Hemolytic disease of the fetus and newborn (HDFN) occurs due to the presence of red blood cell (RBC) alloantibodies in the maternal plasma during pregnancy. Those antibodies cross the placental barrier and enter the fetal bloodstream, bind to erythrocyte antigens, and destroy fetal erythrocytes. Immunoglobulins G (IgG) is actively transported across the placenta and directed against fetal RBCs antigens inherited from the father. Passive blood group antibodies from the mother can continue to affect neonatal red cells after delivery, causing ongoing anemia until the antibody is no longer present, which can be weeks to months after birth. Maternal alloimmunization resulted from exposure to foreign RBCs. It occurs through previous or current pregnancy, previous transfusions, or through an organ transplant. In fetomaternal hemorrhage (FMH), there is spontaneous mixing between fetal and maternal blood circulation. The mixing occurs throughout the pregnancy and increases by 3%, 12%, and 45% in the first, second, and third trimesters, respectively. HDFN due to RBC alloantibodies, especially minor blood groups, rarely occurs in the first stage of pregnancy because the risk of FMH is usually at the later stages, especially during delivery. These antibodies tend to develop after delivery.

**CASE REPORT**

An infant girl was born to a 25-year-old woman at 39 weeks gestation. The baby weighed 2.5 kg and had an Apgar score of 9/10. The baby was noted to have jaundice on day one with a serum bilirubin level of 290 mmol/L. There was a drop in hemoglobin within one day from 20.3 g/dL to 17.0 g/dL with a high reticulocyte count (9.3%) recorded. There was no other cause to suggest neonatal jaundice, such as intrauterine infections and glucose-6-phosphate dehydrogenase deficiency. An urgent peripheral blood film was sent and showed hemolysis with numerous spherocytes and the presence of nucleated RBCs and polychromasia. The baby’s blood group was Rh negative (RhD) positive. Direct Coombs test was positive with IgG specificity. Red cell elution studies...
of infant blood identified the presence of anti-E and anti-Jk^a antibodies. Her red cells phenotyping showed DCEce(R1R2) Jk^a+Jk^b-, which was similar to the father.

Antenatally, the mother had a threatened miscarriage at 13 weeks. She was discharged well without any complications or requiring any blood transfusion. Her blood group was B Rh-positive, and her antibody screening at that time was negative. The pregnancy progressed well without any complications. Post-delivery, her hemoglobin level was 12.0 g/dL with positive antibody screening. In the postnatal period, antibody identification was done and alloantibodies to Jk^a and E antigen were found. The mother’s RBC phenotyping was Jk^a-Jk^b+ and E-e+. Maternal anti-E and the anti-Jk^a antibody titer were determined as 1:512 and 1:32, respectively.

Intravenous immunoglobulin and intensive phototherapy were started for the baby since bilirubin levels were increasing. Simultaneously, we requested fresh whole blood with both antigens negative (E and Jk^a) in anticipation for a possible need for an exchange transfusion from the blood bank. The possibilities for anti E or anti-Jk^a to cause severe jaundice is rare, but based on one case report, the chances to develop severe HDFN cannot be ignored. HDFN due to anti-Jk^a is rare and can cause persistent anemia in the infant. Fortunately, the baby’s two-hourly serum bilirubin level showed a decreasing trend, and she did not require exchange transfusion and was discharged well on day nine. The daily blood investigations are shown in Table 1.

**DISCUSSION**

This case illustrates an uncommon example of HDFN caused by anti-E and anti-Jk^a alloantibodies. The antibodies were identified from the red cell eluate of the baby and the mother’s serum postnatally. In this case, there was no antibody detected antenatally before 13 weeks gestation, and there was no subsequent follow-up for antibody detection during the antenatal period. However, on day one, the baby developed significant jaundice, and an investigation for HDFN demonstrated that both anti-E and anti-Jk^a were present in baby’s red cell eluate and the mother’s serum.

As reported in the literature, the majority of causes for HDFN are due to ABO and Rh incompatibility. However, other minor blood groups have been identified as a cause for HDFN, although the occurrence is rare. A local study among 5163 Malay pregnant mothers reported 51 (0.99%) pregnant women had RBC alloantibodies, and among them, 0.9% were primigravida mothers. Most (66.7%) of the subjects had single alloantibodies, whereas 25.5% had multiple alloantibodies. The most common single alloantibody was anti-E, whereas anti-Le^a and anti-Le^b were the commonest multiple alloantibodies.

In the Rh blood group system, clinically significant alloantibodies other than anti-D such as anti-E and anti-c occur in 1:300 pregnancies, and the risk of HDFN caused by these antibodies is 1:500. Compared to anti-D alloimmunization, anti-E alloimmunization is a less common cause of hemolytic diseases. Its clinical manifestation is associated with more variability and usually less severity as it is a less potent immunogen. There is no obvious agreement regarding critical antibody titer for monitoring the fetus. A study by Joy and colleagues identified a titer of 1:32 or greater as the critical titer of anti-E, while Moran and colleagues observed a poor correlation between maternal alloantibody anti-E titers and severity of HDFN. Anti-E titer is less sensitive in detecting the severity of hemolysis in the subsequent pregnancy. A high

**Table 1: Serial hemoglobin level, reticulocyte count, and liver function test.**

| Laboratory test | Day 1 | Day 2 | Day 3 | Day 5 | Day 6 | Day 7 |
|-----------------|------|------|------|------|------|------|
| Full blood count |      |      |      |      |      |      |
| Hemoglobin, g/dL | 20.3 | 17.7 | 16.1 | 12.4 | 11.9 | 12.6 |
| Reticulocyte, %  | 9.2  | 9.4  | 9.9  | 10.7 | 9.1  | 6.6  |
| Liver function test |      |      |      |      |      |      |
| Total bilirubin, µmol/L | 290 | 294 | 280 | 289 | 216 | 163 |
| Indirect bilirubin, µmol/L | 279 | 281 | 270 | 278 | 205 | 152 |
| Direct bilirubin, µmol/L | 11  | 13  | 10  | 11  | 11  | 11   |
level of suspicion and early recognition of these cases is crucial even with low titters.\textsuperscript{11}

Anti-Jk\textsuperscript{a} was first identified by Allen et al.,\textsuperscript{12} in 1951 during serological testing of an infant diagnosed with HDFN. Jk\textsuperscript{a} antigens are well developed in neonates and can be detected on fetal RBCs as early as 11 weeks. However, only rarely are responsible for severe HDFN, possibly because of its poor immunogenicity.\textsuperscript{13} Prevalence among Asians is even rarer due to the low Jk\textsuperscript{a} allelic frequency.\textsuperscript{14} HDFN associated with anti-Jk\textsuperscript{a} is usually mild. However, Matson et al.,\textsuperscript{15} reported a case of severe HDFN associated with maternal alloantibody anti-Jk\textsuperscript{a}, in whom the infant developed kernicterus and received exchange transfusion. Mittal et al.,\textsuperscript{6} reported a case of moderate HDFN associated with maternal alloantibody anti-Jk\textsuperscript{a} with a titer of 1:64, whereby the infant received intensive phototherapy but escaped exchange transfusion. Previous studies reported that low hemoglobin in infants was associated with the persistence of Jk\textsuperscript{a} antibody coating the red cells for up to seven weeks of life.\textsuperscript{6} Hence, close monitoring of hemoglobin levels in affected infants is warranted for the first few months of life, and due to the presence of multiple maternal alloantibodies, in this case, the risk of developing significant HDFN is prominent.\textsuperscript{10}

The prevalence of E antigen negative red cells in our population is about 80%, but the prevalence of Jk\textsuperscript{a} antigen negative red cells is low, estimated at around 21%.\textsuperscript{16} Hence, to get one unit of both E and Jk\textsuperscript{a} antigen negative red cells is challenging, especially in the Malaysian population, and in emergency situations like this case.

**CONCLUSION**

HDFN caused by a combination of anti-E and anti-Jk\textsuperscript{a} may be moderate in its presentation but highlights the necessity of performing antibody screening for pregnant women. It is recommended at least once during their antenatal follow-up. This is to look for significant alloantibodies other than anti-D. Those mothers found to be alloimmunized should be monitored closely for the measurement of maternal antibody titer and to monitor the fetal wellbeing antenatally. In addition, predicting the management of HDFN in a newborn with positive maternal antibodies should be planned before the baby is delivered.

**Disclosure**
The authors declared no conflicts of interest.