

Pearls & Oysters: Familial epileptic encephalopathy due to metylenetetrahydrofolate reductase deficiency

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PEARLS

- Metylenetetrahydrofolate reductase (MTHFR) deficiency is a rare cause of epileptic encephalopathy.
- The finding of highly elevated plasmatic homocysteine raises the suspicion of MTHFR deficiency.
- Genetic testing for MTHFR deficiency will help differentiate the condition from other homocysteine remethylation defects.

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- The same MTHFR genotype is not strictly predictive of the clinical phenotype and outcome.

Metylenetetrahydrofolate reductase (MTHFR) deficiency is a rare autosomal recessive disease included in the group of homocysteine remethylation disorders. The MTHFR catalyzes the irreversible conversion of 5,10-metylenetetrahydrofolate to 5-methyltetrahydrofolate. Besides the well-known association of thrombophilic defects with *MTHFR* variants and elevated homocysteine, few patients with *MTHFR* mutations have been described, all presenting with neurologic defects, including developmental delay, seizures, progressive neurologic deterioration, and central respiratory failure, eventually evolving into coma.¹ How the enzyme deficiency results in the development of the seizure disorder is unclear.²

We describe 3 patients, 2 siblings and their first-degree cousin, who presented with neurologic symptoms of variable severity leading to the suspicion of MTHFR deficiency. The diagnosis was hypothesized after the detection of increased plasma and urine homocysteine and was confirmed in 2 patients by *MTHFR* gene sequencing. Notwithstanding the same *MTHFR* gene mutations, the severity of manifestations and age at onset of epileptic encephalopathy varied greatly.

CASE REPORT We assessed 3 patients from the same pedigree with epileptic encephalopathy: P1

(female) and 2 siblings P2 and P3 (male and female, respectively) (figure 1). They were all born from non-consanguineous parents after an uneventful pregnancy and delivery. The mother of P1 was the monozygotic twin sister of the mother of P2 and P3.

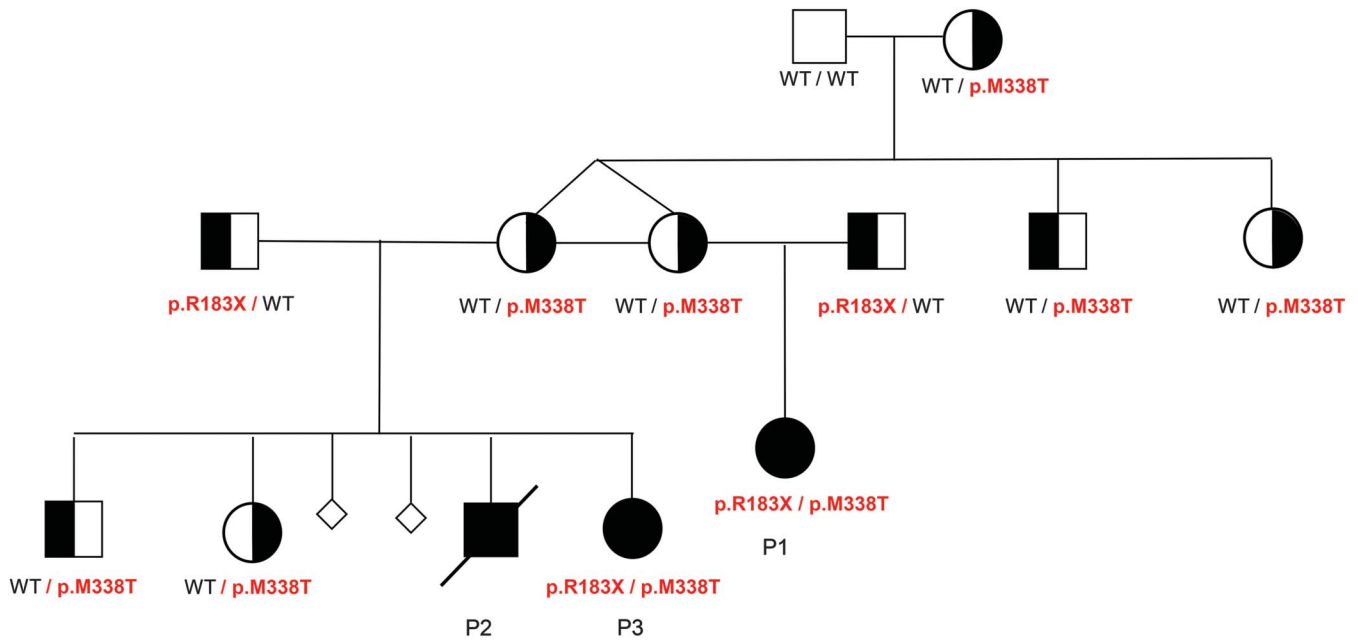
All patients presented within the newborn period with poor sucking, hypotonia, microcephaly, and lethargy. P2, the oldest patient, came to our attention at age 1 month because of infantile spasms later evolving into intractable generalized seizures. His brain MRI showed cerebral atrophy with moderate dilation of lateral ventricles and subarachnoid spaces as well as an abnormal sulcation and gyral pattern in fronto-basal areas. His neurologic condition worsened over time until he became lethargic and comatose, and died from respiratory failure at age 7 months. P1 and P3 were admitted to our department 1 year and 3 years later, respectively, both because of infantile spasms that began at 18 months in the former and at 10 months in the latter. EEGs showed a typical hypsarrhythmic pattern characterized by a chaotic succession of very high-amplitude slow waves randomly interspersed with multifocal asynchronous spikes and sharp waves. P1 responded poorly to initial treatment with ACTH, possibly because of recurrent bacterial infections. Valproic acid and lamotrigine were added and seizures were partially controlled for 2 months. Then, seizures again increased in frequency, necessitating the addition of topiramate. Because of recurrent infections, P3 was initially treated with vigabatrin, then with valproic acid, and finally with topiramate (10 mg/kg/d). Topiramate led to cessation of seizures in both patients. P1 and P3 have been taking topiramate for 36 and 18 months, respectively, and have been seizure-free during that time.

P1 and P3 showed developmental delay with different levels of severity. P1 was able to walk independently at 5 years of age despite reduced muscle tone; she was amiable and sociable but unable to say any words. P3, at 1 year of age, showed severe psychomotor retardation and nonachievement of developmental

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Figure 1 Family pedigree



P1 = patient 1; P2 = patient 2; P3 = patient 3.

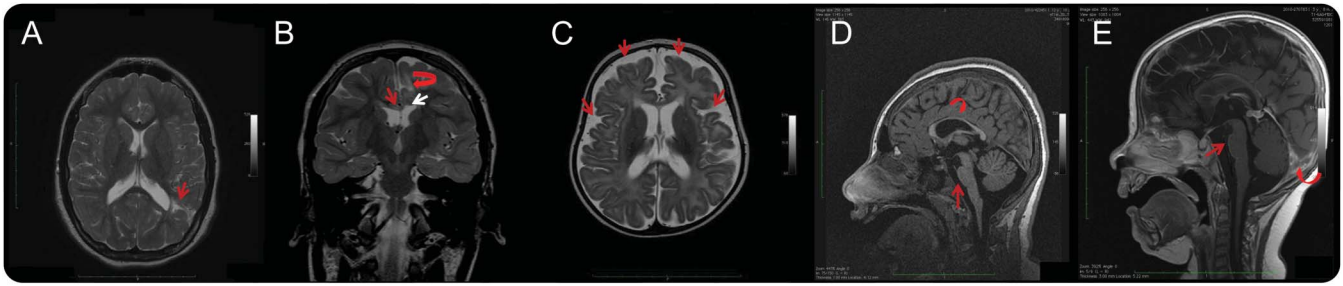
milestones; she also had severe hypotonia with complete head lag and was unable to keep a sitting or standing position. Moreover, at 2 years of age, she needed a tracheostomy after acute respiratory arrest. Brain MRI of both P1 and P3 showed diffuse white matter hyperintensity, dilation of the supratentorial and sub-arachnoidal spaces, hypoplasia of cerebellum and brainstem, and corpus callosum thinning. A left temporal arachnoidal cyst and 2 ischemic areas were also detected in P1 while severe cerebral atrophy especially over the temporal and frontal lobes was evident in P3 (figure 2). There was no history of seizure, stroke, or dementing illness in the other family members. Metabolic investigation in P1 and P3 revealed increased plasma homocysteine levels (average 180.85 $\mu\text{Mol/L}$), reduced plasma methionine levels, and no increase in urinary methylmalonic acid excretion; plasma and urine lactate were increased, whereas vitamin B₁₂ and folate levels, C3 acylcarnitine, and red cell mean corpuscular volume were within normal ranges. Given the high plasma and urine homocysteine levels, low methionine concentration, and absence of macrocytosis and methylmalonic acid, we suspected MTHFR deficiency rather than the most common cause of homocystinuria, namely, cystathionine β -synthase deficiency, which is characterized by high methionine levels.

MTHFR gene sequencing showed that P1 and P3 were compound heterozygous for mutations c.547C>T (p.R183X) and c.1013T>C (p.M338T) in exon 4 and 6, respectively (figure 1). Both mutations in the homozygous state had already been reported in patients with severe MTHFR deficiency.^{3,4} The monozygotic twin

sisters, the mothers of P1 and P3, were heterozygous carriers for the c.1013T>C (p.M338T) mutation, whereas the fathers, who were known to be unrelated to each other, were heterozygous for c.547C>T (p.R183X). Moreover, patients P1 and P3 were heterozygous for the common polymorphism c.677C>T (p.A222V), a genetic risk factor for vascular disease, inherited from their mothers in *cis* with the mutation. P2 was highly suspected of having MTHFR deficiency since he had epileptic encephalopathy with low methionine levels and homocystinuria. Genetic analysis was extended to the siblings of P2 and P3, and to their maternal uncles and maternal grandparents, all of whom, except the maternal grandfather, were carriers of the disease (figure 1).

Once the biochemical diagnosis was made, P1 and P3 were started on folinic acid (1.55 mg/kg/d) and betaine (100 mg/kg/d) therapy.⁵ A slight reduction of plasma homocysteine was obtained after 6 months of therapy, and levels returned to baseline after 18 months of therapy. Some slight improvement in muscle tone was noted during treatment.

DISCUSSION MTHFR deficiency is the most common inborn error of folate metabolism. Besides the metabolic effects, this disorder is associated with a broad spectrum of clinical symptoms: namely, psychomotor retardation, seizures, and progressive neurologic deterioration, as well as myelopathy, psychiatric illness, and cerebral vascular events in older affected individuals.¹ The metabolic diagnosis of this disorder is based on increased plasma homocysteine,



(A) Patient 1. Axial turbo spin echo (TSE) T2-weighted image: thinning and hyperintensity of the corpus callosum near the left parietal ischemic lesion (arrow), with ex vacuo dilation of the ventricles, particularly the left one. (B) Patient 1. Coronal TSE T2-weighted image: note the thinning and the hyperintensity of the corpus callosum (white arrow) near the left frontal and cingulate gyrus ischemic lesion (curved arrow). It extends also to the right side (red arrow), probably related to a partial transcallosal secondary axonal degeneration. (C) Patient 3. Axial TSE T2-weighted image: marked volume reduction of the temporal and frontal lobes (arrows) and diffuse hyperintensity of the white matter related to delayed myelination is shown. (D) Patient 1. Sagittal 3D turbo field echo T1-weighted image: subtle hypoplasia of the pons (arrow) and of the inferior part of the vermis is present. Thinning of the corpus callosum is also evident (curved arrow). (E) Patient 3. Sagittal spin echo T1-weighted image with endovenous contrast medium: note the reduction of the cranio-caudal extension of the pons due to hypoplasia (arrow). Thinning of the corpus callosum is also present. Bleeding in the dural space around the torcula of Herophili is appreciable (curved arrow). Note the enhancement of the dural walls peripheral to the bleeding with contrast administration.

low plasma methionine, and absence of megaloblastic anemia and methylmalonic aciduria. Treatment is targeted toward lowering plasma homocysteine levels and includes oral betaine, methionine, folic acid, B₁₂ vitamin, and 5-methyltetrahydrofolate, which favor homocysteine methylation processes, thereby increasing methionine levels.

In P1 and P3, therapy with folinic acid and betaine was started as soon as the condition was clinically suspected, but it induced only a slight clinical and biochemical improvement. The poor response to treatment may be due to the delayed start of treatment and the coexistence of 2 very harmful mutations, which, together with the p.A222V polymorphism, are known to reduce MTHFR enzyme activity.⁴ Failure of treatment with betaine and folic acid has been reported in 2 other patients with severe MTHFR deficiency.¹

Only one other case of multiple drug-resistant epileptic encephalopathy has been reported.² The epileptic encephalopathy in MTHFR deficiency has been attributed to homocysteine-intermediary metabolites (Hcy-thiolactone, S-nitroso-Hcy, Hcy-disulfides, homocystic acid, and adenosyl-Hcy) that antagonize GABA receptors and activate glutamate-mediated excitotoxicity in the CNS.² The good seizure control in the 2 patients (P1 and P3) treated with topiramate may be due to its capability to counteract the pleiotropic neurobiologic toxic effect of hyperhomocysteinemia.⁶ Although targeted treatment for infantile spasms in MTHFR deficiency has yet to be established, we suggest a trial of topiramate on the basis of the experience with our 2 patients.

Brain imaging findings in MTHFR deficiency are relatively nonspecific and include diffuse cerebral atrophy, cerebellar and brainstem hypoplasia, and

dismyelination.^{7,8} Our 3 cases displayed similar findings, albeit with different degrees of severity. The reduction of brain volume and dysmyelination were more pronounced in P2 and P3 than in P1 (figure 2, A–E). Furthermore, there were 2 ischemic lesions—one in the left parietal lobe and one in the frontal lobe—in P1, which underscores the importance of inactivating *MTHFR* mutations in determining thrombotic events (figure 2A).

Despite the same *MTHFR* mutations and a similar genetic background, the severity of clinical manifestations varied greatly among our patients. P2 and P3 had a more severe form of MTHFR deficiency compared to P1. An earlier onset of the epileptic encephalopathy might be related to a worse prognosis. Environmental factors such as lack of dietary supplements, infections, the use of nitrous oxide in anesthetic practice, and long-term use of antiepileptic drugs could aggravate the condition. Recent evidence suggests that Hcy-thiolactone, a specific homocysteine-derived metabolite, is neurotoxic in vivo. The bleomycin hydrolase and paraxonase 1 enzymes are involved in Hcy-thiolactone catabolism and their diminished function may enhance Hcy-thiolactone neurotoxicity.⁹ The different catabolism of Hcy-derived neurotoxic species might contribute to the variable severity of neurologic manifestations among the patients described. However, differences in epistatic genes may also contribute to different phenotypes.

The neurotoxicity in severe MTHFR deficiency seems to be multifactorial in nature; indeed, it depends on such coexisting variables as environmental factors that worsen homocysteine levels, enzymatic defects in various steps of the remethylation pathway,

different catabolism of reactive homocysteine products, intrinsic neuronal excitability, and different age-dependent vulnerability of the CNS. Therefore, hyperhomocysteinemia may be considered a trigger and not a primary cause of infantile spasms because it increases the susceptibility of the CNS to other independent damaging factors.¹⁰ MTHFR deficiency is an important diagnostic possibility in the differential diagnosis of infantile epileptic encephalopathy. A rapid diagnosis, for which genetic analysis is instrumental, and specific interventions might result in a more favorable outcome.

AUTHOR CONTRIBUTIONS

G. Cappuccio: drafting the manuscript, including medical writing for content, study design. C. Cozzolino, R. Romanelli: analysis and interpretation of genetic data. A. D'Amico, B. Carotenuto: analysis and interpretation of imaging in vivo data of MRI. E. Del Giudice, G. Parenti: drafting/revising the manuscript, including medical writing for content, study concept, supervision and coordination of clinical aspects. F. Salvatore, G. Frisso: drafting/revising the manuscript, including genetic writing for content, study concept, study supervision and coordination.

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DISCLOSURE

The authors report no disclosures relevant to the manuscript. Go to Neurology.org for full disclosures.

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