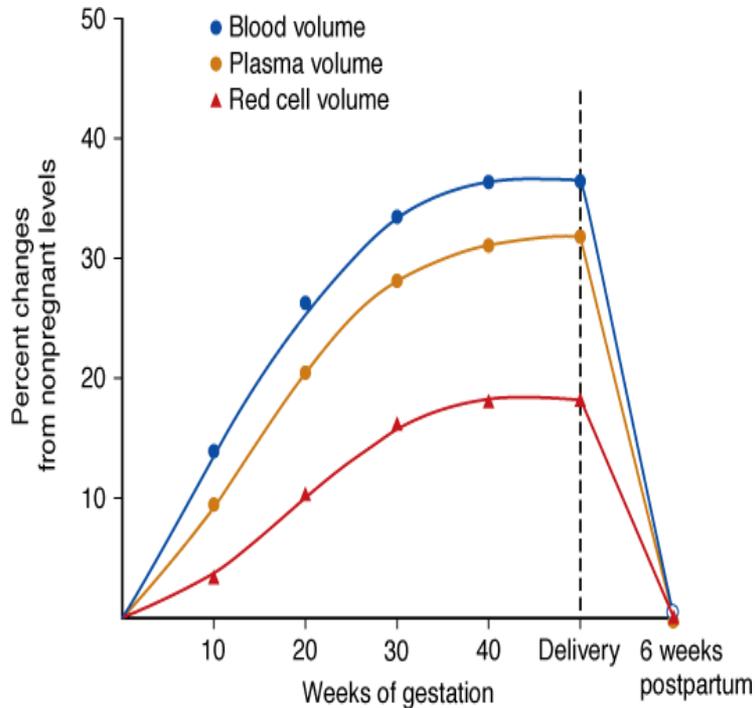
Are the benefits worth the risk of exposure to red cell antigens?

Is the mother compromised by the anemia?

Is the fetus compromised by maternal anemia?

Are there alternatives?

Physiological changes during pregnancy result in a lower Hb level



Blood volume increases by **1.5 L**

Plasma volume increases by **50%**

Red blood cell mass increases by **25%**

Lower hemoglobin levels occur

because plasma volume increases >

red cell volume

Anemia is defined as a hemoglobin level...

Non pregnant : <120 g/L

1st trimester : <110 g/L

2nd and 3rd trimester : < 105 g/L

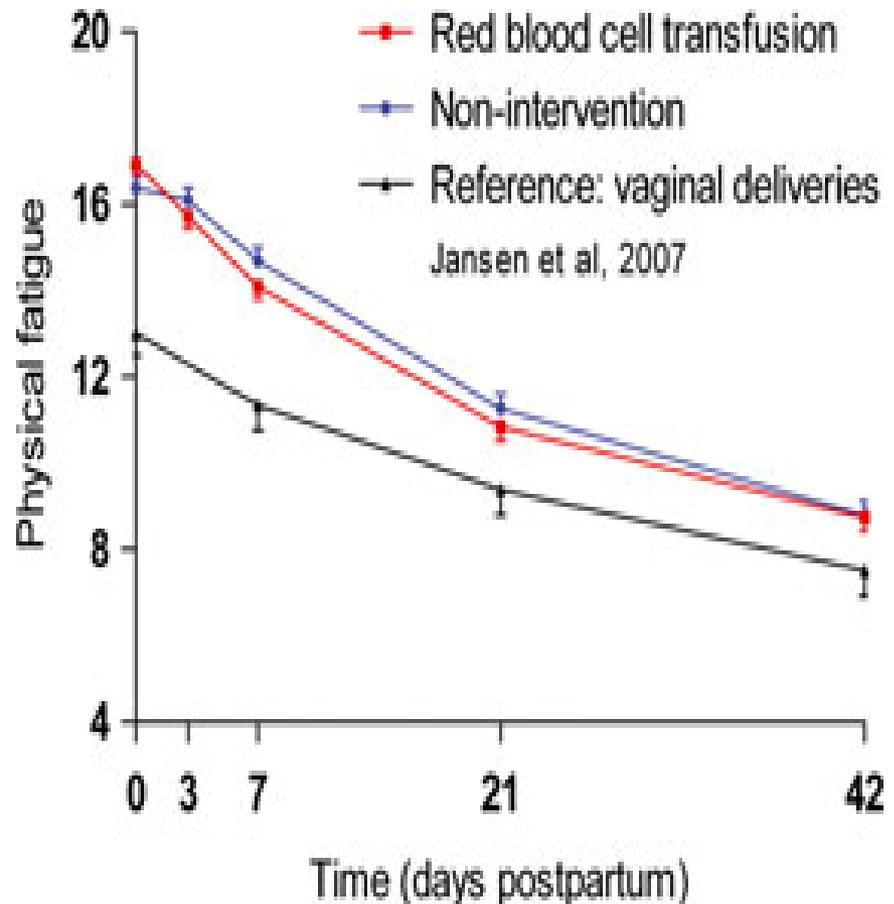
Postpartum : < 100 g/L

Postpartum: Women can tolerate a reduction to 50% of their hemoglobin concentration

Patients: Postpartum with Hb 48- 79 g/l 12–24 hours after delivery (mean age 31 yrs) and **hemodynamically stable**

Randomized to RBC vs no RBC

Fatigue was increased after 7 days but was the same after 42 days



The most common cause of anemia in pregnancy is iron deficiency: in 18%

Marked increase in iron demands in pregnancy

27-30 mg elemental iron in a prenatal vitamin/total iron requirement-1.2 g during the entire pregnancy

Higher frequency with iron deficiency in a prior pregnancy, multiparty, low SES, brief intervals between pregnancy, poor nutritional status, PPH

Diagnosed by ferritin <30 µg/L or fasting iron saturation <15-20%

(* low reticulocyte Hb, low hepcidin, increased soluble transferrin receptor)

Iron preparations

Preparation	Elemental iron (mg)
Oral formulations	
Prenatal vitamin (Ferrous fumarate)	27-35mg/tablet
Ferrous gluconate	35/tablet
Ferrous sulfate	65/tablet
Ferrous fumarate	100/tablet
Polysaccharide-iron Complex	150/capsule
Heme-iron polypeptide	11mg/tablet
Intravenous formulation	
Iron sucrose	20 mg/ml*

* The dose is calculated as (prepregnancy weight (kg) x [(target hemoglobin-actual hemoglobin g/L)/10]) x 2.4 +500. Maximum dose/infusion/day is 300 mg. Doses higher than 300mg need to be administered as divided doses.

Response to IV Iron faster than oral

Response to oral iron

Hb rise by 1 g/L per day after starting iron

Return to normal hemoglobin by 6-8 weeks

Iron stores replenished by 3-6 months

Response to iv iron is 2x as fast

Well tolerated, no hypersensitivity reactions
in 599 patients in a systematic review

Indications: moderate to severe anemia, no
response to oral, side effects to oral iron,
limited time to delivery

* Iv iron reserved for the 2nd and 3rd trimester

Hb levels	n (Intervention/ control)	IV iron sucrose Mean (SD)	Standard therapy Mean (SD)	Adjusted mean difference * 95% CI
Baseline	983/1015	7.7 (0.9)	7.7 (1.0)	-
Change from the baseline at 6wks post randomization	666/629	2.8 (1.3)	1.9 (1.7)	0.95 (0.80, 1.10)
Change from the baseline at delivery	254/221	2.8 (1.4)	2.4 (1.9)	0.45 (0.18, 0.73)
Change from the baseline at 6wks postpartum	697/ 626	3.7 (1.4)	3.0 (1.7)	0.63 (0.48, 0.78)
No anaemia (Hb \geq 11.0gm/dl) at 6wks post randomization	666/629	240 (36.0)	128 (20.3)	2.26 (1.75, 2.92),
No anaemia (Hb \geq 11.0gm/dl) at 6wks postpartum	697/ 626	469 (67.3)	286 (45.7)	2.48 (1.98, 3.10),

Complications because of iron deficiency anemia

Mother:

Fatigue

Anemia may result in increased transfusion rates

Iron supplementation results in a lower incidence of Hb<100 g/L at delivery vs placebo (7% vs. 16%; RR, 0.45 [CI, 0.25 to 0.82])

Fetus:

Potentially associated with adverse fetal outcomes

Obstetrical ultrasound detects fetal compromise by maternal anemia, Hb<60 was associated with fetal compromise

What is the role of erythropoiesis stimulating agents (e.g. erythropoietin)?

Most of the use is in patients with renal failure during pregnancy

It is a large molecule and it is not thought to cross the placenta

Royal College of Obstetrics and Gynecology advises against

The routine use of erythropoiesis stimulating agents

Exceptions

- Patients who do not have blood readily available

- Patients with renal failure

When should patients be transfused in the antepartum setting?

RCOG suggests “There are no firm criteria for initiating red cell transfusion. The decision to provide blood transfusion should be made on clinical and hematological grounds.” In the absence of hemorrhage or hemodynamic instability RBC transfusion is almost always required when the Hb is < 60 g/l and is rarely required when the Hb >100 g/l.”

ACOG suggests “Maternal transfusion should be considered for fetal indications” Hb <60 g/L has been associated with fetal compromise.

25 year old female G1P0 at 27 weeks gestation with vaginal spotting, Hb 70 g/L

She was transfused 1 U PRBC

Postpartum an antibody screen detected allo anti-c

If an alloantibody is detected...

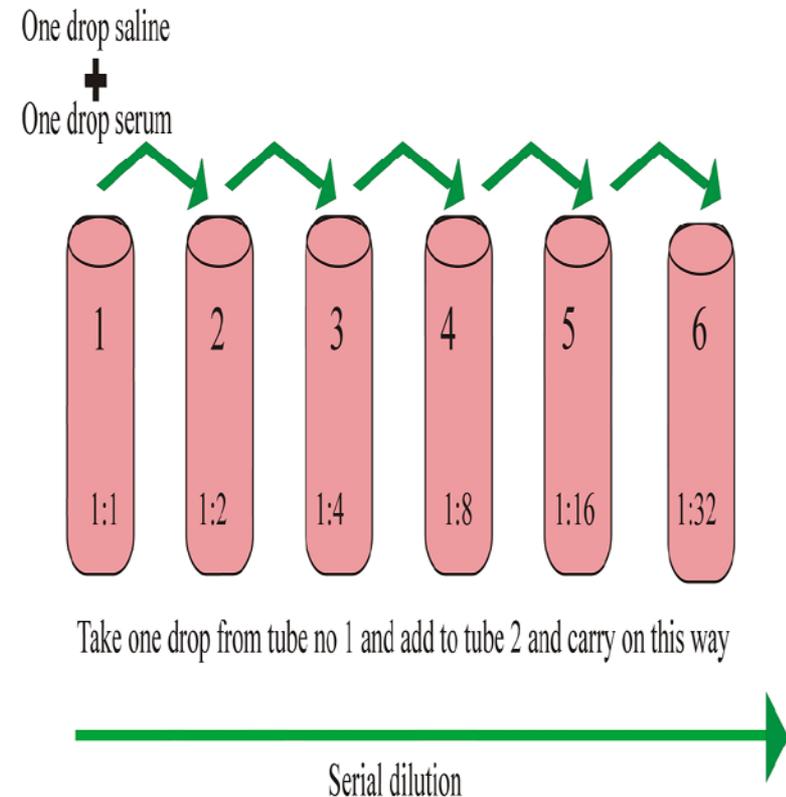
The father's phenotype for the antigen and zygosity

If the fetus is potentially affected

Serial antibody titres: a method to determine strength of the antibody

Antibody titre: greatest dilution with a positive agglutination reaction

cffDNA for D 18 weeks onward if partner is heterozygous



The alloantibody titre should be monitored

D, c

In a primagravida

Every 4 wks in the 1st and 2nd trimester or every 2 wks if there is a rise and in the 3rd trimester

Referral to high risk centers if

For a first pregnancy

When the alloantibody >1:16-32, D, c, E

At any alloantibody titre for K as titres may not correlate with the severity of anemia: K alloantibodies also suppresses fetal erythropoiesis

Subsequent alloimmunized pregnancy where the first had HDFN

Once pregnancy is confirmed

Summary

Overall objective

To appreciate the effect of red cell alloimmunization in women of child bearing age

Alloimmunization leads to risk of HDFN

Alloimmunization from paternal exposure

Know the indications for RhIG to prevent D alloimmunization

RhIG for any potential FMH within 72 hours

Alloimmunization from RBC transfusion

Appreciate the changes in hemoglobin levels during pregnancy, indications and alternatives for transfusion peripartum to prevent red cell alloimmunization

Transfuse if symptomatic mother or fetus appears to be compromised e.g. fetal tachycardia, K negative RBC if transfusion needed



QUESTIONS?
