Case Report

Delayed Diagnosis of Cobalamin E Defect in an Adolescent Patient

Merve Koç Yekedüz, Elif Unal Ince1, Talia İléri1, Mehmet Ertem1, Fatma Tuba Eminoglu

Department of Pediatric Metabolism, 1Department of Pediatric Hematology, Faculty of Medicine, Ankara University, Ankara, Turkey

ABSTRACT

Cobalamin and its metabolites play a critical role in deoxyribonucleic acid synthesis. Disorders of cobalamin metabolism are rare and related with neurological and hematological problems. We report an adolescent patient with cobalamin E (CblE) defect presenting with megaloblastic anemia, mental retardation, cerebral atrophy, cortical visual impairment, white matter changes on brain magnetic resonance imaging, and hyperhomocysteinemia. Homozygous mutation at the c.245C>T in exon 3 of the MTRR gene was identified, which had been found to be related to CblE defect. He was treated with betaine, folic acid, vitamin B6, riboflavin, hydroxycobalamin (OH-B12), and carnitine. During treatment, homocysteine levels decreased over time.

KEYWORDS: Cobalamin E defect, errors of vitamin B12 metabolism, hyperhomocysteinemia, remethylation defects

INTRODUCTION

Cobalamin and its metabolites play a critical and important role in deoxyribonucleic acid (DNA) synthesis and energy metabolism.[1] Cobalamin E (CblE) (methionine synthase reductase [MTRR] gene 5q15.31) is responsible for the remethylation of homocysteine to methionine, and CblE deficiency is an autosomal-recessive rare inborn error of cobalamin metabolism.[2] To date, 19 different mutations have been identified.[3] Remethylation defects are usually characterized by homocystinuria and hypomethioninemia and no methylmalonic acid excretion in urine.[4] This classic triad defines the CblG and CblE subtypes, and final diagnosis must be confirmed by molecular assays [Figure 1].

Disorders of cobalamin metabolism E are rare and usually present with neurological and hematological problems. Patients are often diagnosed in the first 2 years. Common findings are megaloblastic anemia, neurological deficits, mental retardation, psychiatric problems, muscular hypotonia, feeding problems, electroencephalogram (EEG) abnormalities, cerebral atrophy, and nystagmus.[4] Hypertonia, seizures, and ataxia are less frequent. Older children and adults can manifest with psychiatric symptoms and subacute spinal degeneration. Psychiatric symptoms and cognitive defects may be variable.[2]

Early diagnosis and treatment are very important to check for neurological findings. Cobalamin is the cofactor of methionine synthase.[3] Cobalamin treatment is usually given for anemia and biochemical abnormalities in patient, and cyanocobalamin is used for neurological and hematological improvement, but hydroxycobalamin has been more effective than cyanocobalamin.[4,5] Betaine is a potent methyl group donor, which has a role in remethylation of homocysteine to methionine using betaine–homocysteine methyltransferase that bypass methylcobalamin-dependent pathway.[6]

Here, we present a 15-year-old Turkish male patient with a delayed diagnosis of CblE type of homocystinuria with neurological involvement and psychiatric findings.

CASE REPORT

A 15-year-old male patient was admitted to the pediatric outpatient clinic for weakness and fatigue. He was...
referred to psychiatry clinic at 8 years of age for learning difficulties and social isolation at school and was diagnosed with mental retardation and autism. He had been followed by psychiatry clinic since then. He was the second child of consanguineous parents. He was on methylphenidate and risperidone medications. Physical examination showed microcephaly, mental retardation, and Marfan-like long fingers. Laboratory examination revealed normal hemoglobin with macrocytosis (MCV: 99.7 fL). White blood cell and platelet counts were normal. There were hypersegmented neutrophils on peripheral smear with macroovalocytes, suggestive of megaloblastic changes. Initial workup for macrocytosis, including vitamin B12, folic acid, and thyroid function tests were normal, but homocysteine level was very high with 170.1 µmol/L (normal range: 5–14 µmol/L). With homocysteine being very high without vitamin B12 and folic acid deficiency, with mental retardation (IQ: 25–49), and consanguinity, further investigation for possible inherited metabolic disorder was initiated. Methionine level was examined to differentiate the remethylation defects versus transsulfuration defects and it was found to be low (patient: 10.14 µmol/L; normal range: 11–30 µmol/L), leading the diagnosis to remethylation defects [Figure 2]. MTHFR (methylene tetrahydrofolate reductase) gene studies for common mutations showed homozygote C677T gene mutation. MTHFR gene was also sequenced for any other mutation, which was negative and there was no mutation on MTHFR gene to explain the clinical symptoms including megaloblastosis. Vitamin B12 and folic acid were normal. Urine organic acid analysis was examined for other remethylation defects. There was no excretion of methylmalonic acid in urine. On neurological evaluation, he had mild hearing loss, EEG was normal. On cranial magnetic resonance imaging (MRI), there was cerebral atrophy, loss in cerebral white matter, gliotic changes in periventricular white matter, and ischemic lesions in the right frontal operculum. With these findings, there was a strong suspicion for cobalamin defects, especially for CblE/G, and patient’s DNA was sent for genetic analysis. Although waiting for the results of the genetic mutation analysis, treatment was started with betaine (6 mg/day), folic acid (10 mg/day), vitamin B6 (500 mg/day), riboflavin (50 mg/day), hydroxycobalamin (1 mg/week intramuscular), and carnitine (1000 mg/day). Homocysteine level decreased to 48.5 µmol/L (normal: 5–14 µmol/L) and to 32.7 µmol/L respectively. The diagnosis was confirmed with the results of the genetic mutation analysis, which was identified c.245C>T in exon 3 of the MTRR gene in a homozygous state (MTRR gene allele 1: c.245C>T p.[pro82leu], allele 2: c.245C>T p.[pro82leu]). This missense mutation predicts an amino acid change from proline to leucine at position 82 (p.[pro82Leu]) in the mature protein, which was previously been reported.[9] This result was consistent with the diagnosis of CblE type of homocystinuria in the patient and both parents were found to be carriers of the mutation.
Informed consent was received from the family for writing case report.

**DISCUSSION**

The cobalamin E (CblE) defect is a rare inborn error of cobalamin metabolism, leading to impairment of the remethylation of homocysteine to methionine. Earlier biochemical studies proposed that the methionine synthase enzyme might be activated by two different reducing systems, but mutations were reported in only the *MTRR* gene in CblE patients. Proportion of disorders of intracellular cobalamin metabolism attributed to mutation of *MTRR* is less than 5%. Chromosome location of CblE is 5p15.31. To date, 19 different mutations have been identified. Feeding problems, hypotonia, cognitive impairment, and macrocytic anemia are the most frequent symptoms. Patients are often diagnosed with neurological symptoms in the first 2 years. Delay in diagnosis depends on first symptoms and clinical pattern. Patients can show progressive encephalopathy with regression that causes deteriorating functions at school or work. Our patient did not have any neurological symptom at the early ages, including muscle weakness and difficulty in swallowing. Mental retardation, learning difficulties, and social isolation were noted after he started school at 8 years of age. With these symptoms, he was diagnosed with autism and mental retardation and had been followed by psychiatry clinic since then. He was only referred to the department of pediatrics at the age of 15 for fatigue. Epilepsy is common but not specific in remethylation disorders. The seizures and EEG findings are nonspecific. Our patient’s EEG was normal, and he had no seizures at any time. On his brain MRI, gliotic changes were observed on white matter of periventricular area and cerebral atrophy. Our patient was diagnosed late although he had manifestations suggestive of disorders of CblE metabolism.

Macrocytosis is the most important clue to diagnosis. It is not known if the patient had a complete blood count before. Even if the complete blood count was checked, macrocytosis may have been missed because of absence of anemia. Laboratory examination revealed normal hemoglobin with macrocytosis and hyperhomocysteinemia. Our first initial doubt was the combination of neuropsychiatric symptoms and macrocytosis.
The diagnosis of CblE defect is difficult. After clinical suspicion, first, biochemical tests should be carried out and the final diagnosis should be made by showing the genetic mutation. But there are limited laboratories performing these analyses. We were able to send his DNA to the laboratory of University of Zurich. They found the mutation c.245C>T in exon 3 of the MTRR gene in a homozygous state. (MTRR gene allele 1: c.245C>T p.[pro82leu], allele 2: c.245C>T p.[pro82leu]). Both parents were carriers for the same mutation. This result was consistent with the diagnosis of CblE type of homocystinuria.[2]

Cobalamin treatment is usually given for anemia and biochemical abnormalities in patients, and cyanocobalamin is used for neurological and hematological improvement but hydroxycobalamin has been more effective than cyanocobalamin.[4,5] Hydroxycobalamin is beneficial for neurologic development.[4] However, the impact of treatment is less in biochemical parameters and generally hyperhomocysteinemia persists.[7] Betaine and hydroxycobalamin show synergistic effect for metabolic control on decreasing homocysteine.[12] Folate treatment is an adjunctive therapy.[8] Levo-carnitine treatment excretes propionyl group and saves the carnitine level in patients.[9] We used hydroxycobalamin and betaine together. We also added folic acid and carnitine to his treatment. During treatment, homocysteine levels decreased in time.

The established neurological disorders do not regress although prevention is possible with the early administration of specific treatment.[8] In our case, the absence of neurological symptoms in early life and the initial diagnosis of autism spectrum disorders led to a long delay in the initiation of specific therapy. Despite the introduction of treatment, we did not see any clinical improvement in neuropsychiatric symptoms. Early treated patients showed a more favorable neurological outcome.[10]

Homocysteine levels above 45 μmol/L have been reported to be associated with the development of vascular complications in several patients.[11] Identification and treatment of hyperhomocysteinemia, especially in younger people, is of extreme importance.[12] With appropriate treatment, the incidence of thromboembolic complications can be reduced and may even be prevented in late-onset patients.[2] Therefore, in our cases, we aim to keep the homocysteine levels in safe range with treatment.

In summary, patients who are followed with psychiatric problems should be evaluated for underlying inherited metabolic diseases. In case of a patient with megaloblastic anemia and neurological findings, if the vitamin B12 level is normal, one of the remethylation defects should be suspected.

Ethical approval
This article does not contain any studies with human participants or animals performed by any of the authors.

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Conflicts of interest
There are no conflicts of interest.

REFERENCES
1. Kandula T, Peters H, Fahey M. Cobalamin E defect, a rare inborn error of vitamin B12 metabolism: value of early diagnosis and treatment. J Clin Neurosci 2014;21:1815-7.
2. Huemer M, Bürer C, Ješina P, Kožich V, Landolt MA, Suormala T, et al. Clinical onset and course, response to treatment and outcome in 24 patients with the CblE or CblG remethylation defect complemented by genetic and in vitro enzyme study data. J Inherit Metab Dis 2015;38:957-67.
3. Huemer M, Diodato D, Schwahn B, Schiff M, Bandeira A, Benoist JF, et al. Guidelines for diagnosis and management of the cobalamin-related remethylation disorders CblC, CblD, CblE, CblF, CblG, CblJ and MTHFR deficiency. J Inherit Metab Dis 2017;40:21-48.
4. Watkins D, Rosenblatt DS. Inborn errors of cobalamin absorption and metabolism. Am J Med Genet C Semin Med Genet 2011;157C:33-44.
5. Hallam LJ, Sawyer M, Clark AC, Van der Weyden MB. Vitamin B12-responsive neonatal megaloblastic anemia and homocystinuria with associated reduced methionine synthase activity. Blood 1987;69:1128-33.
6. Schwahn BC, Hafner D, Hohlfeld T, Balkenhol N, Laryea MD, Wendel U. Pharmacokinetics of oral betaine in healthy subjects and patients with homocystinuria. Br J Clin Pharmacol 2003;55:6-13.
7. Bartholomew DW, Batshaw ML, Allen RH, Roe CR, Rosenblatt D, Valle DL, et al. Therapeutic approaches to cobalamin-C methylmalonic acidemia and homocystinuria. J Pediatr 1988;112:32-9.
8. Spector R. Affinity of folic acid for the folate-binding protein of choroid plexus. Arch Biochem Biophys 1979;194:632-4.
9. Enns GM, Barkovich AJ, Rosenblatt DS, Fredrick DR, Weisiger K, Ohnstad C, et al. Progressive neurological deterioration and MRI changes in CblC methylmalonic acidemia. J Child Neurol 2003;18:181-6.
acidaemia treated with hydroxocobalamin. J Inherit Metab Dis 1999;22:599-607.

10. Diekman EF, de Koning TJ, Verhoeven-Duif NM, Rovers MM, van Hasselt PM. Survival and psychomotor development with early betaine treatment in patients with severe methylenetetrahydrofolate reductase deficiency. JAMA Neurol 2014;71:188-94.

11. Carrillo-Carrasco N, Sloan J, Valle D, Hamosh A, Venditti CP. Hydroxocobalamin dose escalation improves metabolic control in CblC. J Inherit Metab Dis 2009;32:728-31.

12. Niazi F, Rahman A, Batool U. Hyperhomocysteinemia presenting with complete unilateral intracranial and extracranial carotid occlusion in a young patient. J Coll Physicians Surg Pak 2017;27:101-3.