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CASE REPORT

Congenital amegakaryocytic thrombocytopenia presenting with a new thrombopoietin receptor (MPL) pathogenic variant: An instructive neonatal case

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Case Report

A 3-week-old male patient, child of healthy non-consanguineous Caribbean parents, presented at the hospital because of generalised convulsions. Mother was G6P4, the other children were healthy. He was born at 37 + 1 weeks gestation through vaginal delivery. Apgar scores were 9 at 1 and 5 min and birthweight was 2875 g (p40). Antenatal echographic investigations were normal and the pregnancy was uneventful. Family history revealed an unspecified coagulation disorder in the paternal grandfather.

The morning before presentation, a minor accidental head trauma had occurred and as the child initially did not show any symptoms, the parents did not seek any medical attention. However, after a few hours the patient started having generalised convulsions. At time of presentation at the hospital, he still had right-sided convulsions. Further physical examination revealed no abnormalities, especially no bleeding signs.

Key Points

1 Thrombocytopenia should be considered in neonates with clear clinical signs of mucocutaneous bleeding, but also in atypical presentations (e.g. seizures due to intracerebral haemorrhages). It should not be overlooked by clinicians.
2 If plasma thrombopoietin is elevated in thrombocytopenia, genetic testing for *MPL*-gene variants and other genes associated with thrombocytopenia can be a useful diagnostic tool.
3 Congenital amegakaryocytic thrombocytopenia is a condition that typically presents with severe thrombocytopenia and always progresses into aplastic anaemia. Consequently, haematopoietic stem cell therapy is needed as a definitive cure.

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Blood test showed a decreased haemoglobin of 8.8 g/dL (range 15.0–22.0 g/dL), low reticulocyte count 2‰ (range 6–30‰) with severe thrombocytopenia (platelets 10.0 $\times 10^9$/L (range 150–400 $\times 10^9$/L)), and normal leukocyte count 12.0 $\times 10^9$/L (range 6.0–18.0 $\times 10^9$/L), differentiation without blasts. The mean corpuscular volume was normal and mean platelet volume was 9.4 fL (range 8.0–10.6 fL). Glucose level and electrolytes were within normal ranges, as were prothrombin time and activated partial thromboplastin time. The fibrinogen level was 3.2 g/L (range 1.5–4.0 g/L). Computed tomography scan of the brain revealed bilateral subarachnoid and intraventricular bleeding. Furthermore, a large porencephalic cyst on the right side was shown. These findings were confirmed on MRI scan with contrast. This cyst had a hemosiderin lining, suggestive for antenatal haemorrhage (Fig. 1).

Antibiotics were given for 48 h until blood cultures remained sterile. Almost daily platelet transfusions were needed to keep platelet counts above the preferred level, initially 50.0 $\times 10^9$/L, later 20.0 $\times 10^9$/L. In addition, one red blood cell transfusion was indicated due to anaemia. Due to recurrent episodes of (subclinical) convulsions, levetiracetam was started and raised later. Ten days after presentation, no more (sub)clinical convulsions were reported. Follow-up radiological imaging did not show any new intracerebral abnormalities.

The differential diagnosis of the severe thrombocytopenia included bacterial or viral infections, alloimmune or autoimmune thrombocytopenia, bone marrow infiltrating disorders (e.g. neonatal leukaemia or neuroblastoma) or rare inherited bone marrow failure syndromes (e.g. Wiskott-Aldrich syndrome, thrombocytopenia absent radius syndrome, congenital amegakaryocytic thrombocytopenia (CAMT), Fanconi anaemia, Diamond Blackfan anaemia (DBA), dyskeratosis congenita (DC), radioulnar synostosis with congenital amegakaryocytic thrombocytopenia, Shwachman-Diamond syndrome).

As the child was treated in a health-care centre on a remote island with relatively limited resources, a stepwise approach was necessary. First of all, TORCH screen and viral screening for known common causes of neonatal thrombocytopenia were performed and were negative. There were no indications to suggest a haematological malignancy on the peripheral blood smear. Radiologic studies showed a normally developed radius which excluded thrombocytopenia absent radius syndrome.

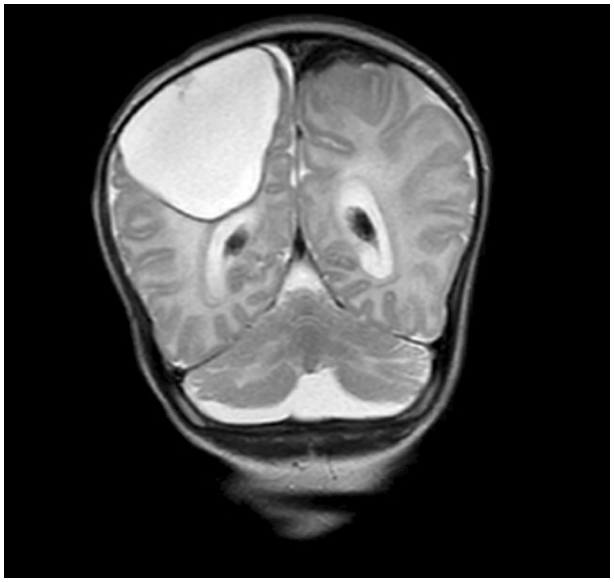

Fig. 1 Magnetic resonance imaging scan. T2 weighted frontal image showing a large porencephalic cyst on the right side and subarachnoidal left and intraventricular bleeding on both sides.

Furthermore, no other skeletal abnormalities were revealed which excluded DBA and radioulnar synostosis with congenital amegakaryocytic thrombocytopenia. The child did not have any physical malformations associated with Fanconi anaemia, DC, Shwachman-Diamond syndrome or DBA. Maternal platelet count was normal, no human platelet antigen (HPA)-1, -3, -5 and/or -15 incompatibility between mother and neonate was present, no maternal alloantibodies against neonatal/paternal HPA were detected and the cross-match between maternal serum and paternal platelets was negative. Free circulating plasma thrombopoietin (TPO) level was significantly elevated (726 IU/L, range 4–120 IU/L), suggestive for decreased platelet production.[1]

Subsequently, a targeted next-generation sequencing gene panel for thrombocytopenia/platelet function disorders was shipped and performed in the Netherlands in both the patient and the parents and was processed in 4 weeks. This demonstrated compound heterozygous variants in the myeloproliferative leukaemia protein (*MPL*) gene of the infant: a single nucleotide deletion in exon 3 (NM_005373.2(MPL):c.378del) and missense variant in exon 7 (NM_005373.2(MPL):c.1000T>C). Both parents were heterozygous for either one of these variants. These findings confirmed the diagnosis of CAMT. Due to the relatively limited resource setting and a clinically stable patient, no additional investigations were performed whilst waiting for the aforementioned results.

Evolution to pancytopenia was noticed at the age of 2 months with a negative trend of the haemoglobin level towards a new minimum of 8.0 g/dL, reticulocytopenia of 2‰ and neutropenia of 0.5×10^9/L (range 2.5–7.5 $\times 10^9$/L). The child was referred to a tertiary centre for haematopoietic stem cell therapy with a HLA-compatible sibling as donor. Neurological development was normal until transfer at the age of 10 weeks.

Discussion

CAMT is a rare autosomal recessive bone marrow failure syndrome, characterised by thrombocytopenia and reduced or absent bone marrow megakaryocytes. Approximately, a total of 100 cases have been reported in the literature. Seventy percent of the cases present at birth with signs of increased bleeding tendency due to thrombocytopenia.[2] Most patients present with hematomas and petechiae.[3] Twenty-one cases were described in the literature with intracranial bleedings. Similar to our case, two of them had both antenatally and postnatally acquired haemorrhages.[2–4] In our case, strikingly, there were no skin manifestations of thrombocytopenia, whilst imaging after minor head trauma, showed both newly acquired intracranial haemorrhages, and a large porencephalic cyst, most likely related to an antenatal intracranial haemorrhage.

Because this patient was admitted in a limited resource setting, a stepwise approach was used. First, the complete blood count and peripheral smear revealed a severe thrombocytopenia with normal-sized platelets, normal coagulation studies and no blasts. Maternal platelet count was normal. The plasma TPO level can be a useful diagnostic tool in guiding the differential diagnosis. A low TPO level is seen in myeloproliferative disorders in newborns where the increasing blast cells bind the free TPO. A normal level is detected in bacterial sepsis. This neonate had highly increased TPO level (>200 IU/mL) which is mainly detected in patients with congenital viral infections, severe asphyxia and amegakaryocytosis. Sometimes slightly elevated levels are observed in neonates with antibody-mediated neonatal platelet function disorders.[1] In the vast majority of cases, an elevated TPO level is characteristic for CAMT. Unfortunately, this diagnostic test is not available in all laboratories. We were also obliged to outsource this test to a tertiary centre.

Since we had a clinical stable child, an uneventful family history and resources were restricted, we opted to first wait for the results of the HPA-screening, the targeted next-generation sequencing gene panel and the viral screening before performing other diagnostic options. As these resulted in the diagnosis of CAMT, we did not perform a bone marrow puncture, chromosomal fragility screen or any other investigation (e.g. telomere length measurements to exclude DC, urinary catecholamines, abdominal ultrasound …). If this was another setting, we would opt to perform a more extensive diagnostic approach.

The molecular causes of this rare disease are homozygous or compound heterozygous variants in the *MPL*-gene coding for the TPO receptor.[5] A literature review described 58 different known variants in the *MPL*-gene responsible for CAMT.[2,3,5] *MPL* is mostly expressed on megakaryocytes and platelets and if their production is decreased, free circulating plasma TPO level rises.[6] Two *MPL*-gene variants were detected in this male infant. The first is a deletion of thymine in exon 3 (NM_005373.2(MPL):c.378del) causing a frameshift and a premature stop codon (p.Phe126Leufs*5). This nonsense variant has already been described as a cause of CAMT and was associated with a CAMT type 1.[1,5] This group is characterised by variants (deletions, frameshift or homozygous or compound heterozygous nonsense variants) with complete loss of function of the TPO receptor. Patients remain thrombocytopenic and have early progression to bone marrow failure and pancytopenia within a median of 22 months.[5]

The second variant in exon 7 (NM_005373.2(MPL):c.1000T>C) was a novel missense variant. This variant causes change of an

evolutionary strong conserved cysteine at amino acid position 334 into arginine (p.Cys334Arg). This variant has not been described before and is not yet reported in the Genome Aggregation Database.[7] Both variants were inherited from the heterozygous parents. Consequently, the child is compound heterozygous for two recessive variants in the *MPL*-gene. Since the patient had severe neonatal thrombocytopenia with elevated plasma TPO and since other causes of neonatal thrombocytopenia were excluded, these two gene variants are assumed to confirm the diagnosis of CAMT. Therefore, we can conclude that the c.1000T>C missense variant is a new pathogenic variant most likely to cause CAMT.

In conclusion, in this report, we describe a neonate with seizures without any other bleeding symptoms, an atypical clinical presentation of CAMT, based on compound heterozygous variants in the *MPL*-gene, one of which has not previously been described.

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