This article has been accepted for publication in BMJ Case Reports, 2019 following peer review, and the Version of Record can be accessed online at http://dx.doi.org/10.1136/bcr-2019-229816
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Case report

Haemolytic disease of the fetus and newborn diagnosed after delivery of a baby to a mother with low anti-E antibody titres

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SUMMARY
The authors report a term male neonate who was born in unexpectedly poor condition with low Apgar scores and low venous cord gas pH. He required admission to the neonatal unit and was found to have developed haemolytic anaemia with associated hydrops, following a presumed severe antenatal insult. Antenatally, low levels of anti-E antibodies (titre 8) had been detected at 28 weeks' gestation. Following the British Society for Haematology and local neonatal team guidance, advice was given for cord direct antiglobulin test, full blood count and bilirubin at delivery. This case highlights the rare case of haemolytic disease of the fetus and newborn on a background of maternal low titre anti-E antibodies.

BACKGROUND
This is a report of a neonate who was unexpectedlyfound to have developed haemolytic anaemia with significant hydrops, likely secondary to a severe antenatal insult, requiring admission to the neonatal unit. Antenatally, the presence of low levels of anti-E antibodies was detected at the 28-week maternal bloods. E antigen belongs to the Rhesus (Rh) blood group system, and after anti-D, anti-c and anti-K, anti-E can also commonly causes haemolytic disease of the fetus and newborn (HDFN). However, from the current literature, in the absence of a history of
HDFN, the severity of haemolysis and thus likelihood of developing hydrops appears low. Yet this case shows that hydrops with fetal compromise can develop in the presence of lower anti-E titres.
The current guidance from the Royal College of Obstetricians and Gynaecologists and the British Society for Haematology is that

> "clinically significant antibodies, other than anti-D, -c or -K, should be excluded or, if present, assessed by titration at the booking appointment and at 28weeks' gestation. If deemed necessary based on a high titre (>32) and/or a past history of HDFN, referral to a specialist in fetal medicine should be made for further assessment".

CASE PRESENTATION
The patient was the second child of a 26-year-old woman. The mother had one previous pregnancy which was uncomplicated antenatally resulting in a normal delivery of a 3.09 kg live baby at term. In the present pregnancy, she booked at 8 weeks' gestation. She had normal dating and fetal anomaly scans. There was no maternal history of epilepsy or seizures during the pregnancy and no history of drug misuse. She had a further growth scan at 28 weeks' gestation because the symphyseal fundal height plotted <10th centile on a customised growth chart. This growth scan was normal. The 28-week blood tests detected a low titre of anti-E antibody of 8. The recommendation from the blood transfusion laboratory was for a cord direct antiglobulin test (DAT) at delivery and no further action. A neonatal alert was sent which further advised a full blood count and bilirubin in addition to cord blood DAT at delivery. She had two further contacts antenatally for a single episode of reduced fetal movements and thrush at 36+1/40 and 38+1/40, respectively.

At 39+1 weeks' gestation, she presented to obstetric care with contractions and was found to be 2–3 cm dilated. She progressed well and delivered a live male infant 14 hours later. Despite being born in apparently good condition initially, the baby collapsed at 1 min of life and required resuscitation including ventilation breaths and facial continuous positive airway pressure (CPAP). Apgar scores were 0 at birth, 4 at 5 min, and 8 at 10 min. A single cord gas was obtained with pH 7.123, base excess of −14.2, lactate of 12.1 mmol/L and haemoglobin of 41 g/L. Cord DAT was IgG 4+ positive with a raised total bilirubin (134 µmol/L) and a Kleihauer test <4 mL, leading to a diagnosis of HDFN.

Birth weight was 2.5 kg (second centile). The baby had signs of moderate hypoxic-ischaemic encephalopathy and was admitted to the neonatal unit for whole body hypothermia (33.0°C–34.0°C), CPAP and inotropic support. On examination he had mild ascites with body wall oedema and hepatomegaly.

INVESTIGATIONS

Admission haemoglobin confirmed congenital anaemia (haemoglobin of 64 g/L). Other admission blood test results showed abnormal coagulation (prothrombin time 20.2 s, fibrinogen 1.2 g/L), severe thrombocytopaenia (platelet count $33x10^9$/L), mildly abnormal liver function tests (alanine transaminase (ALT) 103 U/L; gamma-glutamyltransferase (GGT) 67 U/L; albumin 21 g/L), total bilirubin of 134 µmol/L, conjugated bilirubin of 48µmol/L and acute renal failure. On ophthalmological examination the baby was also found to have bilateral posterior embryotoxon, and echocardiography showed branch pulmonary arterial stenosis. A magnetic resonance brain scan done on day 14 showed a structurally normal brain and no features to suggest any hypoxic-ischaemic brain injury.

TREATMENT

To treat the anaemia and high unconjugated jaundice, he received exchange blood transfusion, for which he was electively intubated, and phototherapy. He received intramuscular vitamin K and also received fresh frozen plasma to treat the prolonged prothrombin time. Metabolic acidosis was corrected by bicarbonate and he was administered intravenous glucose to maintain normoglyceamia prior to introduction of enteral feeds on day 4.

Because of the early and persistent conjugated hyperbilirubinaemia and hepatosplenomegaly, advice was sought from a tertiary paediatric hepatology centre, which recommended ursodeoxycholic acid, vitamin E and vitamin K supplementation. He received human albumin solution with fluid restriction, to which his renal function, oedema and fluid balance responded and improved.

Phenobarbital was started on day 1 due to clinical and electrical seizures. Initial rewarming was attempted after 72 hours; however, due to increased seizures on rewarming, he was cooled for a further 12 hours. Levetiracetam and midazolam were added for persisting seizures. After day 10, midazolam infusion was weaned without recurrence of seizures. Dobutamine was stopped on day 5, and the baby remained haemodynamically stable. He was extubated on day 22 and was self-ventilating in air with no supplementary oxygen requirement by day 31.

OUTCOME AND FOLLOW-UP

On review at 5 months of age, he was thriving with accelerating growth and had no neurological sequelae. He was successfully weaned off levetiracetam and has remained seizure-free with a recent normal repeat electroencephalogram (EEG). He was meeting developmental milestones and there were no concerns with vision or hearing. He has ongoing cardiology and hepatology follow-up but has been discharged from ophthalmology. In view of the cardiac and ophthalmological findings, he was investigated for Alagille syndrome, but genetic analysis was normal; thus, this diagnosis was excluded.

Furthermore, in view of the unexplained neonatal jaundice, a diagnosis of possible familial haemophagocytic lymphohistiocytosis was also considered. His XLP2 expression was comparable with a control sample, suggesting that he does not have XLP2. However granule release assay showed absent cytotoxic granule release on two occasions, initially suggesting a possible defect in a granule release pathway. However subsequent analysis showed no pathogenic mutations present in the genes related to FHL3-5, GS2 and XLP2, and so it was presumed that the initial abnormal cytotoxic granule release tests had simply reflected his initial newborn state.

DISCUSSION

This baby was born in an unexpectedly poor condition with moderate neonatal encephalopathy related to perinatal compromise. After review of the case and subsequent investigation, the most likely cause for this was fetal compromise due to mild asphyxiation associated with fetal anaemia, which was felt to be due to chronic haemolysis secondary to anti-E-related HDFN.

Anti-E is reported to be the most common non-Rh-D antibody cause of HDFN with a prevalence of 14%–30%.[1–4] Case reports and series in current literature report varying severity of HDFN; however,

severe cases appear to occur in the context of isolated maternal titres of anti-E >1:16.[5–8] For example Joy *et al* [5] reported a series of 32 pregnancies at risk of HDFN from anti-E of which a maternal titre of 1:32 or greater predicted all anaemic fetuses. In contrast, however, Moran *et al* [6] reported 122 pregnancies over a 29-year period in which anti-E was identified as the only maternal autoantibody. Of these, 62 pregnancies were affected by HDFN of varying severity (Walker classification 1971), and titres ranged from 1/1, that is, where anti-E was only detected using enzyme techniques, to higher titres up to 1/1600. Of interest, the single very severe case was associated with a maternal titre of 1/1.

Moran *et al* [6] recommended that regardless of maternal anti-E titre, a direct antihuman globulin test and haemoglobin measurement should be performed on cord blood, unless the fetus is known to be Rh-E-negative, which was the action taken in this case.

Following detection of maternal anti-E antibodies, identification of Rh-E status of the fetus would help further stratify which pregnancies are at risk. This, like the current practice for antenatal diagnosis of Rh-D status (for Rh-D-negative mothers), can be performed with validated cell-free fetal DNA tests of maternal blood, which as well as being a non-invasive method of testing Rh-E status would also avoid potential discrepancies in reported paternity as would be the case with paternal phenotyping.[9 10]

The current paucity of literature suggests this is a relatively rare case; however, given the severity of the haemolysis and significant neonatal adverse resultant effects associated with this earlier low maternal anti-E titre, we suggest that more intensive fetal monitoring may be warranted in cases where anti-E antibody is detected, irrespective of titre.

LEARNING POINTS

- Adherence to current screening guidelines for the management of women with anti-E antibodies did not detect a baby, who then suffered moderate neonatal encephalopathy requiring therapeutic hypothermia and haemolytic disease of the newborn requiring exchange transfusion.
- Expectant parents should be reminded of the nature of *screening* tests, including that there are always some false positives and negatives.
- Consideration should be given when revising the guidelines for antenatal screening for abnormal antibodies as to whether the evidence in this area supports a change.

Contributors: TTML and PC co-wrote the main case report, with oversight, editing and review from EP-S, who initiated the writing of the case report following review of the case locally.
Funding: The authors have not declared a specific grant for this research from anyfunding agency in the public, commercial or not-for-profit sectors.
Competing interests: None declared.
Patient consent for publication: Parental/guardian consent obtained.
Provenance and peer review: Not commissioned; externally peer reviewed.

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