

Whole genome sequencing identified a novel copy number variant on TPK1 gene in a Chinese family with thiamine metabolism dysfunction syndrome 5

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Abstract

Background

Thiamine metabolism dysfunction syndrome 5 (THMD5) is a rare autosomal recessive neurometabolic disorder caused by biallelic pathogenic variants in the *TPK1* gene. Copy number variations (CNVs) involving *TPK1* are extremely rare. We aimed to characterize the clinical and genetic features of two affected siblings from a Chinese family with THMD5.

Methods

The medical history, neurological examination, neuroimaging, laboratory findings, and treatments were collected from the proband and his affected older brother. Whole genome sequencing (WGS) was performed on all family members. Identified variants were validated by Sanger sequencing and interpreted according to ACMG/ClinGen guidelines.

Results

The proband, a 31-year-old male, exhibited recurrent episodic encephalopathy, ataxia, dysarthria at the age of 13, and then developed spastic gait. His older brother had an onset of more severe phenotype at the age of four, showing symmetric basal ganglia lesions and elevated serum lactate. WGS revealed compound heterozygous variants in *TPK1* including a known pathogenic missense variant and a novel tandem duplication affecting exon 7 that was predicted to cause reading frame disruption and nonsense-mediated decay. The proband received regular high-dose thiamine (100 mg/day) with stabilization of symptoms, whereas his older brother had low-dose thiamine (30 mg/day) and discontinued treatment, leading to progressive deterioration.

Conclusions

Our study expands the genetic spectrum of *TPK1* by reporting a novel CNV and highlights the diagnostic superiority of WGS in detecting CNVs. Furthermore, the divergent clinical trajectories between the two siblings underscore that high-dose and lifelong thiamine therapy is critical to prevent neurological decline in patients with THMD5.

Introduction

Thiamine metabolism dysfunction syndrome 5 (THMD5) is a rare autosomal recessive inherited metabolic disease, characterized by recurrent encephalopathy, progressive ataxia, dystonia, psychomotor retardation, and elevated lactic acid level in early childhood^[1, 2]. In severe situation, children manifested with Leigh syndrome which resulted in rapid progression of symptoms and severe disability

with high mortality^[3]. Due to less common in adult patients, the diagnosis of THMD5 is challenging in adult neurology departments. Thiamine pyrophosphokinase (TPK), encoded by *TPK1* gene, played a key role in converting thiamine into thiamine pyrophosphate (TPP), the biologically active form of thiamine which served as essential coenzyme for multiple metabolic processes including carbohydrate, fatty acids, and branched chain amino acids metabolism^[1]. Homozygous or compound heterozygous mutations in the *TPK1* gene caused TPK deficiency and thiamine metabolism dysfunction, contributed to the pathogenesis of THMD5^[4]. To date, over 30 pathogenic variants of *TPK1* gene had been reported in published literature, encompassing multiple variant types^[3, 5]. The missense variants were the most common type, accounting for the majority of reported *TPK1* gene variants. Few patients with copy number variant (CNV) of *TPK1* gene had been documented before.

Here, we report two affected siblings in a Chinese family with THMD5 caused by compound heterozygous single nucleotide variation (SNV) c.382C > T (p. Leu128Phe) and CNV (NC_000007.13: g.144285435_144289647dup) in *TPK1* gene, which expanded the spectrum of *TPK1* variants.

Methods

Subjects and clinical data collection

This study was approved by the medical ethics committee at the First Affiliated Hospital of Sun Yat-sen University and conducted according to the Declaration of Helsinki. Written informed consent was obtained from the patients.

Clinical data including demographic information, medical history, physical examination, laboratory results, electrophysiology examinations, brain magnetic resonance imaging (MRI) scanning results, cognitive function and neuropsychological assessment results from the patients were collected and analyzed. Treatments and follow-up results were also collected.

Whole genome sequencing (WGS) and validation of variants

The peripheral blood samples of proband, his affected older brother, his healthy younger brother and his parents were collected for whole genome sequencing and further sanger sequencing for verification. Whole genome sequencing was performed with the use of TruSeq DNA PCR-Free Library Preparation (Illumina) and conducted on Illumina sequencing platforms. Samples were sequenced to an average depth of 30× on human nuclear genome. Within the scope of the whole genome, at least 98% sequences achieved a minimum of 10× coverage, with no less than 90% sequences achieved a minimum of 20× coverage. The sequencing data was analysed using the Gene Analysis ToolKit (GATK) software. Fastq files were aligned to the human reference genome19 (hg19 on <https://genome.ucsc.edu/>) using BWA software. Sanger sequencing was performed to validate the single nucleotide variants found by whole genome sequencing in the proband and his family members.

Variants interpretation

The Variant Effect Predictor (VEP) software was used to interpret single nucleotide variations and small insertions/deletions. Meanwhile, variations were screened based on genetic disease databases, variation databases and large-scale population sequencing databases, such as ClinVar (<https://www.ncbi.nlm.nih.gov/clinvar/>), OMIM (<https://www.omim.org/>), HGMD (<https://www.hgmd.cf.ac.uk/>), and gnomAD (<https://gnomad.broadinstitute.org/>). Moreover, several internationally recognized computational algorithms including SIFT, PolyPhen-2, and FATHMM v2.3 were utilized to predict the pathogenicity of missense variants. SNVs and small insertions/deletions were classified in accordance with standards and guidelines for the interpretation of sequence variants recommended by the American College of Medical Genetics and Genomics (ACMG)^[6]. CNVs were interpreted and classified according to the technical standards for the interpretation and reporting of constitutional copy-number variants reported by ACMG and the Clinical Genome Resource (ClinGen)^[7].

Results

The proband was 31-year-old male patient from a non-consanguineous family (family pedigree was showed in Fig. 1A). He initially developed recurrent episodic encephalopathies with bilateral lower limbs weakness, ataxia, dysarthria and blurred vision at the age of 13. Neurological examination at that time showed decreased muscle strength in both lower limbs, nystagmus and pyramidal tract signs. His symptoms were alleviated completely after treatments with glucocorticoids. He experienced six episodes of encephalopathy during the following 6-years though complete remission was acquired with the treatment of glucocorticoids for each time. At the age of 19, he developed progressively worsen weakness and stiffness in lower limbs, accompanied with spastic gait, difficulty walking and dysarthria. The physical examination at the age of 29 revealed horizontal nystagmus of the eyeballs, dysarthria, and spastic gait during walking. The deformed bending of fingers and toes as well as mild muscle atrophy of both hands were observed (Fig. 1B). Hypertonia was observed in all limbs though muscle strength was normal (5/5). Deep tendon reflexes were hyperactive in all limbs. Positive Hoffman and Babinski signs were observed. No sensory deficits or cognitive impairments were detected. His older brother, a 33-year-old male, developed similar weakness of lower limbs, gait difficulty and dysarthria after acute infectious disease at the age of 4, and gained partial remission with treatments. However, spasticity of lower limbs, gait disturbance, dysarthria and intelligence impairment continue to deteriorate with ages in his teens. His symptoms were significantly more severe than that of the proband patient, manifesting difficulty on walking or standing independently in his thirties. The proband's parents and younger brother revealed no clinical manifestations, which indicated possible recessive inheritance pattern in the present pedigree.

Brain MRI of the proband patient demonstrated mild cerebral, cerebellar, and brainstem atrophy with slightly significant atrophy in bilateral parietal cortex (Fig. 1C). Electrophysiology examinations showed normal nerve conduction velocity, while increased duration and amplitude of motor unit action potential were detected in right quadriceps femoris and left anterior tibialis, indicating neurogenic damages on the detected muscles. The proband's serum lactate level was normal. The mini-mental state examination

scale (MMSE) scores, Montreal Cognitive Assessment scale (MoCA) scores, and Alzheimer's Disease Assessment scale- Cognitive (ADAS-Cog) scores of proband were 30/30, 26/30, and 8/70 respectively, demonstrating approximately normal cognitive function. The Neuro-psychiatric inventory (NPI) score of proband was 0, indicating absence of mental behavior damage. As for the proband's older brother, brain MRI showed typical symmetric bilateral signal alterations (hyperintensity on T2-weighted images while hypointensity on T1-weighted images) in basal ganglia, selectively involving the lenticular nucleus (Fig. 1D). In addition, atrophy on cerebral cortex and cerebellum were detected on brain MRI. Electrophysiology examinations revealed increased duration and amplitude of motor unit action potential on right gastrocnemius and anterior tibialis muscles. The serum lactate level of the proband's older brother was 3.65mmol/L, which was higher than reference value (reference ranges 1.3-2.49mmol/L). The MMSE scores, MoCA scores, and ADAS-Cog scores were 20/30, 12/30, and 21.7/70 respectively, indicating mild cognitive impairments. The NPI scores were 30, indicating emotional and behavioral damages.

WGS revealed a missense variant NM_022445.4:c.382C > T (p. Leu128Phe) and a novel CNV NC_000007.13: g.144285435_144289647dup in *TPK1* on the proband patient and his older brother. The missense variant, c.382C > T (p. Leu128Phe), is absent or at a low frequency in several global human genome databases such as Exome Sequencing Project, 1000 Genomes Project, Exome Aggregation Consortium, and gnomAD. This variant had been reported and predicted to be pathogenic in previous studies^[8, 9]. Moreover, the variant p. Leu128Phe was suggested to cause *TPK1* deficiency in a published literature^[10]. Sanger sequencing determined the variant c.382C > T (p. Leu128Phe) in the proband patient, and this variant was identified in his older brother, his mother, and his younger brother except his father (Fig. 2). According to the 2015 ACMG guidelines, the variant c.382C > T was classified as "pathogenic" (PS1 + PS3 + PM2 + PM3 + PP1). The CNV, NC_000007.13: g.144285435_144289647dup, is a 4.2 kb tandem repeated sequences consisting of the exon 7 and its adjacent introns of *TPK1* gene located in the chromosome 7q35. By visually inspecting the duplicative region at the *TPK1* locus in the proband's, proband's father's, proband's mother's, proband's older brother's and proband's younger brother's WGS data, we identified that the split-read sequences of the breaking points at left and right ends of the duplicative region were identical completely and matched in the proband, proband's father and proband's older brother (Fig. 3), suggesting duplication was in tandem. The occurrence of reading frame disruption and non-stop decay (NMD) was predicted. According to the 2020 ACMG and ClinGen CNV interpretation guidelines, the CNV NC_000007.13: g.144285435_144289647dup was classified as "pathogenic" (score 1.2 points).

Based on the clinical characteristics, brain MRI features, family-based linkage analysis, and genetic analysis results, the proband and his older brother were diagnosed with THMD5. For the proband, vitamin B1(100mg/day) was administered regularly though improvement of symptoms like gait disturbance, spasticity of limbs, and dysarthria were not observed. Otherwise, the proband's older brother received treatment of vitamin B1 (30mg/day) for a short period and subsequently refused to

attend our clinic and stopped supplement of vitamin B1. His spasticity on lower limbs and gait difficulty progressed gradually.

Discussion

This study presented two siblings of THMD5 in a Chinese family caused by compound heterozygous variants in the *TPK1* gene, identified via WGS analysis. The genetic architecture in this pedigree consisted of a previously reported pathogenic missense variant (c.382C > T, p. Leu128Phe) and a novel CNV, predicted to result in a tandem duplication of exon 7 and adjacent introns in the second *TPK1* allele. Our report expanded the genetic spectrum of *TPK1* gene mutation associated THMD5 and provided insight on analyzing genotype-phenotype correlations of this disease. We also reinforced the diagnostic superiority of WGS in identifying complex genetic lesions like chromosome structure variations and non-coding region variations.

Thiamine, also known as vitamin B1, is an essential water-soluble vitamin for human beings and involved in energy metabolism in brain tissue as a key cofactor. There are four known genes (*SLC19A2*, *SLC19A3*, *SLC25A19*, and *TPK1*) involved in the transport and metabolism of thiamine, and the defect of each gene is related to diverse phenotype^[11]. The *TPK1* gene is located on chromosome 7q35 and contains nine exons. It encodes thiamine pyrophosphokinase (TPK) protein, which consists of 243 amino acids and catalyzes the conversion of thiamine to its active form, thiamine pyrophosphate (TPP) in cytoplasm^[12]. Genetic defects of *TPK1* results in THMD5 with a wide range of disease phenotypes include severe Leigh syndrome and mild recurrent ataxia^[3]. To date, over 30 pathogenic variants which distribute across all exons without a single predominant hotspot have been identified, and the missense variants are the most common variants, followed by nonsense, frameshift, and splice-site variants. In 2011, Mayr et al. first reported five individuals with *TPK1* gene variations from three different families and showed a reduced concentration of TPP in the muscle and blood of patients. Furthermore, immunoblot analysis revealed that the TPK protein levels were decreased in the muscle extracts of patients^[1]. Biochemical studies in a previous publication showed that variation c.479 C > T (p.Ser160Leu) likely change the dimension of TPK protein thereby possibly affecting its enzyme activity^[2]. However, study results from Huang et al. indicated that variants like p.Ser160Leu, p.Trp202Gly, and p.Ser28Leu were likely to disrupt thiamine binding rather than decrease enzymatic activity of, providing new insights on potential mechanism underlining THMD5^[13]. The missense variant c.382 G > A (p. Leu128Phe), identified in our family, was reported in three previous publications and proven to be pathogenic^[8–10]. This variant had been confirmed to be located in the thiamine binding domain in a highly conserved amino acid^[9]. Functional studies on variant p. Leu128Phe indicated that it severely impaired the enzymatic activity of *TPK1*, by reducing catalytic efficiency about 30.9% of wild-type^[10]. Large duplications or deletions affecting *TPK1* gene have been rarely documented but are increasingly recognized with comprehensive genomic analyses. The novel CNV identified in our family, containing tandem duplication of the exon 7 and adjacent introns on *TPK1* gene, expanded the genetic spectrum. This CNV is predicted to result in NMD with partial or complete loss of function on TPK1 protein, probably cause a severe biochemical

defect. However, further biochemical and functional studies are required to validate this hypothesis. Noteworthy, the majority of reported *TPK1* gene variants were identified via whole exome sequencing (WES) or targeted gene panels. While WES or targeted gene panels are excellent on detecting SNVs and small indels within selective capturing of target regions, they have limitations in sufficiently covering coding exons and reliably detecting larger CNVs, particularly those that are intragenic or having breakpoints in non-coding regions^[14]. WGS offers a comprehensive, single-assay solution by providing complete coverage of the coding region across the entire genome, enabling the simultaneous detection of point mutations, small indels, and CNVs with high resolution^[15]. It can precisely map CNV breakpoints and determine *cis/trans* phasing of compound heterozygous variants, which were crucial in our cases.

Recently, 32 patients of THMD5 with biallelic variants in *TPK1* have been well documented in multiple literatures^[1–3, 5, 8–10, 13, 14, 16–25]. The age of onset for most patients was predominantly in infancy or early childhood, with ages ranging from 1 month to 7 years old. The most common clinical presentations were recurrent episodes of encephalopathy often triggered by acute infectious disease, gait ataxia, and dystonia, which presented in over or approximately half of all patients. Other symptoms and signs like dysarthria, seizures, nystagmus, spasticity and psychomotor regression were also observed in partial cases. Few patients manifested as loss of hearing or optic atrophy. In our study, the proband and his affected sibling manifested as recurrent episodes of encephalopathy, ataxia, dysarthria, nystagmus and limbs spasticity, which aligned robustly with the established THMD5 phenotype though the onset age in the proband was later than that of average reported. The brain MRI manifestation included a wide spectrum ranging from bilateral, symmetric signal abnormalities (T2/FLAIR hyperintensity) in the basal ganglia, the cerebellum and brainstem to normal neuroimaging. Specifically, the involvement of bilateral putamen and caudate nuclei is highly characteristic in patients with THMD5. These lesions place THMD5 firmly within the neuroimaging spectrum of Leigh syndrome. Biochemical studies in previous reports revealed elevated lactic acid in serum and cerebrospinal fluid especially during episodes of encephalopathy. Notably, the proband's older brother in our study showed significant symmetric lesions in the putamen and the elevation of serum lactate level was observed, while the proband patient lacked typical brain MRI manifestation and the serum lactate level was normal. It was noticeable that the symptoms and brain MRI changes of the proband's older brother were more severe than his younger sibling, suggesting possible clinical phenotypic heterogeneity on patients with THMD5. In a previous study, Li et al. also observed that two affected siblings in a Chinese family with THMD5 showed different disease progression and treatment outcomes^[16]. The proband in our study was initially misdiagnosed as recurrent encephalomyelitis and received glucocorticoids treatment for several times. However, the childhood onset, progressive development of symptoms in the late second decade and positive family history lead to the suspect of inherited metabolic disease, and genetic analysis was performed to confirm the diagnosis of THMD5.

The therapeutic efficacy of thiamine in THMD5 has been consistently demonstrated across multiple case series and individual reports^[2, 4, 16]. Patients who received high-dose thiamine (typically 100–300 mg/day or 10–30 mg/kg/day) showed marked reduction in the frequency and severity of acute

metabolic crises, with stabilization or even improvement of neurological function^[5, 8]. In a comprehensive analysis of 28 reported patients, Zhao et al. observed that among those treated with thiamine, approximately 70% demonstrated significant clinical improvement, particularly when therapy was initiated early in the disease course^[8]. Dallan et al. similarly noted that treatment with the phosphorylated thiamine active bioform (TPP) is more effective in patients presenting with the recurrent ataxia phenotype, especially when initiated promptly at symptom onset^[3]. However, treatment response is not universal. Approximately 20–30% of reported patients showed only partial or no clear improvement^[8]. Factors associated with poor response included very early onset (infancy), presence of Leigh syndrome phenotype with established structural brain damage before treatment initiation, and biallelic null mutations resulting in complete absence of TPK1 protein^[3, 8, 11]. Emerging evidence suggests that the molecular nature of *TPK1* mutations significantly influences therapeutic outcomes. Patients carrying at least one missense allele with residual enzymatic activity generally exhibit more robust responses to thiamine supplementation compared to those with biallelic null variants (nonsense, frameshift, or large deletions)^[8, 11]. The c.382C > T (p.Leu128Phe) variant identified in our family has been previously characterized functionally. Importantly, patients carrying this variant have shown favorable responses to thiamine therapy when treatment is maintained consistently. In the report by Zhao et al., a patient compound heterozygous for c.382C > T and a frameshift variant showed complete resolution of symptoms with regular thiamine supplement (200mg/day), though symptoms recurred upon treatment discontinuation on two separate occasions, demonstrating the requirement for lifelong therapy^[8]. The two affected siblings in our family demonstrate markedly different outcomes following thiamine therapy, providing a compelling illustration of the importance of treatment adherence and timing. The proband, diagnosed at age 31, received regular thiamine supplementation (100 mg/day). Although his symptoms did not show significant improvement, they did not progress further during follow-up. The absence of deterioration, despite his relatively late age with treatment, suggests that thiamine therapy may have arrested further neurodegeneration. In contrast, the proband's older brother received only a short course of thiamine (30 mg/day) and subsequently discontinued treatment. His spasticity and gait difficulty progressed gradually, culminating in severe disability with inability to walk or stand independently by his thirties. This divergent clinical trajectory between the two siblings carrying identical compound heterozygous *TPK1* mutations strongly supports the efficacy of thiamine therapy in modifying disease progression. We therefore strongly advocate for immediate initiation of high-dose thiamine upon genetic or even clinical suspicion of THMD5, and consistent therapy is essential to prevent progressive neurological deterioration.

Conclusion

In this study, we report two siblings in a Chinese family with THMD5 caused by compound heterozygous *TPK1* variants: a known missense mutation c.382C > T (p.Leu128Phe) and a previously unreported 4.2 kb tandem duplication involving exon 7 and adjacent introns. This novel CNV, identified through WGS, expands the known genetic spectrum of *TPK1*. Clinically, the two affected siblings exhibited marked phenotypic heterogeneity that the older brother presented with earlier onset, typical Leigh-like basal

ganglia lesions on brain MRI, elevated lactate, and cognitive impairment, while the proband had a later onset, milder brain atrophy, and preserved cognition. The striking difference in disease progression between the two patients with identical genotypes probably correlated with high-dose thiamine treatment adherence. We therefore advocate for immediate initiation of high-dose thiamine upon clinical or genetic suspicion of THMD5 and emphasize the necessity of lifelong adherence to prevent irreversible neurological damage. Our findings also support the broader use of WGS in the diagnostic workup of inherited metabolic disorders, particularly when initial testing fails to identify a molecular diagnosis.

Declarations

Conflict of Interest

All authors declare that they have no conflict of interest.

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Author Contribution

HR and YX contributed to the conception and design of this study. HJ and HS collected and analyzed the data. HR drafted the manuscript. YX reviewed the whole paper, figures and tables. All the authors read and approved the final manuscript.

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The authors would like to thank the patients for providing their consent to the publication of this study.

Data Availability

The datasets used and analyzed during the current study are available from the corresponding author on reasonable requests.

References

1. Mayr JA, Freisinger P, Schlachter K, et al. Thiamine pyrophosphokinase deficiency in encephalopathic children with defects in the pyruvate oxidation pathway [J]. *Am J Hum Genet.* 2011;89(6):806–12. 10.1016/j.ajhg.2011.11.007.
2. Banka S, De Goede C, Yue WW, et al. Expanding the clinical and molecular spectrum of thiamine pyrophosphokinase deficiency: a treatable neurological disorder caused by TPK1 mutations [J]. *Mol Genet Metab.* 2014;113(4):301–6. 10.1016/j.ymgme.2014.09.010.
3. Dallan A, Reynolds G, Canavese C, et al. The wide phenotypic spectrum of thiamine metabolism dysfunction syndrome 5 and its treatment [J]. *Orphanet J Rare Dis.* 2025;20(1):160. 10.1186/s13023-025-03665-9.
4. Marcé-Grau A, Martí-Sánchez L, Baide-Mairena H, et al. Genetic defects of thiamine transport and metabolism: A review of clinical phenotypes, genetics, and functional studies [J]. *J Inherit Metab Dis.* 2019;42(4):581–97. 10.1002/jimd.12125.
5. Mascarenhas S, Yeole M, Rao LP, et al. Report of a novel recurrent homozygous variant c.620A > T in three unrelated families with thiamine metabolism dysfunction syndrome 5 and review of literature [J]. *Clin Dysmorphol.* 2024;33(4):160–6. 10.1097/mcd.0000000000000490.
6. Richards S, Aziz N, Bale S, et al. Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology [J]. *Genet Med.* 2015;17(5):405–24. 10.1038/gim.2015.30.
7. Riggs ER, Andersen EF, Cherry AM, et al. Technical standards for the interpretation and reporting of constitutional copy-number variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics (ACMG) and the Clinical Genome Resource (ClinGen) [J]. *Genet Med.* 2020;22(2):245–57. 10.1038/s41436-019-0686-8.
8. Zhao D, Liu M, Jiang H, et al. Thiamine pyrophosphokinase deficiency: report of two Chinese cases and a literature review [J]. *Front Pediatr.* 2023;11:1173787. 10.3389/fped.2023.1173787.
9. Zhu B, Wu J, Chen G, et al. Whole Exome Sequencing Identifies a Novel Mutation of TPK1 in a Chinese Family with Recurrent Ataxia [J]. *J Mol Neurosci.* 2020;70(8):1237–43. 10.1007/s12031-020-01568-x.
10. Li S, Zhou L, Yu H, et al. [Clinical and genetic analysis of a case with Thiamine metabolism dysfunction syndrome 5] [J]. *Zhonghua Yi Xue Yi Chuan Xue Za Zhi.* 2021;38(9):873–6. 10.3760/cma.j.cn511374-20200428-00310.
11. Ortigoza-Escobar JD, Molero-Luis M, Arias A, et al. Treatment of genetic defects of thiamine transport and metabolism [J]. *Expert Rev Neurother.* 2016;16(7):755–63. 10.1080/14737175.2016.1187562.

12. Zhao R, Gao F, Goldman ID. Molecular cloning of human thiamin pyrophosphokinase [J]. *Biochim Biophys Acta*. 2001;1517(2):320–2. 10.1016/s0167-4781(00)00264-5.
13. Huang W, Qin J, Liu D, et al. Reduced thiamine binding is a novel mechanism for TPK deficiency disorder [J]. *Mol Genet Genomics*. 2019;294(2):409–16. 10.1007/s00438-018-1517-3.
14. Invernizzi F, Panteghini C, Chiapparini L, et al. Thiamine-responsive disease due to mutation of *tpk1*: Importance of avoiding misdiagnosis [J]. *Neurology*. 2017;89(8):870–1. 10.1212/wnl.0000000000004270.
15. Marwaha S, Knowles JW, Ashley EA. A guide for the diagnosis of rare and undiagnosed disease: beyond the exome [J]. *Genome Med*. 2022;14(1):23. 10.1186/s13073-022-01026-w.
16. Li X, Huang Z, Chen Y, et al. Case report of two affected siblings in a family with thiamine metabolism dysfunction syndrome 5: a rare, but treatable neurodegenerative disease [J]. *BMC Neurol*. 2022;22(1):373. 10.1186/s12883-022-02887-9.
17. Wang J, Chen X, Mei A. [Clinical characteristics and genetic analysis of a Chinese pedigree affected with thiamine pyrophosphokinase deficiency] [J]. *Zhonghua Yi Xue Yi Chuan Xue Za Zhi*. 2021;38(2):141–4. 10.3760/cma.j.cn511374-20191208-00627.
18. Rüsç CT, Wortmann SB, Kovacs-Nagy R, et al. Thiamine Pyrophosphokinase Deficiency due to Mutations in the TPK1 Gene: A Rare, Treatable Neurodegenerative Disorder [J]. *Neuropediatrics*. 2021;52(2):126–32. 10.1055/s-0040-1715628.
19. Eckenweiler M, Mayr JA, Grünert S, et al. Thiamine Treatment and Favorable Outcome in an Infant with Biallelic TPK1 Variants [J]. *Neuropediatrics*. 2021;52(2):123–5. 10.1055/s-0040-1715631.
20. Au LWC, Lee HHC, Sheng B, et al. Movement disorders associated with thiamine pyrophosphokinase deficiency: Intrafamilial variability in the phenotype [J]. *Clin Neurol Neurosurg*. 2020;199:106258. 10.1016/j.clineuro.2020.106258.
21. Zhu L, Wu R, Ye Z, et al. Identification of two novel TPK1 gene mutations in a Chinese patient with thiamine pyrophosphokinase deficiency undergoing whole exome sequencing [J]. *J Pediatr Endocrinol Metab*. 2019;32(3):295–300. 10.1515/jpem-2018-0363.
22. Nyhan WL, McGowan K, Barshop BA. Thiamine phosphokinase deficiency and mutation in TPK1 presenting as biotin responsive basal ganglia disease [J]. *Clin Chim Acta*. 2019;499:13–5. 10.1016/j.cca.2019.07.034.
23. Mahajan A, Sidiropoulos C. TPK1 mutation induced childhood onset idiopathic generalized dystonia: Report of a rare mutation and effect of deep brain stimulation [J]. *J Neurol Sci*. 2017;376:42–3. 10.1016/j.jns.2017.02.063.
24. Fraser JL, Vanderver A, Yang S, et al. Thiamine pyrophosphokinase deficiency causes a Leigh Disease like phenotype in a sibling pair: identification through whole exome sequencing and management strategies [J]. *Mol Genet Metab Rep*. 2014;1:66–70. 10.1016/j.ymgmr.2013.12.007.
25. Thompson ZE, Boyd NK, Khoshnood MM, et al. Thiamine metabolism dysfunction syndrome 5 (THMD5) mimicking acute disseminated encephalomyelitis [J]. *Am J Med Genet A*. 2023;191(12):2868–72. 10.1002/ajmg.a.63376.

Figures

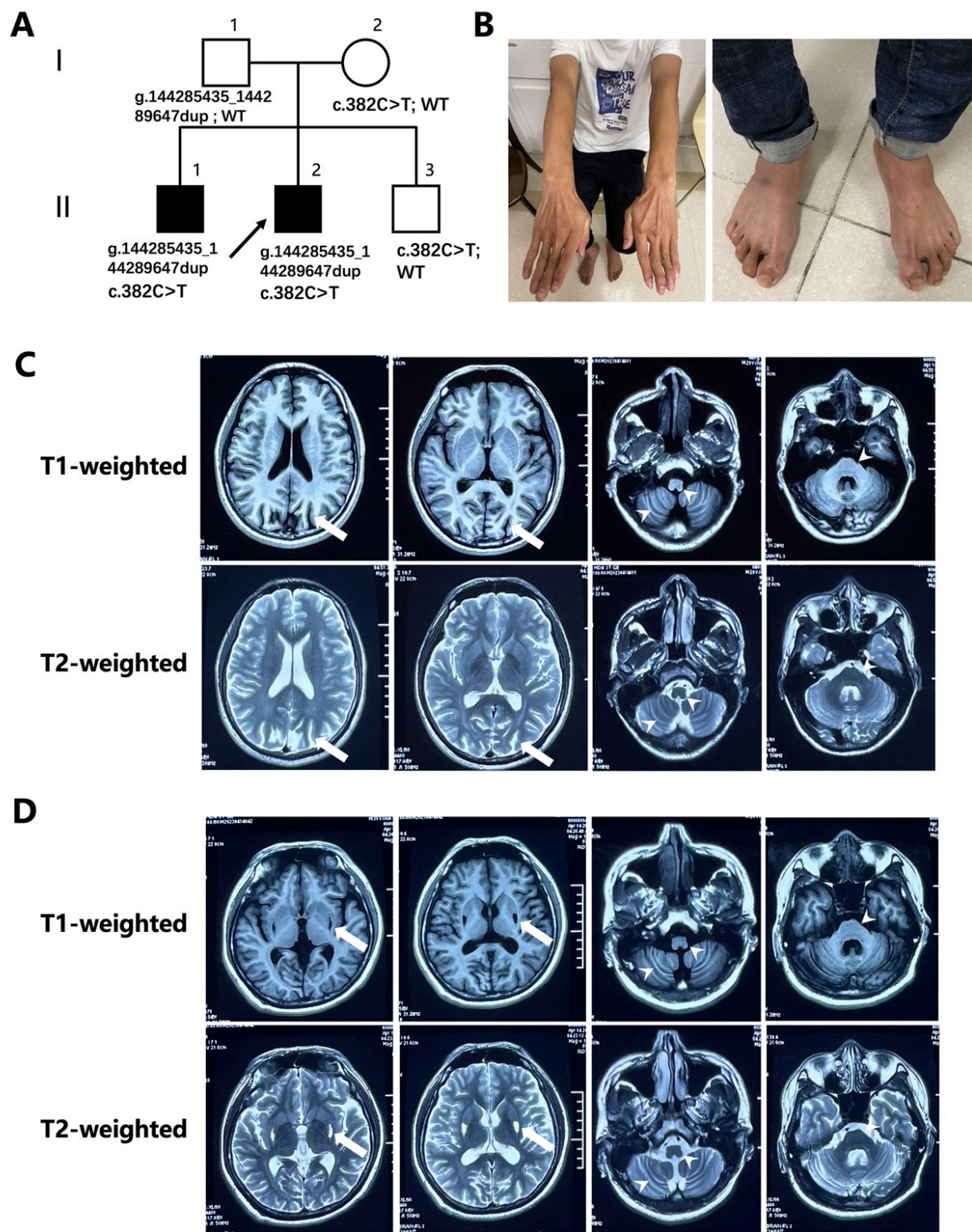


Figure 1

Clinical features of the Chinese family with THMD5 caused by *TPK1* variants.

(A) Pedigree of the family with identified *TPK1* variants. Blank symbols = unaffected; filled symbols = affected; squares = male; circles = female; arrow = the proband; WT = wild-type. (B) *Left*: Mild muscle atrophy of bilateral hands; *Right*: Deformed bending of toes. (C) Brain MRI T1 and T2-weighted images of the proband showed mild atrophy in bilateral parietal cortex (white arrows), brainstem and cerebellum (white arrowheads). (D) Brain MRI T1 and T2-weighted images of the proband's older brother demonstrated symmetric bilateral signal alterations in basal ganglia (white arrows) and significant atrophy in the brainstem and cerebellum (white arrowheads). THMD5 = Thiamine metabolism dysfunction syndrome 5; *TPK1* = Thiamine pyrophosphokinase 1; MRI = Magnetic resonance imaging.

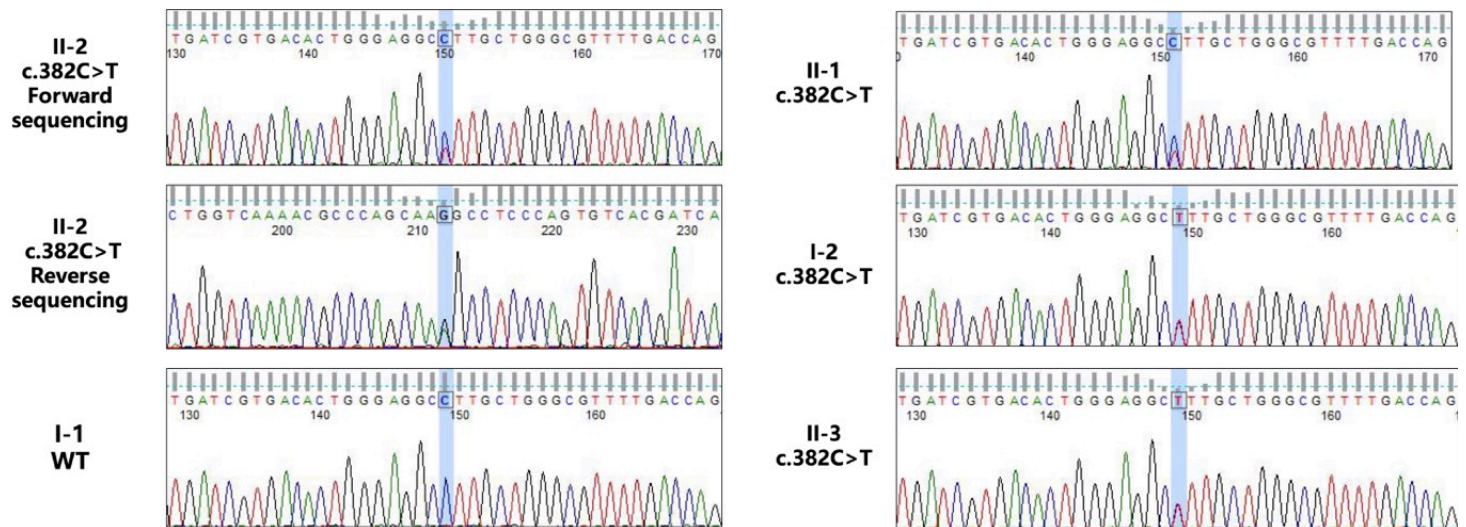


Figure 2

Sanger sequencing chromatograms showed the heterozygous mutation in *TPK1* in the Chinese family with THMD5. The variant c.382C>T was identified in the proband using forward sequencing (left top) and reverse sequencing (left middle). The variant c.382C>T was absent in the proband's father (left bottom). The proband's older brother (right top), the proband's mother (right middle) and the proband's younger brother (right bottom) had heterozygous c.382C>T variant. THMD5 = Thiamine metabolism dysfunction syndrome 5 ; *TPK1* = Thiamine pyrophosphokinase 1.



Figure 3

Aligned sequence reads of the breaking points at left and right ends of the repeated region (NC_000007.13:g.144285435_144289647dup) visualized in the IGV browser from the WGS study. The panel order from top to bottom is the proband, proband's father, proband's older brother, proband's mother and proband's younger brother. The split-read sequences at left and right ends of the repeated region (showed in red boxes) were completely identical in the proband, proband's father and proband's older brother, but were not in proband's mother and proband's younger brother. IGV = Integrative Genomics Viewer; WGS = whole genome sequencing.