

CASE REPORT

A RARE CASE OF THIAMINE-RESPONSIVE MEGALOBLASTIC ANAEMIA SYNDROME: A DISORDER OF HIGH-AFFINITY THIAMINE TRANSPORT

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A three years old boy presented with sensory neural hearing loss since birth, Diabetes mellitus and anaemia. On investigation he was found to be suffering from Thiamine Responsive Megaloblastic Anaemia (TRMA) a very rare condition diagnosed in our settings.

Keywords: TRMA, Megaloblastic Anaemia, Thiamine, Haemoglobin, Bone Marrow, Prenatal Testing

INTRODUCTION

The condition is caused by a deficiency of a thiamine (vitamin B₁) transporter protein which means that the body is unable to effectively utilize thiamine from the diet. Thiamine responsive megaloblastic anaemia syndrome is listed as a 'rare disease' by the Office of Rare Diseases (ORD) of the National Institutes of Health (NIH) USA. Consortium of European partners, currently defines a condition rare when it affects 1 person per 2,000. They list Thiamine responsive megaloblastic anaemia syndrome as a 'rare disease'. Twenty-five affected individuals reported to date have been diagnosed.¹ The diagnosis of TRMA should be suspected in patients with syndrome of diabetes including hearing loss and anaemia, even if the latter is only very mild and particularly in the case of consanguinity. The bone marrow aspirate usually shows megaloblastic changes and ringed sideroblasts.² TRMA syndrome gene maps to a region on chromosome 1q23.2–23.3.³ The clinical features of TRMA, resembling in part those found in typical mitochondrial disorders with complex deficiency, may be caused by a secondary defect in mitochondrial energy production.⁴ Designated 'SLC19A2' as a member of the solute carrier gene super family, this gene is mutated in all TRMA kindred's studied to date. The product of the SLC19A2 gene is a membrane protein which transports thiamine (vitamin B₁) with sub-micromolar affinity.⁵ Cells from TRMA patients are uniquely sensitive to thiamine depletion to the nanomolar range, while pharmacologic doses of vitamin B₁ ameliorate the anaemia and Diabetes.⁶

CASE REPORT

A three years old boy was referred to us by our worthy hospital Paediatrician for the diagnosis and management of anaemia. On taking of comprehensive history from his father, the boy was the only child of their parents. He was born at a local hospital and had hearing loss since birth. He was observed to have developed gradual pallor about a year and a half after birth. The child was transfused twice with red cell concentrates for correction of

anaemia. The physical and systemic examination revealed an anaemic child with sensory neural hearing loss. Initial Lab investigations showed anaemia (Hb 6.1 gm/dl) and peripheral blood macrocytosis (MCV 110 fl/l). Bone marrow examination was performed to determine the cause of macrocytosis. Megaloblastosis with hypercellular marrow was seen. The bone marrow iron was increased with moderate number of siderocytes and sideroblasts. On the basis of bone marrow examination a diagnosis of Megaloblastic anaemia was made. Here the child's father gave a valuable clue that two of the child's paternal first cousins suffered from exactly similar condition. This led to a bit of brain storming by the authors and a differential diagnosis of Thiamine-Responsive Megaloblastic Anaemia Syndrome was kept in mind. The diagnosis of TRMA should be suspected in patients with syndrome of diabetes including hearing loss and anaemia. The fasting blood glucose levels done two weeks apart revealed a very high glucose content (16.2 mmol/l). We decided to get the molecular diagnosis. The child was taken to the United States where his DNA analysis confirmed the 1q23.2–23.3 mutation. The child has been on Thiamine therapy for the past six months. His anaemia has significantly improved (Hb 10.6 gm/dl). The child is transfusion independent since the start of therapy. The patient is on insulin therapy at the moment.

DISCUSSION

Thiamine responsive megaloblastic anaemia syndrome (TRMA), an autosomal recessive disorder caused by the deficiency of thiamine transporter protein, is the association of diabetes mellitus, anaemia and deafness.⁷

The water-soluble micronutrient thiamine is required for normal tissue growth and development in humans. Thiamine is accumulated into cells through the activity of two cell surface thiamine transporters (hTHTR1 and hTHTR2), which are differentially targeted in polarized tissues. Mutational dysfunction of hTHTR1 is associated with the clinical condition of thiamine-responsive megaloblastic anaemia: the

symptoms of which are alleviated by thiamine supplementation.⁸

Diabetes in this syndrome is due to an insulin insufficiency that initially responds to thiamine supplements; however, most patients become fully insulin dependent after puberty.⁹

Cochlear histological analysis showed a pattern uncommon for sensor neural hearing loss: selective loss of inner hair cells after 1–2 weeks on low thiamine and significantly greater inner than outer hair cell loss after longer low-thiamine challenges. Such a pattern is consistent with the observed discrepancy between ABR and OAE threshold shifts. The possible role of thiamine transport in other reported cases of selective inner hair cell loss is confirmed.¹⁰ Retinal abnormality and visual disturbances occur in thiamine-responsive megaloblastic anaemia.¹¹

Megaloblastic changes in the bone marrow are morphologically quite distinctive, and the several causes of this condition, including specific nutrient deficiencies, metabolic errors, and certain drugs, are well described.¹² Among the more obscure causes of megaloblastic anaemia is the acronymic curiosity thiamine-responsive megaloblastic anaemia, the use of mass spectrometry in conjunction with stable isotope-labelling techniques has made it possible to unlock doors along previously inaccessible hallways of gene function analysis in the metabolomic maze. The door to TRMA was thus opened by Boros *et al*,¹³ who have pioneered the use of Stable Isotope-based Dynamic Metabolic Profiling (SIDMAP) as a key to better understanding of changes in substrate flow as a basis for drug mechanisms and disease. Teaming up with the Boston group who first identified the loss of function mutation in the high-affinity, low-capacity thiamine transporter in TRMA, the authors have pinpointed the cause of disruption of nucleic acid synthesis that leads ultimately to premature apoptosis in this intriguing genetic disorder. Through tracking the stable¹⁵ C-labelled glucose in fibroblasts from patients with TRMA, these authors concluded that the underlying lesion in this condition resides in the pentose cycle, specifically the transketolase enzyme, which requires thiamine pyro-phosphate as a cofactor¹⁴. Through a consideration of the several interconnected pathways of glycolysis, the tricarboxylic acid cycle, and ribose synthesis, the authors defined substrate flux in TRMA and normal wild-type fibroblasts grown in both low- and high-thiamine medium. They concluded that defective high-affinity thiamine transport in TRMA leads to a critical reduction in *de novo* generation of ribose with consequent cell-cycle arrest that triggers precocious apoptosis. Their results clearly demonstrate a selective and time-dependent loss of ribose synthesis

in TRMA patients that is most marked under thiamine-deprived culture conditions and is partially restored by thiamine supplementation, explaining the clinical responsiveness of TRMA patients to high doses of thiamine. Use of the powerful tools provided by SIDMAP and related techniques that use even more sensitive accelerator mass spectrometry with ultra-low-dose labelling techniques provides the promise to address, perhaps *in vivo*, similar unanswered questions involving the molecular basis for disease. Applying these methods to the study of the more common conditions that cause megaloblastic anaemia, but that are still shrouded in mystery, could ultimately shed similar light on their mechanism.

The diagnosis of TRMA is based on an obligate triad of clinical features described above.¹⁵ Examination of the bone marrow reveals megaloblastic anaemia with erythroblasts often containing iron-filled mitochondria (ringed sideroblasts). *SLC19A2*, which encodes the high-affinity thiamine transporter, is the only gene known to be associated with TRMA. All individuals with the diagnostic phenotypic triad evaluated by sequence analysis have identifiable mutations in the *SLC19A2* gene.¹⁶ Sequence analysis of *SLC19A2* DNA is available clinically. That reduced nucleic acid production through impaired transketolase catalysis is the underlying biochemical disturbance that likely induces cell cycle arrest or apoptosis in bone marrow cells and leads to the TRMA syndrome in patients with defective high-affinity thiamine transport.¹⁷ Pharmacological dose thiamine normalizes haematological abnormalities and their effects on the course of diabetes mellitus. Thiamine induces a remarkable haematological response and improvement in the diabetic control but has no effect on deafness.¹⁸

Treatment of TRMA focuses on lifelong use of pharmacologic doses (25–75 mg per day) of thiamine (Vitamin B₁) in affected individuals.¹⁹ Surveillance to monitor the efficacy of the oral thiamine therapy as well as disease progression should be performed at least yearly and includes: haematologic tests (CBC, reticulocyte count), assessment for glucose intolerance (fasting serum glucose concentration, OGTT, urine analysis), and hearing, ophthalmologic, and cardiac evaluations.²⁰

TRMA is inherited in an autosomal recessive manner. At conception, the sibs of an affected individual have a 25% chance of being affected, a 50% chance of being an asymptomatic carrier, and a 25% chance of being unaffected and not a carrier. Once an at-risk sib is known to be unaffected, the risk of his/her being a carrier is 2/3. Prenatal testing is available for families in which the disease-causing mutations have been identified.²¹

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