ERYTHROBLASTOSIS FETALIS WITHOUT AN OBVIOUS CAUSE
Case Study by Jim Perkins, MD (©2010)

ANSWERS:

1. How might you explain this infant's positive DAT and apparent hemolytic disease?

   This infant appears to have all of the classic features of hemolytic disease of the fetus and newborn (HDFN) including anemia, icterus (an elevated bilirubin level), and erythroblastosis. The problem with this diagnosis is that the mother's blood group antibody detection test (antibody screen) is negative and mother and baby are both group O, so there is no apparent incompatibility to explain the positive DAT on the infant's RBCs. To explain this one must remember that the antibody screen will only detect antibodies directed against those antigens present on the screening cells. The screening cells, like most individual's RBCS, are not expected to express most low frequency antigens, some of which can mediate HDFN. So the mother could have an antibody directed against a low frequency antigen that the infant has inherited from his father yet still have a negative antibody screen.

2. What test should be performed next? Explain your answer.

   To investigate this possibility a crossmatch should be performed between the mother's plasma and the father's RBCs. If the infant expresses a low frequency antigen that the mother has an antibody against, he must have inherited it from the father who would express it as well.

3. What is your immunohematologic and clinical diagnosis?

   The positive crossmatch of mother's plasma with father's RBCs demonstrates blood group incompatibility. As mentioned above, the clinical presentation is classic, so all of the criteria for a diagnosis of HDFN are present.

4. What would you like to do now?

   It is of interest to identify the specificity of the mother's antibody. Note, however, that this is largely an academic concern; failure to identify the antibody would not change the diagnosis. In order to identify the antibody the mother's plasma can be tested against cells expressing low frequency antigens. Although maintenance of large libraries of such cells is generally restricted to reference laboratories, manufacturers of antibody identification panel cells typically test them for certain low frequency antigens such as C^w, V, Js^a, Kp^a, and Lu^a, and may list additional antigens as well. The inventory of such cells can be increased by saving outdated panels.

   It is also of interest to determine the titer of the antibody. Although the threshold titer at which significant HDFN occurs cannot be strictly generalized for different antibody specificities and different titration methods, one would not expect a very low titer to cause this degree of hemolysis.

5. What is the definitive immunohematologic diagnosis? Discuss the nature of the causative antibody and target antigen?

Testing with cells identified as bearing low frequency antigens allowed us to identify the mother as having anti-Wright^a (Wr^a). Strictly speaking, one would also like to show that the infant carried the Wr^a antigen, but we did not have an anti-Wr^a serum to demonstrate this. Nonetheless, it is implausible that the mother would have some other antibody that could cause this infant's problems.

The titer of 64 appears consistent with HDFN. Again note that strictly speaking one can't say that this is an anti-Wr^a titer since the father's cells were used as a target and these were not typed for Wr^a. One could be determining the titer of another antibody against a low frequency antigen on the father's cells, but again, that is implausible.

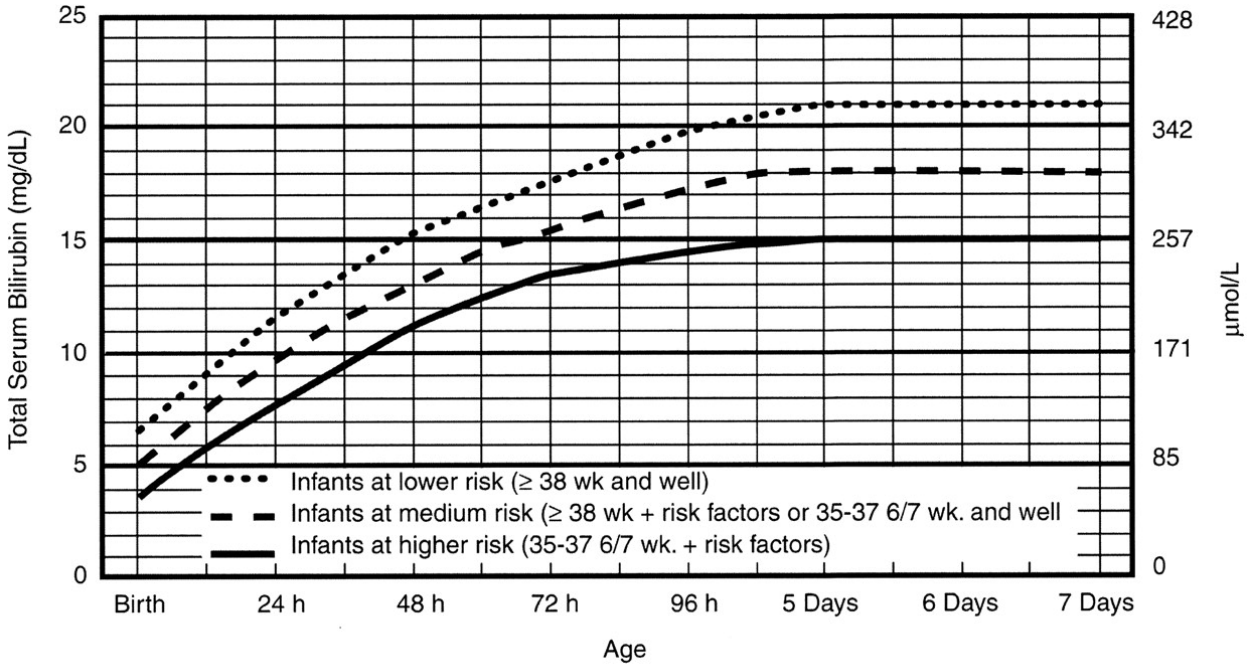
Wr^a belongs in the Diego blood group system of antigens expressed on the "band 3" protein. Band 3 is an important membrane constituent, making up 20% of the total RBC membrane protein. Like the Rh proteins it is a multipass protein, traversing the membrane 12 times. Band 3 functions as the major anion exchange protein, mediating the critical exchange of chloride and bicarbonate ion. It also binds to the red cell skeleton, tethering the lipid bilayer to it, and abnormalities of band 3 are associated with hereditary spherocytosis and other disorders of RBC morphology. The Wr^a/Wr^b dimorphism is caused by a single amino acid substitution. Wr^a is expressed on the RBCs of 1 in 1000 to 10,000 individuals in various European populations that have been studied.

Anti-Wr^a is present in the serum of 1 to 2% of blood donors and higher frequencies in patient studies, so in many instances it must be formed in response to some stimulus other than Wr^a on RBCs. It was first described in a case of HDN and is associated with both immediate and delayed hemolytic reactions (see "IHTR in a Patient with a Negative Antibody Screen" in the transfusion reaction cases). The antibodies may be IgG and/or IgM but do not activate complement.

6. What are the principles of treatment for this neonate?

The first concern is to control the bilirubin level in order to prevent kernicterus. This infant's bilirubin level of 9.9g/dL at birth was well above the level of 5g/dL at which phototherapy is indicated for a newborn with HDFN, and it was started immediately. The American Academy of Pediatrics (AAP) guideline for bilirubin levels which should prompt phototherapy are shown in the following figure.

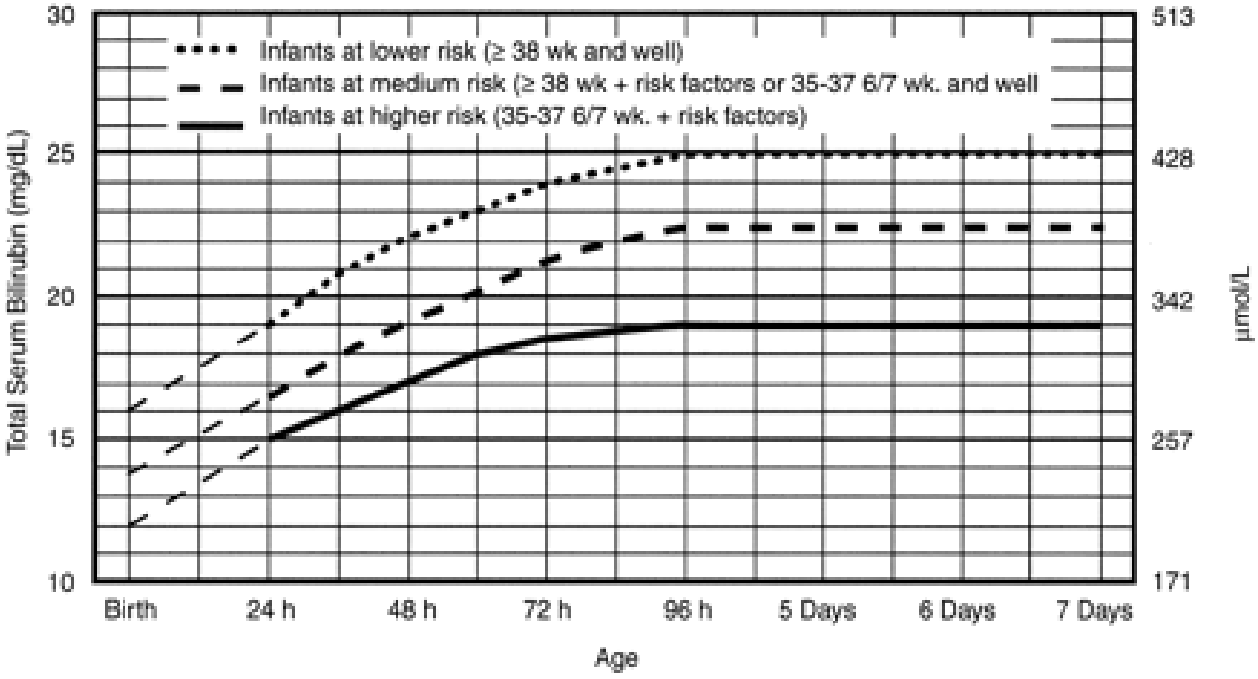
ERYTHROBLASTOSIS FETALIS WITHOUT AN OBVIOUS CAUSE: pg 3



- Use total bilirubin. Do not subtract direct reacting or conjugated bilirubin.
- Risk factors = isoimmune hemolytic disease, G6PD deficiency, asphyxia, significant lethargy, temperature instability, sepsis, acidosis, or albumin < 3.0g/dL (if measured)
- For well infants 35-37 6/7 wk can adjust TSB levels for intervention around the medium risk line. It is an option to intervene at lower TSB levels for infants closer to 35 wks and at higher TSB levels for those closer to 37 6/7 wk.
- It is an option to provide conventional phototherapy in hospital or at home at TSB levels 2-3 mg/dL (35-50mmol/L) below those shown but home phototherapy should not be used in any infant with risk factors.

The high bilirubin level coupled with significant anemia at birth prompted the laboratory to prepare for possible exchange transfusion. Close surveillance of the bilirubin was initiated, but a level prompting exchange was never reached. The AAP guidelines for bilirubin levels which should prompt exchange transfusion are shown in the second figure.

ERYTHROBLASTOSIS FETALIS WITHOUT AN OBVIOUS CAUSE: pg 4



- The dashed lines for the first 24 hours indicate uncertainty due to a wide range of clinical circumstances and a range of responses to phototherapy.
- Immediate exchange transfusion is recommended if infant shows signs of acute bilirubin encephalopathy (hypertonia, arching, retrocollis, opisthotonos, fever, high pitched cry) or if TSB is ≥5 mg/dL (85 µmol/L) above these lines.
- Risk factors - isoimmune hemolytic disease, G6PD deficiency, asphyxia, significant lethargy, temperature instability, sepsis, acidosis.
- Measure serum albumin and calculate B/A ratio (See legend)
- Use total bilirubin. Do not subtract direct reacting or conjugated bilirubin
- If infant is well and 35-37 6/7 wk (median risk) can individualize TSB levels for exchange based on actual gestational age.

A second concern regards anemia, and twice-a-day blood counts were ordered. The patient's hematocrit fell to 17 gm/dL on the 6th day of life, and the patient was transfused two aliquots of RBCs from a single donor on the 6th and 7th days. Thereafter the bilirubin fell and the infant was discharged.