Early Onset Methylmalonic Aciduria and Homocystinuria cblC Type With Demyelinating Neuropathy

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Methylmalonic aciduria and homocystinuria, cblC type, is the most common inborn error of vitamin B_{12} (cobalamin) metabolism. The recent cloning of the disease gene, *MMACHC*, has permitted genotype-phenotype correlation. In a 1-year-old girl, compound heterozygous c.271dupA and c.616C>T mutations in *MMACHC* were identified as causing an early onset methylmalonic aciduria and homocystinuria, cblC type, which was complicated by sensorimotor peripheral demyelinating neuropathy. © 2010 by Elsevier Inc. All rights reserved.

Introduction

Patients with methylmalonic aciduria and homocystinuria have defective metabolism of vitamin B_{12} (cobalamin) in its two metabolically active forms: methylcobalamin and adenosylcobalamin. Methylmalonic aciduria and homocystinuria, cblC type (OMIM #277400), is the most frequent inborn error of vitamin B_{12} metabolism; based on the age of onset, two distinct clinical phenotypes (early onset and late onset) have been demonstrated [1,2]. In 2006, Lerner-Ellis et al. [3] reported the identification of a gene responsible for this disorder, located on chromosome region 1p34.1: *MMACHC* [methylmalonic aciduria (cobalamin deficiency) cblC type, with homocystinuria] (OMIM *609831). The functions of the *MMACHC* gene product are not completely understood, but it apparently acts as an intracellular trafficking cobalamin chaperone. Reported here are clinical and biochemical findings in a 1-year-old child with early onset methylmalonic aciduria and homocystinuria, cblC type, complicated by peripheral sensorimotor demyelinating neuropathy, who carried compound heterozygous c.271dupA and c.616C>T mutations in the *MMACHC* gene.

Case Report

The patient, a girl, was the second child of healthy, nonconsanguineous Italian parents. She was born at the 37th gestational week with an uneventful delivery. Intrauterine growth retardation had been noticed since the seventh month of pregnancy. Her weight at birth was 2.150 kg (10th to 25th centile), and her head circumference was 30 cm (<3rd centile). Apgar scores were 7 and 8 at 1 and 5 minutes, respectively. She had poor sucking, feeding difficulties, and severe hypotonia from birth.

The child was presented at 7 months of age with marked hypotonia, converging strabismus, and psychomotor delay. Neurologic examination revealed poor eye contact, strabismus, horizontal nystagmus, microcephaly, and severe axial and segmental hypotonia with reduced movement and areflexia. Routine laboratory examination findings were normal for hemogram and for erythrocyte mean corpuscular volume, glucose, transaminase, vitamin B_{12}, folate, ammonium, lactate, creatine kinase levels, celiac disease screening, and serum free thyroid hormones. Similarly, there were no abnormal findings from funduscopic examination, brainstem acoustic evoked potentials, and abdominal and cardiac ultrasound. Cranial magnetic resonance imaging revealed a mild delay in myelination, and electroencephalograms displayed poorly organized background activity with occasional spikes and wave over the left occipitoparietal regions. No seizures were noticed.

Because of severe hypotonia and areflexia, electrophysiologic studies in the upper and lower limbs were performed, revealing uniform and symmetric slowing of both sensory and motor conduction velocities and normal amplitude of the compound sensory nerve and muscle nerve potentials (Tables 1 and 2). Moreover, the sensory nerve action potential of sural, medial plantar, and superficial peroneal nerves was bilaterally absent (Table 2). No temporal dispersion was detected. Distal latency values were at the upper limit of normal range for age

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(Tables 1 and 2). Electromyographic evaluation of the bilateral anterior tibial and gastrocnemius muscles was unremarkable.

Further investigations revealed elevated plasma homocysteine, at 160 μmol/L (normal range, 2.6-10.3 μmol/L); markedly elevated urinary methylmalonic acid, at 600 μg/mg creatinine (normal value, <5 μg/mg creatinine); and decreased plasma methionine, at 4 μmol/L (normal range, 9-42 μmol/L). Studies on the patient's cultured skin fibroblasts revealed low incorporation of both [^{14}C]propionate and [^{14}C]methyltetrahydrofolate, slightly low uptake of [^{57}Co]cyanocobalamin, and a decreased synthesis of both adenosylcobalamin and methylcobalamin.

The diagnosis of methylmalonic aciduria and homocystinuria, cblC type, was confirmed by mutational analysis, which demonstrated heterozygosity for the 271dupA and 616C>T mutations in the *MMACHC* gene. Treatment with oral hydroxycobalamin, vitamin B$_6$, folic acid, carnitine, and betaine slightly decreased metabolite levels, although without complete normalization and without clinical improvement.

At the age of 12 months, the patient's clinical course was complicated by interstitial pneumonia with severe progression of neurologic impairment. The patient died 4 days later of respiratory failure and cardiac arrest.

Discussion

With more than 400 cases described, methylmalonic aciduria and homocystinuria, cblC type, is the most frequent inborn error of vitamin B$_{12}$ metabolism [2]. Two clinical forms have been described, based on age at onset (namely, an early and a late onset form). The early onset form, with the appearance of symptoms within the first year of life, includes failure to thrive, feeding difficulties, mild dysmorphic features [4], and ophthalmologic and cardiac anomalies [5,6]. Neurologic symptoms of the early onset form include hypotonia, progressive developmental delay, microcephaly, myelopathy, seizures,

Table 1. Motor nerve conduction findings at age 7 months in a case of early onset methylmalonic aciduria and homocystinuria, cblC type, complicated by sensorimotor peripheral demyelinating neuropathy

Measure	Patient Value	Normal Value
Right peroneal nerve		
Conduction velocity, m/s	30	>31
Response amplitude, mV	3.5	>2
Distal latency, ms	3.1	<3.2
Left peroneal nerve		
Conduction velocity, m/s	25	>31
Response amplitude, mV	3.6	>2
Distal latency, ms	3	<3.2
Right posterior tibial nerve		
Conduction velocity, m/s	22	>31
Response amplitude, mV	7	>2
Distal latency, ms	3	<3.2
Left posterior tibial nerve		
Conduction velocity, m/s	24	>31
Response amplitude, mV	7.5	>2
Distal latency, ms	3.1	<3.2
Left median nerve		
Conduction velocity, m/s	24	>26
Response amplitude, mV	3	>2
Distal latency, ms	2.6	<3.2
Right median nerve	not tested	

Table 2. Sensory nerve conduction findings at age 7 months in a case of early onset methylmalonic aciduria and homocystinuria, cblC type, complicated by sensorimotor peripheral demyelinating neuropathy

Measure	Patient Value	Normal Value
Right median nerve		
Conduction velocity, m/s	25	>39
Response amplitude, μV	15	>14
Distal latency, ms	2.2	<2.4
Left median nerve		
Conduction velocity, m/s	25	>39
Response amplitude, μV	15	>14
Distal latency, ms	1.8	<2.4
Right ulnar nerve		
Conduction velocity, m/s	25	>34
Response amplitude, μV	12	>10
Distal latency, ms	1.9	<2.5
Left ulnar nerve	not tested	
Sural nerve	no response	
Medial plantar nerve	no response	
Superficial peroneal nerve	no response	

and hydrocephalus [1]. A milder phenotype is described in patients with the later onset form, in which behavioral disturbances and neurologic impairment are noticed after the age of 4 years [1,7].

In 2006, Lerner-Ellis et al. [3] reported the identification of *MMACHC*, the gene responsible for methylmalonic aciduria and homocystinuria, cblC type. Although this disorder has been demonstrated to be a heterogeneous condition, identification of the gene involved has allowed study of phenotype-genotype correlation. To date, 56 mutations have been detected in 366 individuals with methylmalonic aciduria and homocystinuria, cblC type. The most common mutation, c.271dupA (p.Arg91LysfsX14), accounts for 42% of all pathologic alleles; the second most common mutation, c.394C>T (p.R132X), represents 20% of affected alleles [2]. Individuals with c.394C>T tend to present with late onset disease, whereas patients with c.271dupA tend to present in infancy. Moreover, in the published cases of patients homozygous for c.271dupA mutation [2,8,9], the phenotype was invariably early onset.

In the present case, mutational analysis of the *MMACHC* gene revealed compound heterozygosity for 271dupA and a missense mutation, 616C>T. In patients who are compound heterozygotic for c.271dupA and a missense mutation, the late onset variant of disease is frequently seen [2,8], suggesting that the transcripts containing these missense alleles are translated into proteins with residual function. In the cohorts reported, only a few individuals with early onset methylmalonic aciduria and homocystinuria, cblC type, were compound heterozygotic for the 271dupA mutation and a missense mutation [2,9].

The c.616C>T (p.Arg206Trp) missense mutation found in the present case has been described previously only in

a homozygous Turkish patient with an early onset form of the disease [2]. To our knowledge, the present report is the first publication of a case with compound heterozygosity for the 271dupA and 616C>T mutations in the *MMACHC* gene. Clinical heterogeneity and variability in phenotype and age of onset of disease may also be related to the presence of polymorphisms or mutations in other genes in the cobalamin pathway, or to *cis*-acting polymorphisms or *trans*-acting factors interacting with the mutant form of protein and environmental factors (e.g., diet). Moreover, individuals with later onset of the disease may have developed mild symptoms that went unrecognized for some time before the formal diagnosis [2,9].

In the present case, magnetic resonance imaging at the age of 7 months indicated delayed myelination, and electroneurographic studies revealed the presence of a demyelinating neuropathy. White matter damage and demyelination in central nervous system is a well established feature of methylmalonic aciduria and homocystinuria, cblC type, and cranial magnetic resonance imaging typically reveals diffuse supratentorial white matter swelling at presentation and a variably severe white matter loss at later stages of the disease, sometimes associated with delayed myelination [10]. In contrast, peripheral neuropathy is more frequently reported in the late onset form, and never in the early onset form. No detailed neurophysiologic studies are available for the late onset cases reported; nerve biopsy specimens display demyelinating [11], axonal [12], and mixed axonal and demyelinating peripheral neuropathy [13]. Although in the present case the lack of nerve biopsy complicates the precise diagnosis, electrophysiologic study indicated a sensorimotor demyelinating polyneuropathy. Despite slowing, the absence of temporal dispersion probably indicates a homogeneity of the pathologic process at this stage of the disease (7 months of age). Moreover, sensory nerve action potential was absent in the lower limbs, indicating a major involvement of sensory nerves at onset.

Several hypotheses have been put forward to explain the underlying mechanism of white matter involvement in methylmalonic aciduria and homocystinuria, cblC type. Disease results from combined impaired synthesis of adenosylcobalamin and methylcobalamin, the latter leading to decreased activity of methionine synthase and a subsequent reduced availability of methionine for conversion to the universal methyl donor *S*-adenosylmethionine [14]. Myelin basic protein, a major constituent in central and peripheral nervous system myelin, is believed to participate in the tight compaction of myelin sheath through association with acidic lipids on the cytosolic side of the membrane bilayer. Myelin basic protein is methylated by a specific methyltransferase requiring *S*-adenosylmethionine as the methyl donor [15]. A decrease in the methylation of myelin basic protein could cause conformational change and splitting of the myelin sheet, leading to peripheral demyelinating neuropathy. An alternative hypothesis is a reduction in the methylation of lipids required for maintenance of the myelin sheet [14]. Further potential mechanisms for

neurotoxicity and white matter involvement have been related to homocysteine accumulation, which is responsible for vascular endothelial damage or direct neurotoxicity.

In conclusion, clinical, biochemical, neurophysiologic, and molecular findings have been presented for a patient with early onset methylmalonic aciduria and homocystinuria, cblC type, who was compound heterozygous for the 271dupA and 616C>T mutations in the *MMACHC* gene; the case was complicated by peripheral sensorimotor demyelinating neuropathy. The present findings expand the clinical spectrum of early onset methylmalonic aciduria and homocystinuria, cblC type, to include early onset sensorimotor demyelinating neuropathy among the neurologic symptoms. Further studies into disease mechanisms are warranted, to better understand the phenotypic presentation and to clarify genotype-phenotype correlations for this disorder.

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