

# Megaloblastic anaemia, diabetes and deafness in a 2-year-old child



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Megaloblastic anaemia in childhood usually occurs as a result of dietary folate deficiency or, rarely, congenital disorders of vitamin B<sub>12</sub> metabolism.

We present a 2-year-old girl with megaloblastic anaemia and insulin-dependent diabetes mellitus, both of which proved responsive to pharmacological doses of thiamine. She was also found to have sensorineural hearing loss.

Also known as Rogers' syndrome, thiamine-responsive megaloblastic anaemia is the result of inactivating mutations in a gene encoding a thiamine transporter.

A clinical diagnosis is supported by characteristic bone marrow findings and can be confirmed by demonstrating apoptosis in skin fibroblasts cultured in thiamine-depleted medium. Where available, DNA sequencing is definitive.

There is rapid reticulocytosis after thiamine administration. We recommend a trial of therapy for megaloblastic anaemia not responding to folate and vitamin B<sub>12</sub>, especially in a deaf and/or diabetic child.

First described by Rogers, thiamine-responsive megaloblastic anaemia (TRMA) is a rare autosomal-recessive disorder characterised by megaloblastic anaemia, insulin-dependent diabetes mellitus and sensorineural deafness.<sup>1</sup>

The diagnosis is supported by a megaloblastic bone marrow, particularly the presence of ringed sideroblasts. Paranuclear iron-laden mitochondria are seen in normoblasts on electron microscopy.<sup>2</sup> The diagnosis can be confirmed by demonstrating apoptosis in fibroblasts cultured in thiamine-free medium.<sup>3</sup> DNA sequencing has demonstrated mutations in the TRMA gene; a thiamine transporter gene SLC19A2.<sup>4</sup>

The condition responds variably to thiamine. Anaemia resolves but a degree of macrocytosis persists. Insulin requirements usually decrease but the deafness is irreversible.

We present a 2-year-old girl with the condition, and review the literature of this disorder.

## Case report

A 2-year-old girl was seen at a regional hospital at the age of 1 year with severe macrocytic anaemia and diabetic

ketoacidosis (DKA). She was transfused and commenced on folate and was discharged off insulin without further monitoring of her blood glucose. At follow-up she was found to have a chronic macrocytic anaemia with a high serum lactate dehydrogenase. A presumptive diagnosis of a congenital haemolytic disorder was made, and she was maintained on folate.

After another episode of DKA, she was referred to our service. There was no history of neonatal jaundice, and no family history of anaemia or consanguinity. Her motor milestones were normal but she had never acquired meaningful speech, being able only to babble.

She was well nourished, pale without jaundice and there were no dysmorphic features. She was profoundly deaf. Audiological assessment confirmed severe bilateral sensorineural hearing loss. Control of her diabetes required 18 units of insulin daily.

Her haemoglobin concentration was 5.4 g/dl with a mean cell volume of 101 fl, and a reticulocyte count of 68 x 10<sup>9</sup>/l (1.4% corrected). The white cell count and differential count were normal, as was the platelet count. The smear revealed oval macrocytes and poikilocytosis. Red cell folate (2 114 ng/ml), serum vitamin B<sub>12</sub> (615 pg/ml) and ferritin (184 ng/ml) levels were normal. Investigations for

haemolysis were negative. The glycosylated haemoglobin level was 5.7% (normal 2.9 - 4.6%), indicating satisfactory glycaemic control. Bone marrow examination revealed a hypercellular marrow with marked erythroid hyperplasia and trilineage dysplasia. Erythropoiesis was markedly megaloblastic but the granulocyte series had only occasional giant bands. Megakaryocytes had both exploded and bare nuclei. A Perl's Prussian blue stain showed numerous ringed sideroblasts (20%). Owing to technical difficulties, electron microscopy failed to demonstrate iron deposits in the mitochondria.

Suspecting the diagnosis of TRMA, we administered an intramuscular loading dose of 100 mg of thiamine, followed by 50 mg daily by mouth. There was a rapid reticulocytosis – the corrected reticulocyte count rose from 1.4% to 10.2% within a week, and the haemoglobin concentration increased from a post-transfusion value of 7.7 g/dl to 9.8 g/dl after 14 days. Her daily insulin requirements fell to 4 units.

Fibroblasts were later obtained from a skin biopsy, and cultured in Dulbecco's minimal essential medium supplemented with 10% fetal calf serum. Substitution with commercially obtained medium specifically formulated to be thiamine-free failed to induce the characteristic apoptosis. This was possibly because of trace amounts of thiamine present in the fetal calf serum used. DNA sequencing was not available.

The patient was given a hearing aid, and discharged to the regional hospital on lifelong thiamine supplementation.

Three years after diagnosis the patient maintains a haemoglobin concentration of 12.2 g/dl with a mean cell volume of 92 fl, and requires 3 - 5 units of insulin daily. She has been referred to a school for deaf children.

## Discussion

Rogers *et al.*<sup>5</sup> first reported TRMA in 1969. They described an 11-year-old girl with megaloblastic anaemia, diabetes and deafness. The molecular biology has recently been elucidated.

This autosomal-recessive disorder is the result of mutations of the SLC19A2 gene located on chromosome 1. First identified in 1999,<sup>6</sup> the product of this gene is a high-affinity thiamine transporter<sup>3</sup> that mediates facilitated transport of thiamine across cell membranes. The rapid transport of thiamine by this facilitated transport system appears to be essential only for haematopoietic, pancreatic islet and auditory nerve cell function. Passive uptake by a separate low-affinity, high-capacity system appears adequate to protect other tissues from intracellular thiamine depletion. Hence TRMA patients with adequate dietary thiamine seldom manifest the classic signs of severe thiamine deficiency typical of

beriberi, including peripheral neuropathy, cardiomyopathy and central nervous system involvement.

A consequence of intracellular thiamine depletion is a decrease in activity of thiamine-dependent transketolase, thereby critically reducing the synthesis of ribose via the non-oxidative arm of the pentose phosphate pathway.<sup>7</sup> Lack of ribose for nucleic acid synthesis leads to cell-cycle arrest and triggers apoptosis. It has been suggested that cumulative cell loss in sensitive tissues explains why the clinical manifestations are not apparent in early infancy.<sup>8</sup> The gradual decline in glucose tolerance seen in our patient is consistent with this hypothesis.

Thiamine supplementation in pharmacological doses can compensate to a degree by increasing passive uptake via the low-affinity system. The ability of pluripotent haematopoietic stem cells to repopulate the bone marrow after the death of thiamine-deficient red cell precursors explains the recovery of the red cell indices. Ongoing macrocytosis demonstrates that their thiamine-replete successors remain sensitive to the high-affinity transport defect.<sup>3</sup> Low-turnover pancreatic beta cells and auditory nerve cells presumably require thiamine primarily to maintain their adenosine triphosphate (ATP) levels by oxidative phosphorylation, rather than for nucleic acid synthesis. Beta cells seem to be of intermediate sensitivity to intracellular thiamine depletion, demonstrating a degree of recovery with supplementation, whereas auditory nerve cells are exquisitely sensitive, presumably because of their higher energy requirements. Here ongoing apoptosis results in irreversible deafness.

Our patient ran a typical course with rapid improvement in the anaemia and a partial response with regard to insulin requirements. The sensorineural hearing loss has not improved, and this is consistent with the experience of others.<sup>1</sup>

We suggest that the typical clinical presentation together with megaloblastosis in the face of normal vitamin B<sub>12</sub> and folate levels should prompt a therapeutic trial of thiamine. Fibroblast culture or DNA sequencing would be definitive.

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