Thiamine Deficiency in Childhood With Attention to Genetic Causes: Survival and Outcome Predictors

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Primary and secondary conditions leading to thiamine deficiency have overlapping features in children, presenting with acute episodes of encephalopathy, bilateral symmetric brain lesions, and high excretion of organic acids that are specific of thiamine-dependent mitochondrial enzymes, mainly lactate, alpha-ketoglutarate, and branched chain keto-acids. Undiagnosed and untreated thiamine deficiencies are often fatal or lead to severe sequelae. Herein, we describe the clinical and genetic characterization of 79 patients with inherited thiamine defects causing encephalopathy in childhood, identifying outcome predictors in patients with pathogenic SLC19A3 variants, the most common genetic etiology. We propose diagnostic criteria that will aid clinicians to establish a faster and accurate diagnosis so that early vitamin supplementation is considered.

Thiamine or vitamin B1 is a critical cofactor involved in energy metabolism and in the synthesis of nucleic acids, antioxidants, lipids, and neurotransmitters.1,2 Thiamine is a water-soluble essential nutrient obtained from cereals, meat, eggs, legumes, and vegetables. In the absence of adequate thiamine intake, limited tissue storage may be depleted in 4 to 6 weeks.3 Thiamine requires specific transporters for the absorption in the small intestine and for cellular and mitochondrial uptake (thiamine transporter-1, encoded by SLC19A2, thiamine transporter-2, encoded by SLC19A3, and mitochondrial thiamine diphosphate carrier, encoded by SLC25A19). Within the cellular compartment, thiamine is converted into thiamine diphosphate by thiamine phosphokinase (TPK1), the metabolically active form of thiamine, which acts as a cofactor of several thiamine-dependent enzymes.

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in the cytosol, peroxisomes, and mitochondria (Fig 1).

Specifically, in the mitochondria, thiamine diphosphate (TDP) acts as a cofactor of the PDHc (pyruvate dehydrogenase complex), OGDHC (oxoglutarate dehydrogenase complex), and BCODC (branched chain 2-oxo acid dehydrogenase complex).

Infantile beriberi and Wernicke's encephalopathy are rare life-threatening and reversible causes of secondary thiamine deficiency that still occur in vulnerable populations. Infantile beriberi presents in infants breastfed by mothers with inadequate intake of thiamine or receiving low thiamine-content formula, whereas Wernicke's encephalopathy is described in sick children that undergo medical or surgical procedures such as gastrointestinal resections, parenteral nutrition, chemotherapy, etc.

In recent years, genetic defects in thiamine transport and metabolism have been described in childhood, with overlapping clinical, biochemical, and radiological features to those observed in secondary forms, and a good response to vitamin supplementation. Well-defined clinical phenotypes have been recognized in the following defects: (1) SLC19A2 (thiamine transporter-1) causes Roger's syndrome or thiamine responsive megaloblastic anemia (OMIM 249270); (2) SLC19A3 (thiamine transporter-2; ThTR2) is responsible for biotin thiamine responsive basal ganglia disease (BTRBGD; OMIM 607483); Leigh's syndrome (LS); infantile spasms with lactic acidosis; and Wernicke-like encephalopathy; (3) TPK1 (thiamine pyrophosphokinase) and transported inside the mitochondria by the SLC25A19 (mitochondrial TDP transporter). Human TDP-dependent enzymes comprise TK (transketolase), HACL1 (2-hydroxyacyl-CoA lyase 1), PDHc (pyruvate dehydrogenase complex), OGDHC (oxoglutarate dehydrogenase complex), and BCODC (branched chain 2-oxo acid dehydrogenase complex). NADPH = nicotinamide adenine dinucleotide phosphate.
In this study, we provide a global overview of the numerous genetic and acquired etiologies of thiamine deficiency in childhood, with specific attention to inherited defects of thiamine transport and metabolism. We analyze the clinical features and long-term outcomes in a multiethnic cohort of 79 SLC19A3, SLC25A19, and TPK1 patients, evaluate how thiamine/biotin treatment modifies the natural history of SLC19A3 patients, and identify the clinical parameters that may help predict neurological outcome and guide further therapeutic interventions. We propose fundamental features to suspect inherited thiamine defects against external or secondary causes of thiamine deficiency, and suggest diagnostic criteria that will help clinicians to establish faster and accurate diagnosis so that early vitamin supplementation is considered.

Materials and Methods

Study Design

We conducted a multicenter cohort study by reviewing data from patients with inherited defects in thiamine transport and metabolism. We invited 44 investigators that had published patients with inherited thiamine defects and/or patients with LS and mitochondrial disorders in PubMed. In total, 21 investigators accepted to participate, 17 centers did not have patients to include, and, last, six colleagues did not answer or refused to participate in the study.

A systematic analysis in MEDLINE (through PubMed) was performed to search for secondary causes of thiamine deficiency. We included the following keywords: #1 beriberi, #2 Wernicke’s encephalopathy, and #3 secondary thiamine deficiency, from January 2010 to February 2017. We analyzed predisposing factors, consanguinity, clinical, biochemical and radiological features, mortality, treatment, recovery, and neurological and radiological sequelae.

Study Population

We included patients with two pathogenic variants of the SLC19A3, TPK1, and SLC25A19 genes. Patients with SLC19A2 mutations were excluded from the study, because they did not have significant involvement of the central nervous system (CNS). The responsible clinician at each collaborating center collected data via a questionnaire that consisted of 131 items including: demographic data, family and perinatal history, genetic defects, gene-related phenotype, early developmental milestones, age of disease onset, triggering events, clinical neuroimaging and biochemical data at disease onset, thiamine and biotin supplementation and follow-up. In surviving patients presenting with dystonia, disability was evaluated using part b of the Burke–Fahn–Marsden scale (BFMDS). This questionnaire evaluates the dystonic patient’s ability to perform everyday activities and has been used previously in SLC19A3 patients. One hundred twenty-nine patients with nuclear encoded complex I deficiency and 324 patients with PDHc deficiency were collected through a review of the research literature in order to perform a comparative survival analysis. The time of death of nuclear encoded complex I patients was quantified according to methods described by Ortigoza-Escobar et al. References of patients collected with PDHc deficiency included for comparative survival analysis appear in the Supplementary Material.

Standard Protocol Approvals, Registration, and Patient Consent

This study was approved by the Ethics Committee of the Hospital Sant Joan de Déu, Barcelona, Spain. Informed consent was obtained from all patients.

Statistical Analysis

Statistical analyses were performed using IBM SPSS Statistics 23 software (IBM Corp., Armonk, NY). The quantitative variables were reported either in terms of the normal distribution mean, standard error of the mean (SEM), and the range; or in terms of the median and interquartile range (IQR). The Mann–Whitney U test was applied to evaluate differences in numerical variables between groups. The chi-square test and Fisher’s exact test were used to test the association between categorical variables. Multiple logistic regression analysis was performed to further investigate the relationship between the binary response variable and potential predictors of survival. The Kaplan–Meier survival analysis was used to compare the survival rates of the SLC19A3-deficient patients, patients with PDHc, and nuclear-encoded complex I-deficient LS. Differences in survival between the groups were evaluated using the log rank test. All statistical tests were two-sided and performed at a 0.05 significance level.

Results

Inherited Thiamine Defects

We identified 70 patients with SLC19A3 disease, 4 patients with TPK1 disease, and 5 patients with SLC25A19 disease. The patients were diagnosed at 21 centers: UK (n = 4); United States, Germany, and Finland (n = 3); Saudi Arabia (n = 2); and The Netherlands, Spain, Israel, France, and Sweden (n = 1). Genotypes were established in all patients, including P76 who had a similar disease course to his sibling with TPK1 deficiency and in whom the same mutations were confirmed using residual DNA. Complete clinical data sets were available in 65 of 70 patients with the SLC19A3 mutation and in all patients with SLC25A19 and TPK1 mutations. Magnetic resonance imaging (MRI) data were available in all except 7 SLC19A3 patients who were diagnosed post-mortem (P39–P45). None of the patients were found to have additional clinical or biochemical abnormalities suggestive of other genetic diseases.
The 70 patients with SLC19A3 deficiency (mean age at assessment ± SEM, 9.5 ± 0.9 years; range, 1 month–40 years old; 36 males; 51%) were born between 1975 and 2015. Of these, 63 (90%) had been previously reported. Consanguinity was reported in 51 (73%) patients and 44 (62%) had other affected family members. Arabs formed the largest ethnic group (58 of 70, 82%; Saudi Arabian n = 41, Moroccan n = 11, Iraqi n = 3, Kurdish n = 2, and Kuwaiti n = 1), followed by white European (10 of 70, 14%; Spanish n = 3, Portuguese n = 2, German n = 2, Finnish n = 2, and Hispanic n = 1), and African/Afro-Caribbean (2 of 70, 2.8%; Supplementary Table 1).

**Clinical Phenotype**

Supplementary Table 1 summarizes the demographic, genetic, clinical, and radiological features in the entire patient cohort. The frequency of the main clinical features appears in Figure 2. Fetal distress was noted in P2, P29, and P61 and acute presentation during the newborn period (around 4 weeks of age in all cases) was reported in 9 (12%) patients. In the vast majority, the developmental milestones were average, except in 7 cases (P28, P29, P53, P57, P60, P61, and P66).

The median age at disease onset was 3 years, the range was 1 month to 34 years, and the IQR was 1 to 2.8 years. The trigger events (39 of 70 patients; 55%) were viral (n = 30) or bacterial (n = 4) infection, trauma (n = 3), profuse exercise, and vaccination (n = 1, each). Fifteen (21%) patients were classified as LS and 53 (75%) as biotin thiamine responsive basal ganglia disease (BTRBGD) attributed to their positive responses to thiamine/biotin treatment. Twins (P35 and P36) with a positive family history (siblings of P34) were identified before the onset of symptoms.

Twenty-six patients experienced more than one encephalopathic episode before the initiation of vitamin supplementation (2 episodes n = 12, 3 episodes n = 7, Supplementary Table 1).
4 episodes \( n = 4 \), and 5 or more episodes \( n = 3 \) (P32, P33, and P49)). Most episodes of neurological deterioration were triggered by infection or stress. Intensive care was required in 5 of the patients (P1, P3, P48, P68, and P69).

A minority of patients (10 of 62; 16%) had an insidious onset of symptoms characterized by psychomotor regression, hyperactivity and attention deficit, unsteadiness, ataxia, walking, or stiffness of the limbs.

Systemic features of mitochondrial disease (cardiomyopathy, cardiac conduction defects, renal tubulopathy, or facial dysmorphism) were absent in our series. P4 developed a steroid-sensitive nephrotic syndrome.

Seizures were classified according to the International League Against Epilepsy as follows: generalized seizures (\( n = 31 \)); focal seizures (\( n = 6 \), including 2 patients [P30 and P31] with epilepsia partialis continua); epileptic spasms (P64, \( n = 1 \)); and West syndrome (P48, \( n = 1 \)). Thiamine and biotin treatment controlled seizures in patients effectively, except in P38, P53, and P59, who also received antiepileptic drugs, and P49, P61, and P64, who developed drug-resistant epilepsy.

**Ancillary Testing**

Few patients showed increased cerebrospinal fluid (CSF) lactate (5 of 29 patients; mean ± SEM, 3.7 ± 1.0 mmol/l; range, 2.1–7.1; normal voiding [NV] < 2), increased blood lactate (20 of 40; mean ± SEM, 4.1 ± 0.4 mmol/l; range, 2.1–8.6; NV < 2), metabolic acidosis (6 of 36) and increased blood alanine (4 of 15; mean ± SEM, 666 ± 129 μmol/l; range, 450–1,037; NV < 439). The lactate/pyruvate ratio was below the normal limit in 2 patients. Increased blood lactate levels were negatively correlated with the age of onset in the SLC19A3 patients (\( p < 0.001 \)).

Abnormal organic acid profiles were recorded in 8 of 50 patients, with high excretion of isobutyric, 2-OH-isovaleric and 2,4-di-OH-butyric (P1), alphaketoglutaric (P2), lactic (P34 and P52), 3-OH-butyric (P37), 2-OH-glutaric, glutaric, succinic, and 2-keto adipic acid (P41), and 4-OH-phenyllactic (P46).

MRI and magnetic resonance spectroscopy (MRS) were available in 61 of 70 (87%) and 43 of 70 (61%) patients, respectively. All symptomatic patients had lesions at onset, involving bilateral caudate (\( n = 55 \)), putamen (\( n = 57 \)), cortico/subcortical areas of the cerebral hemispheres (\( n = 40 \)), ventromedial region of the thalamus (\( n = 38 \)), cerebellum (\( n = 23 \)), brainstem (\( n = 14 \)), periaqueductal region (\( n = 12 \)), spinal cord (\( n = 11 \)), and the globus pallidus (\( n = 6 \); Figs 2 and 3). Acute MRIs indicated swelling and chronic MRIs indicated volume loss and necrotic changes. MRS detected a lactate peak in 55% (24 of 43) patients within the affected areas. Stroke-like lesions or mammillary body lesions were not identified.

Statistical analysis showed that deceased patients had more frequent involvement of the globus pallidus and brainstem than surviving patients (3 of 15 vs 3 of 55; Mann–Whitney \( U \) test, \( p = 0.001 \); and 4 of 15 vs 10 of 55; Mann–Whitney \( U \) test, \( p = 0.009 \), respectively).

Oxidative phosphorylation (OXPHOS) activity was normal in the muscle and skin biopsies of 6 patients, with the exception of P41 who showed 56% of complex IV activity in fibroblasts. None of the patients had ragged red fibers.

**Treatment and Outcome**

Fifty-one patients received vitamin supplementation during the acute encephalopathic episode. The time from disease onset to vitamin initiation was very broad (median, 14 days; IQR, 4–180). Forty-four patients had a significant clinical recovery within hours or days of vitamin initiation: They regained alertness, improved feeding, had a better control of seizures, and gradually recovered previously acquired milestones. Four more patients showed a mild improvement, and 3 patients did not improve at all.

Fifty-five patients were alive at the time of recruitment (mean follow-up, 5.2 ± 0.7 years; range, 2 weeks–22 years). All of them received thiamine (thiamine hydrochloride; mean dose, 20 mg/kg/day; range, 5–55) and 47 received biotin (mean dose, 5 mg/kg/day; range, 1–30). Both vitamins were administered orally in most patients, although some patients received intravenous supplementation in the acute episode. The neurological examination was normal in 26 patients at the time of assessment and they were symptom-free, whereas 27 had developed some neurological sequelae (Supplementary Table 1). No further decompensating episodes of encephalopathy, dystonia, or other neurological symptoms were recorded after vitamin supplementation in these patients, except for P61 who received inadequate vitamin doses. Additionally, blood alanine levels and the organic acid profiles in urine were normal in patients receiving vitamin supplementation. Only P52 had slightly elevated blood lactate (2.3 mmol/l). The twins (P35 and P36) who were treated presymptomatically with thiamine alone were symptom-free at the time of assessment (5 years).

Fifteen patients (21%) died; the majority of them from central respiratory failure (6.1 ± 1.9 years at death; range, 4 weeks–20 years). Deceased patients were younger at onset compared to surviving patients (mean age ±
Among the 15 deceased patients, 4 had received vitamin supplementation (P1, P30, P41, and P49).

**Disability Score**

The BFMDS questionnaire was administered to 34 SLC19A3 patients with dystonia (9.8 ± 1.8 points [mean ± SEM]; range, 0–30). Higher BFMDS scores were identified in patients with a previous history of developmental delay (19.5 ± 4.1 vs 7.7 ± 1.7; Mann–Whitney U test, $p = 0.017$) and in patients with disease onset before 6 months of age (23.7 ± 2.8 vs 7.9 ± 1.7; Mann–Whitney U test, $p = 0.01$). A positive, and almost significant, correlation was observed between the BFMDS scores and the time from disease onset to thiamine initiation (Pearson correlation, $r = 0.340; p = 0.053$; Fig 4D).

**Survival Analysis**

In the Kaplan–Meier analysis (Fig 4), treated SLC19A3 patients had a longer mean survival length than non-treated patients (A; 28.99 vs 17.23 years; log rank test, $p < 0.0001$). Additionally, mean survival length was longer in homozygous c.1264A>G SLC19A3 patients than in patients with other mutations (B; 29.88 vs 15.52 years; log rank test, $p < 0.0001$). Homozygous c.1264A>G patients were comparable to patients with other mutations with respect to age at disease onset and age at treatment initiation. However, a significant difference was observed between both groups in the number of treated patients (39 of 44 [88%] treated c.1264A>G patients vs 13 of 22 [59%] treated patients with other mutations; $p = 0.006$). Mean survival length was longer in the 70 SLC19A3 patients than in 129 patients with nuclear-encoded complex I deficiency (C; 28.0 vs 11.5 years; log rank test, $p < 0.001$). Similar results were obtained.
when the SLC19A3 and Complex I patients were divided into two age groups: those with disease onset before 6 months (11.3 vs 2.9 years; log rank test, $p < 0.0001$) and those with a later onset (33.3 vs 22.6 years; log rank test, $p = 0.003$). No significant differences were observed in the mean survival length between 70 SLC19A3 and 324 PDHc patients in both age groups. However, an almost significant difference was observed when selecting male PDHA1 patients.

**SLC19A3 Gene Mutations**

We identified 13 SLC19A3 pathogenic mutations (Supplementary Table 1; Fig 4) in the 70 patients. Five of these mutations were novel (c.91T>G, c.157A>G, c.503_505delCGT, c.516_delC, and c.833T>C). Fifty-nine patients were homozygous for the following missense mutations: c.1264A>G (n = 47); c.20C>A (n = 7); c.157A>G (n = 3); c.68G>T (n = 1); and c.541T>C (n = 1). Eleven patients were compound heterozygotes. The most frequently occurring mutation in our cohort, c.1264A>G, was present in patients with Arab ethnic backgrounds, including Saudi Arabian, Moroccan, Kurdish, and Kuwaiti patients. The next most common mutation, c.20C>A, occurred exclusively in subjects from the province of Al Hoceima in Northern Morocco (n = 7; 3 pedigrees). A splice mutation, c.980-14A>G, was observed in 5 compound heterozygote individuals, all of them of white European origin.

**SLC25A19.** We recruited 4 consanguineous Arabic patients from Israel (homozygous for c.373G>A)\(^\text{12}\) and a new white European German patient diagnosed by our group (P75). The phenotype of this girl, aged 21 years, was similar to previously reported cases (Supplementary Table 1). The symptoms

![FIGURE 4: The figure shows Kaplan-Meier survival curves (A, B, C) and the correlation between the Burke-Fahn-Marsden Disability Scale and the time elapsed between disease onset and thiamine supplementation in SLC19A3 patients (D). (A) Comparison between treated (n = 51) vs untreated (n = 19) SLC19A3-deficient patients (log rank test, $p < 0.0001$). (B) Comparison between c.1264A>G homozygous mutation (n = 44; 39 [88%] treated patients) vs other mutations (n = 22; 13 [59%] treated patients) in SLC19A3-deficient patients (log rank test, $p < 0.0001$). (C) Comparison between SLC19A3 patients (n = 70), nuclear-encoded complex I deficient Leigh syndrome (n = 129), and male PDHc-deficient patients attributed to PDHA1 deficiency (n = 145). When comparing SLC19A3 and nuclear-encoded complex I deficient Leigh syndrome, differences reach statistical significance (log rank test, $p < 0.001$). When comparing SLC19A3 and male PDHc patients, differences did not reach statistically significance (log rank test, $p = 0.06$). (D) Correlation between the BFMD5 (y-axis) and the time elapsed between disease onset and thiamine supplementation (x-axis, days, log-scale) in SLC19A3 patients ($r = 0.34$; $p = 0.053$).](image-url)
were triggered by febrile illness between 20 months and 6.5 years and consisted of acute encephalopathy, dysarthria, and episodic flaccid weakness. They had elevated levels of lactate in the CSF at onset (2.9–4.2mmol/l; NV < 2), but normal systemic mitochondrial biomarkers. MRI showed T2 hyperintensity and necrosis in the caudate and putamen in all patients. Additionally, P74 had T2 hyperintensity (cavitated lesion) in the medial thalamus.

Thiamine treatment (400–600mg/day) administered 3 years after onset led to substantial improvement in peripheral neuropathy and gait in P71 and P72, whereas P74 and P73, treated 9 and 12 years later, respectively, continued to have significant ambulatory impairment. All patients are currently alive. They have mild-to-severe bilateral pes equinus, axonal polyneuropathy, and dystonia. The disease severity was clearly milder in P71, P72, and P75, supporting the efficacy of thiamine treatment in preventing further disease progression.

Interestingly, P73 stopped thiamine treatment because of an apparent lack of benefit. Four years later, a severe episode of flaccid paralysis occurred with fever, and intravenous thiamine (1.500mg) led to clinical recovery within 24 hours.

**TPK1.** Four **TPK1** patients (P76, P77, and P79, previously reported)\(^{17,18}\) suffered from LS triggered by febrile illness between 1 month and 2.5 years of age (Supplementary Table 1). Brain lesions developed in the cerebellum and dentate nuclei (n = 4), striatum (n = 3), thalamus, globus pallidus (n = 2), brainstem, and spinal cord (n = 1). P76 showed lactic acidosis (3.0mmol/L; NV < 1.77) whereas none had increased lactic acid in the CSF. Increased excretion of organic acids was recorded in 3 of 4 patients: lactic acid (P76), glutaric acid (P77), and mildly increased alpha-ketoglutaric acid and dicarboxylic acid (P79). P76 and P78, who did not receive thiamine supplementation, died at the age of 29 and 6 months, respectively. P77 and P79 are currently aged 4 and 7 years, respectively. P77 receives a combination of thiamine (15mg/kg/day), biotin (1mg/kg/day; P77), and ketogenic diet, and P79 receives thiamine (500mg/day) alone. Both patients show severe neurological sequelae, with spasticity, hypotonia, dystonia, developmental delay, and high scores on the BFMDS (27 and 14, respectively).

**Secondary Thiamine Deficiency**

A total of 153 patients (beriberi, N = 88; Wernicke’s encephalopathy, N = 65) were collected, aged between 2 weeks and 17 years of life.\(^4,19–42\) A summary of the main characteristic features collected in our database is provided in Figure 6. Predisposing factors were reported in...
100% of cases. Systemic, cardiovascular, and respiratory symptoms were recorded in all patients with beriberi and a few cases with Wernicke encephalopathy. The majority of cases showed increased blood (53 of 55; 96%) and CSF lactate (12 of 13; 92%) and metabolic acidosis (17 of 34; 50%). Low blood thiamine levels were detected in 100% of cases that were analyzed. The thalamus, periaqueductal region, and mammillary bodies were more frequently involved, as opposed to patients with inherited thiamine defects, who showed more-frequent alterations in the caudate and putamen. Clinical improvement after thiamine supplementation was constant, and less than 20% showed mortality or neurological sequel.

**Discussion**

Thiamine diphosphate, the metabolically active form of thiamine, is essential for energy production in the CNS. In brain regions with high metabolic demands, thiamine deficiency attributed to exogenous (nutritional) or endogenous (genetic) defects can trigger a metabolic crisis, and Wernicke's encephalopathy is a model of cerebral thiamine deficiency. Primary and secondary conditions leading to thiamine deficiency have overlapping features in children, both presenting with acute episodes of encephalopathy, bilateral symmetric brain lesions, and biomarkers of mitochondrial dysfunction, such as lactic acid accumulation in different tissues and high excretion of organic acids. In both scenarios, early thiamine supplementation may lead to clinical recovery within a few hours or days, and brain lesions may be reversible on MRI.

In our international study group of 79 patients with inherited thiamine defects, the vast majority of children were born to consanguineous patients; they presented between the age of 1 and 6 years in the context of a febrile illness, with acute/recurrent encephalopathy, basal ganglia lesions, dystonia, hypotonia, spasticity, ataxia, and seizures. Lactic acid accumulation in the CNS was identified by MRS in half of the SLC19A3 patients, and in all SLC25A19 cases by CSF analysis, suggesting brain energy failure, as in other cases of Leigh’s encephalopathy. Also, abnormal organic acids were identified in a few SLC19A3 and TPK1 cases. These included organic acids that were specific of thiamine-
dependent mitochondrial enzymes, such as lactic acid (PDHc), alpha-ketoglutarate (OGDHC), and branched chain keto-acids (BCODC), and other nonspecific organic acids caused by generalized mitochondrial dysfunction. These biomarkers did not predict outcome in our series.

Fundamental features to suspect primary thiamine defects against secondary etiologies were the frequency of consanguineous families, the absence of predisposing factors for beriberi and Wernicke's encephalopathy, the lack of cardiovascular or respiratory features, and the sparing of mamillary bodies on MRI (Fig 6).

A characteristic feature that distinguished patients with mutation in SLC25A19 from the other genetic defects was the presence of peripheral neuropathy. This phenotype was initially described in Spiegel et al., and, to our knowledge, no further patients have been reported since then. We identified a novel missense variant in the SLC25A19 gene in a 20-year-old woman with striatal necrosis and peripheral neuropathy, thus providing clinical evidence to the recognition of this phenotype. In contrast to SLC19A3 and SLC25A19, patients with mutation in TPK1 had an earlier onset of symptoms, they showed variable response to thiamine supplementation, and developed a more-severe phenotype with higher morbidity and mortality.

Based on the frequency of the main clinical and radiological features, and on specific biomarkers, we propose diagnostic criteria for the three known inherited thiamine defects with prominent neurological involvement (Table 1). Total thiamine levels in blood are reduced in patients with secondary deficiencies, but are normal in genetic conditions, in which the quantification of thiamine isoforms, either in blood, CSF, fibroblasts, or muscle cells, is critical for the diagnosis.

More than one third of SLC19A3 patients had several recurrent encephalopathic episodes before vitamin supplementation. Most of them were born after the first description of the disease by Ozand et al in 1998, suggesting that this is an underdiagnosed disorder. Thiamine and biotin supplementation led to prompt and significant clinical recovery in most SLC19A3 patients. Survival analyses showed that the mean survival length was longer in patients who received thiamine and biotin compared to nontreated patients. No further episodes of encephalopathy, dystonia, or other neurological disturbances were noted after vitamin supplementation. Additionally, patients had effective control of seizures without the need of antiepileptic drugs and nonspecific biomarkers of mitochondrial dysfunction remained within normal limits. More important, half of treated patients had no disability at all, and neurological examination was normal at the time of assessment. A positive correlation was observed between the disability scores and the time spent between disease onset and thiamine initiation, meaning that the earlier the therapeutic intervention, the lower the sequel observed, although these data did not reach statistical significance. It was likely that the deceased patients with severe disabilities were missing relevant data, representing an important bias against a significant correlation in this analysis.

We identified clinical parameters that may help predict neurological outcomes before the initiation of vitamin supplementation. Patients with symptoms presenting within the first 6 months of life had a shorter survival curve and higher scores on the disability scale. Moreover, the distributions of brain lesions in the deceased SLC19A3 patients were more diffuse, with a significantly higher involvement of the globus pallidus and brainstem. This is in accord with previous reports that indicate a subgroup of patients with thiamine transporter 2 deficiency presenting with fatal infantile LS, who died very early in the disease course, or were left with severe neurological sequel and extensive brain injury despite the initiation of vitamin treatment. In our cohort, 4 children died despite vitamin supplementation. Pretreatment predictors of poor responses in these patients were extensive brain involvement (P49), early onset of the disease (P41), or the ocurrence of septicemia in a child receiving intensive care (P1).

Based on these data, we believe that patients presenting with LS should be treated with a vitamin cocktail including thiamine and biotin, given that the phenotype of inherited thiamine defects may be clinically indistinguishable from other genetic disorders leading to Leigh's encephalopathy. However, therapeutic interventions should be individualized in those cases presenting clinical predictors of poor neurological recovery.

SLC19A3 patients showed longer survival length than patients with complex I deficiency, with more than 60% of the SLC19A3 patients surviving at 20 years of age. Previous studies have demonstrated poor survival rates in LS patients. Factors associated with poor prognosis in LS patients were the onset of symptoms within the first 6 months, the presence of cardiomyopathy, and brainstem involvement. As opposite to these features, SLC19A3 patients in our series had a median age at disease onset of 3 years, they did not show cardiac involvement, and brainstem lesions were observed in a minority of cases. More important, thiamine supplementation prolonged survival in SLC19A3 patients, whereas no effective treatment is available for complex I deficiency patients. We did no observe differences on survival between SLC19A3 and PDHc patients; however, an almost significant difference...
was observed when selecting male patients with PDHA1 deficiency, who are known to have a poorer survival than females. PDHc patients benefit from ketogenic diet\textsuperscript{57} and thiamine supplementation,\textsuperscript{58} which reduce neurological features and the frequency of hospitalizations.

A high proportion of \textit{SLC19A3} patients in our cohort were homozygous for the missense variant, c.1264A>G, all of them belonging to the Arab ethnic group. These patients had a longer survival than patients with other mutations. The higher proportion of patients

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<th>TABLE 1. Suggested Criteria for the Diagnosis of Inherited Thiamine Defects With Prominent Neurological Involvement</th>
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<td><strong>Required</strong></td>
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<td>1. <strong>Clinical criteria</strong></td>
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<td>a. \textit{SLC19A3}: Acute or recurrent episodes of encephalopathy (decreased consciousness, irritability) with two or more of the following: (1) dystonia, (2) hypotonia, (3) bulbar dysfunction, (4) ataxia, and (5) seizures. Of note, 16% of patients may have an insidious onset of symptoms (psychomotor regression, clumsy or abnormal gait, and stiff limbs).</td>
</tr>
<tr>
<td>b. \textit{SLC25A19}: Acute or recurrent episodes of encephalopathy with: (1) progressive peripheral neuropathy or (2) severe congenital microcephaly with brain malformations.</td>
</tr>
<tr>
<td>c. \textit{TPK1}: Acute or recurrent episodes of encephalopathy, with two or more of the following: (1) dystonia, (2) hypotonia, (3) ataxia, (4) seizures, and (5) developmental delay. Of note, some patients may have a nonepisodic early-onset global developmental delay.</td>
</tr>
<tr>
<td>2. <strong>Biochemical criteria</strong></td>
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<tr>
<td>a. Normal total thiamine blood levels</td>
</tr>
<tr>
<td>b. Low free-thiamine in CSF and/or fibroblasts (\textit{SLC19A3})</td>
</tr>
<tr>
<td>c. Low TDP in blood, muscle, and/or fibroblasts (\textit{TPK1})</td>
</tr>
<tr>
<td>d. High excretion of alpha-ketoglutaric acid in urine (common in \textit{TPK1} and \textit{SLC25A19}, rare in \textit{SLC19A3}).</td>
</tr>
<tr>
<td>3. <strong>Radiological criteria</strong></td>
</tr>
<tr>
<td>a. MRI pattern compatible with Leigh’s syndrome (\textit{SLC19A3}, \textit{SLC25A19}, \textit{TPK1}) or Wernicke’s encephalopathy (\textit{SLC19A3})</td>
</tr>
<tr>
<td>i. \textit{SLC19A3}: Symmetrical T2W hyperintensity of caudate, putamen, cortico/subcortical areas, and/or ventromedial thalamus. No involvement of mammillary bodies.</td>
</tr>
<tr>
<td>ii. \textit{SLC25A19}: Symmetrical T2W hyperintensity in the caudate and putamen.</td>
</tr>
<tr>
<td>iii. \textit{TPK1}: Symmetrical T2W hyperintensity in basal ganglia and cerebellum (dentate nuclei).</td>
</tr>
<tr>
<td>4. <strong>Therapeutic criteria</strong></td>
</tr>
<tr>
<td>a. Clinical improvement after thiamine supplementation.</td>
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<tr>
<td><strong>Supportive</strong></td>
</tr>
<tr>
<td>1. Consanguinity</td>
</tr>
<tr>
<td>2. Trigger event (infection, vaccination, trauma, intense physical activity, etc.).</td>
</tr>
<tr>
<td>3. Absence of predisposing factors of beriberi or Wernicke’s encephalopathy.</td>
</tr>
<tr>
<td>5. Increased lactate in blood and/or CSF.</td>
</tr>
<tr>
<td>6. Normal OXPHOS and PDHc activity in muscle and fibroblast.</td>
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<tr>
<td>7. Increased lactate on MRS.</td>
</tr>
</tbody>
</table>

\textit{CSF} = cerebrospinal fluid; \textit{TDP} = thiamine diphosphate; \textit{MRI}, magnetic resonance imaging; \textit{T2W} = T2 weighted; \textit{OXPHOS} = oxidative phosphorylation; \textit{PDHc} = pyruvate dehydrogenase complex; \textit{MRS} = magnetic resonance spectroscopy.
receiving thiamine and biotin in the group of homozygous c.1264A>G patients compared to patients with other mutations may account for these differences. In line with this observation, transfection studies demonstrated that the c.1264A>G mutation led to a protein with null-transport activity, which is difficult to correlate with a more-benign phenotype.

Among patients from European countries, we identified 4 compound heterozygous cases with c.74dupT/c.980-14A>G that had in common a late disease onset, 3 of them in adolescence and adulthood, with an excellent response to thiamine. Likewise, a recently described patient with the same mutations presented an adult-onset subacute leukencephalopathy with an effective response to thiamine.

Our results confirm previous reports on a characteristic pattern of brain injury in inherited thiamine defects that will help in the early recognition and differential diagnosis. The majority of SLC19A3 patients showed striatal lesions in combination with other affected brain areas, including cortico/subcortical regions of the cerebral hemispheres, ventromedial thalamic nuclei, cerebellum, brainstem, periaqueductal region, spinal cord, and globus pallidus, in order of decreasing frequency. As previously mentioned, deceased patients had a greater involvement of the brainstem and globus pallidum. Lesions in the respiratory centers, located in the medulla oblongata and pons, may be responsible for acute respiratory failure, the most frequent cause of death in this cohort, as in other LS patients. Moreover, bilateral lesions in the globus pallidus were also related to severe dystonia and poor prognosis in patients with glutaric aciduria type I and in carbon monoxide intoxication. Isolated striatal necrosis was a common feature of SLC25A19 patients. In contrast, TPK1 patients showed basal ganglia lesions in combination with involvement of the cerebellum and dentate nuclei.

The small size of the sample was an important limitation for this study. Also, recruitment was restricted to published cases and individual centers, leading to possible selection bias toward more-severe presentations. These are common limitations for research in rare diseases, where the limited number of patients hampers the recognition of the full spectrum of severities. Also, a more-accurate assessment on neurodevelopment should be warranted in future studies.

In summary, this international study describes the natural history of 79 patients with inherited defects in thiamine transport and metabolism. We confirm that thiamine defects manifest with acute brain injury in the first decade of life in the vast majority of patients. We demonstrate how vitamin supplementation modifies the survival curve of SLC19A3 patients and identify statistically significant predictors of neurological outcome that may guide clinicians in further therapeutic interventions. Our study indicates a better prognosis than other causes of LS, encouraging clinicians to suspect the disease and to make an early diagnosis and accurately prescribe treatment. We also contribute with diagnostic criteria for inherited thiamine defects and help differentiate them from secondary causes of thiamine deficiency.

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Author Contributions


Potential Conflicts of Interest

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