Characterization of a Novel Pathogenic Mutation Causing Mitochondrial Neurogastrointestinal Encephalopathy

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Case report

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Abstract

Background Mitochondrial neurogastrointestinal encephalopathy (MNGIE) is a rare autosomal recessive disorder caused by mutations in TYMP gene, encoding nuclear thymidine phosphorylase (TP). MNGIE mainly presents with gastrointestinal symptoms and is mostly misdiagnosed in many patients as malabsorption syndrome, inflammatory bowel disease, anorexia nervosa, and intestinal pseudo-obstruction. Up to date, more than 80 pathogenic and likely pathogenic mutations associated with the disease have been reported in patients from a wide range of ethnicities. The objective of this study was to investigate the underlying genetic abnormalities in a 25-year-old woman affected with MNGIE. Case Presentation The patient was a 25-year-old female referred to our center with the chief complaint of severe abdominal pain and diarrhea for two years that had worsened from two months prior to admission. The clinical and para-clinical findings were in favor of mitochondrial neurogastrointestinal encephalopathy syndrome. Subsequent genetic studies revealed a novel, private, homozygous nonsense mutation in TYMP gene (c. 1013 C>A, p.S338X). Sanger sequencing confirmed the new mutation in the proband. Multiple sequence alignment showed high conservation of amino acids of this protein across different species. Conclusion The detected new nonsense mutation in the TYMP gene would be very important for genetic counseling and subsequent early diagnosis and initiation of proper therapy. This novel pathogenic variant would help us establish future genotype-phenotype correlations and identify different pathways related to this disorder.

Background

Mitochondrial neurogastroinstestinal encephalopathy (MNGIE – OMIM# 603041) is a rare multisystem autosomal recessive disorder caused by homozygous or compound heterozygous mutations in the nuclear-encoded thymidine phosphorylase gene (*TYMP*; 131222) on chromosome 22q13, the first gene whose role was defined at molecular level in the defects of intergenomic communication [1]. Impairment in this enzyme with resultant decreased enzyme activity leads to accumulation of the enzyme's substrates, thymidine and deoxyuridine, which in turn leads to an imbalance in the intra-mitochondrial nucleotide pool and multiple deletions, point-specific mutations and depletions in mitochondrial DNA (mtDNA) [2, 3]. Recent evidence has suggested that the accumulation of these nucleosides is the main culprit for the development of the molecular and phenotypic aberrations reported in this disorder [4].

First described in 1976, MNGIE usually presents in the first to fifth decades of life, with a progressive clinical course leading to death at the mean age of 37 [5-7]. The clinical manifestations of MNGIE are severe gastrointestinal dysmotility, cachexia, extraocular muscle weakness with resultant ptosis and ophthalmoplegia, sensorimotor neuropathy, and leukoencephalopathy [8]. More than 120 patients with diagnostic features consistent with MNGIE have so far been reported in the literature, with more than 80 pathogenic and likely pathogenic mutations (https://www.ensembl.org) associated with the disease identified in patients from a wide range of ethnicities [9, 10].

Herein, we report on a patient with MNGIE with a novel homozygous mutation in *TYMP* gene, along with the clinical, laboratory and imaging findings.

Case Presentation

Clinical Presentation

A 25-year-old female was referred to our center with the chief complaint of severe abdominal pain and diarrhea for two years that had worsened from two months prior to admission. She had significant weight loss during this period; weighing 36.5 kg with a height of 160 cm, her body mass index (BMI) was 14.3 kg/m^2 at the time of admission. Her past medical and surgical history was only significant for one undocumented episode of seizure at the age of three and appendectomy three years before. On interview, she denied fear of weight gain, laxative abuse, and self-induced vomiting. Her parents were consanguineous. There was no history of sibling loss or any similar symptoms in other family members.

Clinical examination revealed a cachectic lady with external ophthalmoplegia, ptosis, right lower quadrant scar of the McBurney (oblique) incision for appendectomy, decreased muscle power in the upper (4/5 MRC muscle scale) and lower (3/5 MRC muscle scale) extremities, and absent deep tendon reflexes. Abdominopelvic sonography revealed mild free fluid in the abdominal cavity and increased thickness in the bowel wall (4.8 mm) and a 16×11-mm cortical cyst in the upper pole of the left kidney with thin septation. A diagnostic esophago-gastro-duodenoscopy showed diffuse severe erythema and congestion in the body, fundus, and antrum of the stomach with moderate chronic gastritis in pathologic examination and deformity of the duodenal bulb with decreased folds in D2 part of the duodenum. On colonoscopy, the ileocecal valve was stenotic; biopsy revealed submucosal fibrosis with lymphoid proliferation and focal ulceration. Echocardiography was normal except for mild pericardial effusion.

Her complete blood count indicated the presence of microcytic hypochromic anemia. Biochemistry revealed low total protein level (4.1 g/dl; reference range: 6.6–8.8 g/dl) and albumin (2.2 g/dl; reference range: 3.5–5.2 g/dl). Serum lactate level (35.3 mg/dl; reference range: 4.5–19.8 mg/dl) was elevated. Fecal occult blood test was positive with moderately elevated fecal calprotectin level (58.5 µg/g; reference range: <15 µg/g) suggestive for the inflammatory process. Cerebrospinal fluid analysis revealed marked elevation of protein level (122 mg/dl; reference range: 15–45 mg/dl). Serology tests for HIV, HBV, and HCV were negative. Serum anti-tissue trans-glutaminase antibodies, anti-phospholipid antibodies, anti-cardiolipin antibodies, lupus anti-coagulants, β_2 microglobulin, anti-nuclear antibody (ANA), anti-double stranded DNA (anti-dsDNA), C-ANCA, and P-ANCA were also within the normal limits. Serum levels of complement factors were also found to be altered: C_3 level (65 mg/dl; reference range: 90–180 mg/dl) was decreased; C_4 level (11.4 mg/dl; reference range: 10–40 mg/dl) was within the lower limit of the normal range. Blood TP activity was not measured for lack of laboratory resources.

The electrodiagnostic evaluation showed a neurogenic pattern on needle electromyography (EMG), conduction block in sensory nerves, and decreased compound muscle action potential (CMAP) in motor

nerves with decreased conduction velocity and prolonged F-wave latency. Brain MRI with contrast showed leukoencephalopathy with diffusely increased T_2 signal intensity in both cerebral hemispheres white matter. Hypersignal intensity in splenium of corpus callosum was also observed (Figure 1).

Molecular Analysis

After obtaining informed consent, a blood sample was obtained from the patient. The whole blood sample was prepared in EDTA tube. Genomic DNA was then extracted from peripheral leukocytes using the QIAamp DNA Blood Mini Kit (Qiagen, Germany).

Whole Exome Sequencing (WES) was performed on Illumina NextSeq500 instrument to a sequence close to 100 million reads. WES result was then analyzed using different bioinformatics tools and databases such as BWA aligner, GATK and ANNOVAR [11-13]. WES uncovered a novel, private, homozygous stopgain mutation in *TYMP* gene (NM_001113756: exon7: c. 1013C>A: p.S338X, Chr: 50526392).

Subsequently in order to confirm this novel mutation using Sanger sequencing, the region of interest was amplified using PCR on the DNA of the proband using following primers: F–TYMP-E7: 5'-ACTTAAGGGACCTGGTCACCAC-3' and R–TYMP-E7: 5'-AGCCTCTGACCCACGTCGA-3' (PCR product: 594bp). Then, the amplicon was sequenced with both forward and reverse primers using ABI BigDye Terminator Cycle Sequencing Kit (Applied Biosystems®, USA). Sanger sequencing result was analyzed by NCBI BLAST (https://blast.ncbi.nlm.nih.gov) and CodonCode Aligner (http://www.codoncode.com/aligner/). Sanger sequencing confirmed this mutation as homozygous in the proband (Figure 2).

This mutation has not been reported yet in main variant databases and our public database (Bayangene). The mutation occurred in domains described as Glycos_transf_3 in Pfam database or Nucleoside phosphorylase/phosphoribosyltransferase catalytic domain in Gene3D database which affects this domain and other domains such PYNP_C, Pyrimidine nucleoside phosphorylase C-terminal domain, TIGR02644, THYMIDINE PHOSPHORYLASE and TP_PyNP which are described by a different database (Table 1).

To reveal the conservation of amino acid sequence of TYMP protein across various species, multiple sequence alignment analysis by BLAST available on ExPASy (https://web.expasy.org/cgi-bin/blast/blast.pl) was also performed. Multiple sequence alignment showed high conservation of amino acids of this protein across different species, mainly in the mutated region and after this position. (Figure 3)

Discussion And Conclusions

A group of mitochondrial disorders are characterized by mutations in the nuclear genome affecting expression and replication of the genes on the mitochondrial genetic material. Progressive external ophthalmoplegia (PEO) was the first disease identified in this group caused by defects in the intergenomic communication [14]. MNGIE, a rare progressive multisystem autosomal recessive disorder caused by a mutation in *TYMP* gene is also a member of this group of disorders.

TYMP gene which encodes the cytosolic enzyme named thymidine phosphorylase, TP, is located at chromosome 22q13.33. This protein catalyzes phosphorylation of mitochondrial dThd and dUrd to thymine and uridine, respectively (Figure 4). TP plays an important role in the metabolic pathways of various cells including those in the brain, muscle, RBCs, WBCs, and bone marrow, relying on the salvage pathway for recovering nucleosides [15, 16]. MNGIE usually presents with symptoms of gastrointestinal dysfunction, such as gastrointestinal motility disorders, gastro-esophageal reflux, dysphagia, abdominal pain and distention, and diarrhea leading to severe weight loss and cachexia [17]. At this stage of the disease, most of the patients are misdiagnosed as malabsorption syndrome, inflammatory bowel disease (IBD), anorexia nervosa, or intestinal pseudo-obstruction, often leading to unnecessary medical interventions and delay in diagnosis of up to 10 years [6, 18-20]. Ptosis, ophthalmoparesis, hearing loss, and sensory-motor neuropathy constitute the most common neurologic features of patients with MNGIE [8]. Due to the high metabolic activity of extraocular muscles, deterioration in their function resulting in ophthalmoplegia or ophthalmoparesis occurs early in the course of the disease that parallels the disease progression [21]. Neuroimaging studies such as brain MRI and magnetic resonance spectroscopy (MRS) might yield a clue about the diagnosis of MNGIE, with the absence of leukoencephalopathy ruling out MNGIE in most cases. Unlike the patient reported here, who had involvement of splenium of the corpus callosum, it is relatively spared in most individuals [8, 10, 22]. Our patient was found to have leukoencephalopathy, with diffuse T_2 hyperintensity in both cerebral hemispheres white matter on brain MRI, though, she had normal cognitive function. This, in turn, is likely to be due to the impaired blood-brain barrier function in these patients leading to edema in lieu of demyelination [23].

Other disorders with phenotypes similar to MNGIE, caused by mutations in *RRM2B* and *POLG* genes have been reported [24, 25]. Therefore, it is prudent to test the individuals suspected of having MNGIE for these genes as well. In our patient whole exome sequencing was done and no mutations in these genes were detected.

To the best of our knowledge, around 80 pathogenic mutations in the *TYMP* gene have so far been reported. Attempts to draw a genotype-phenotype correlation in this disorder have mostly been discouraging, except for c. 622G>A variant (p.Val208Met), producing less severe TP dysfunction, leading to a late-onset disease [26-28].

Current treatment modalities for MNGIE mainly focus on restoration of the activity of TP and lowering the circulatory levels of the nucleoside substrates. Hematopoietic stem cell transplantation (HSCT) has so far been used to restore TP enzyme activity in patients with MNGIE. A retrospective analysis of 24 patients who underwent HSCT for the treatment of MNGIE revealed a survival rate of 37.5% after a median follow-

up of almost four years. Most of the deaths were attributed to the transplantation with MNGIE complications leading to death in a quarter of the patients [29]. It was found that younger patients without gastrointestinal dysmotility and liver disease receiving HSCT from an HLA-matched donor would benefit mostly from this type of treatment, highlighting the importance of diagnosis in the momentous days early in the course of the illness, when HSCT would change the course of the disease [29]. Hemodialysis and peritoneal dialysis have also been proposed as treatment modalities in these patients intending to remove the nucleosides from the peripheral circulation. A prospective study evaluating a 29-year-old patient with MNGIE who underwent extensive hemodialysis for one year also revealed that it has only a transient effect on the serum and urine levels of nucleosides with no long-term effects; there were no changes in the level of the toxic metabolites in the CSF in both short-term (within 24 hours) and long-term (at months 6 and 12) [30]. Our patient in this report underwent hemodialysis with only mild improvements in the gastrointestinal symptoms. These findings cast doubt on the efficacy of dialysis in the treatment of MNGIE. Other therapeutic modalities including platelet infusion, which was also performed in our patient, and orthotopic liver transplantation have also been reported for the treatment of this disease in the literature [4, 31].

The mutation found initially by WES and subsequently confirmed using Sanger sequencing is predicted to disrupt the proper function of TYMP protein since different reports have identified frameshift mutations before and after this region resulting in the impaired TYMP [32-34].

Our patient had many of the clinical, laboratory, and imaging features seen in MNGIE. The detected novel nonsense mutation in the *TYMP* gene would be of importance for genetic counseling and subsequent early diagnosis and initiation of proper therapy. On account of the wide clinical spectrum of signs and symptoms presented by patients with MNGIE, molecular diagnostic methods would be of paramount importance.

Abbreviations

MNGIE Mitochondrial neurogastrointestinal encephalopathy

TP Thymidine phosphorylase

mtDNA Mitochondrial DNA

ANA Anti-nuclear antibody

anti-dsDNA anti-double stranded DNA

EMG Electromyography

CMAP Compound muscle action potential

WES Whole Exome Sequencing

PEO Progressive external ophthalmoplegia

IBD Inflammatory bowel disease

MRS Magnetic resonance spectroscopy

HSCT Hematopoietic stem cell transplantation

Declarations

Ethics Approval and Consent to Participate

The Ethics Committee of the Persian BayanGene Research and Training Center approved the study protocol. This investigation was conducted in accordance with the ethical principles and recommendations outlined in the Declaration of Helsinki.

Consent for publication

The patient consented to the publication of the case and accompanying clinical and genetics data. Written informed consent was obtained from the patient.

Availability of data and materials

All data are available from the corresponding author on request.

Competing Interests: None to declare.

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Authors' contributions

MAF conceived and designed the study, collected, assembled and interpreted NGS data. PH, SEJ and VRO did the clinical evaluation. MS, SBJ and MY did the genetic studies. PH drafted the manuscript. HD did the bioinformatics studies. MM and HD revised the manuscript. All authors have read and approved the manuscript.

References

1. Nishino I, Spinazzola A, Hirano M: **Thymidine phosphorylase gene mutations in MNGIE, a human mitochondrial disorder**. *Science* 1999, **283**(5402):689-692.
2. Nishigaki Y, Marti R, Copeland WC, Hirano M: **Site-specific somatic mitochondrial DNA point mutations in patients with thymidine phosphorylase deficiency**. *J Clin Invest* 2003, **111**(12):1913-

1921.

3. Nishino I, Spinazzola A, Papadimitriou A, Hammans S, Steiner I, Hahn CD, Connolly AM, Verloes A, Guimaraes J, Maillard I *et al*: **Mitochondrial neurogastrointestinal encephalomyopathy: an autosomal recessive disorder due to thymidine phosphorylase mutations**. *Ann Neurol* 2000, **47**(6):792-800.
4. Yadak R, Sillevis Smitt P, van Gisbergen MW, van Til NP, de Coo IF: **Mitochondrial Neurogastrointestinal Encephalomyopathy Caused by Thymidine Phosphorylase Enzyme Deficiency: From Pathogenesis to Emerging Therapeutic Options**. *Front Cell Neurosci*, **11**:31.
5. Cardaioli E, Da Pozzo P, Malfatti E, Battisti C, Gallus GN, Gaudiano C, Macucci M, Malandrini A, Margollicci M, Rubegni A *et al*: **A second MNGIE patient without typical mitochondrial skeletal muscle involvement**. *Neurol Sci*, **31**(4):491-494.
6. Lara MC, Valentino ML, Torres-Torronteras J, Hirano M, Marti R: **Mitochondrial neurogastrointestinal encephalomyopathy (MNGIE): biochemical features and therapeutic approaches**. *Biosci Rep* 2007, **27**(1-3):151-163.
7. Okamura K, Santa T, Nagae K, Omae T: **Congenital oculoskeletal myopathy with abnormal muscle and liver mitochondria**. *J Neurol Sci* 1976, **27**(1):79-91.
8. Hirano M, Nishigaki Y, Marti R: **Mitochondrial neurogastrointestinal encephalomyopathy (MNGIE): a disease of two genomes**. *Neurologist* 2004, **10**(1):8-17.
9. Baris Z, Eminoglu T, Dalgic B, Tumer L, Hasanoglu A: **Mitochondrial neurogastrointestinal encephalomyopathy (MNGIE): case report with a new mutation**. *Eur J Pediatr*, **169**(11):1375-1378.
10. M H: **Mitochondrial Neurogastrointestinal Encephalopathy Disease**. In.; 2005 Apr 22 [Updated 2016 Jan 14].
11. Li H, Durbin R: **Fast and accurate short read alignment with Burrows-Wheeler transform**. *Bioinformatics* 2009, **25**(14):1754-1760.
12. McKenna A, Hanna M, Banks E, Sivachenko A, Cibulskis K, Kernytsky A, Garimella K, Altshuler D, Gabriel S, Daly M *et al*: **The Genome Analysis Toolkit: a MapReduce framework for analyzing next-generation DNA sequencing data**. *Genome Res*, **20**(9):1297-1303.
13. Wang K, Li M, Hakonarson H: **ANNOVAR: functional annotation of genetic variants from high-throughput sequencing data**. *Nucleic Acids Res*, **38**(16):e164.
14. Zeviani M, Servidei S, Gellera C, Bertini E, DiMauro S, DiDonato S: **An autosomal dominant disorder with multiple deletions of mitochondrial DNA starting at the D-loop region**. *Nature* 1989, **339**(6222):309-311.
15. Suomalainen A, Isohanni P: **Mitochondrial DNA depletion syndromes--many genes, common mechanisms**. *Neuromuscul Disord*, **20**(7):429-437.
16. Young JD, Yao SY, Sun L, Cass CE, Baldwin SA: **Human equilibrative nucleoside transporter (ENT) family of nucleoside and nucleobase transporter proteins**. *Xenobiotica* 2008, **38**(7-8):995-1021.

17. Filosto M, Scarpelli M, Tonin P, Testi S, Cotelli MS, Rossi M, Salvi A, Grottolo A, Vielmi V, Todeschini A *et al*: **Pitfalls in diagnosing mitochondrial neurogastrointestinal encephalomyopathy**. *J Inherit Metab Dis*, **34**(6):1199-1203.
18. Laforce R, Jr., Valdmanis PN, Dupre N, Rouleau GA, Turgeon AF, Savard M: **A novel TYMP mutation in a French Canadian patient with mitochondrial neurogastrointestinal encephalomyopathy**. *Clin Neurol Neurosurg* 2009, **111**(8):691-694.
19. Monroy N, Macias Kauffer LR, Mutchinick OM: **Mitochondrial neurogastrointestinal encephalomyopathy (MNGIE) in two Mexican brothers harboring a novel mutation in the ECGF1 gene**. *Eur J Med Genet* 2008, **51**(3):245-250.
20. Taanman JW, Daras M, Albrecht J, Davie CA, Mallam EA, Muddle JR, Weatherall M, Warner TT, Schapira AH, Ginsberg L: **Characterization of a novel TYMP splice site mutation associated with mitochondrial neurogastrointestinal encephalomyopathy (MNGIE)**. *Neuromuscul Disord* 2009, **19**(2):151-154.
21. Wang HF, Wang J, Wang YL, Fan JJ, Mo GL, Gong FY, Chai ZM, Zhang J, Meng HX, Li CX *et al*: **A novel thymidine phosphorylase mutation in a Chinese MNGIE patient**. *Acta Neurol Belg*, **117**(1):259-267.
22. Vissing J, Ravn K, Danielsen ER, Duno M, Wibrand F, Wevers RA, Schwartz M: **Multiple mtDNA deletions with features of MNGIE**. *Neurology* 2002, **59**(6):926-929.
23. Szigeti K, Sule N, Adesina AM, Armstrong DL, Saifi GM, Bonilla E, Hirano M, Lupski JR: **Increased bloodâ€“brain barrier permeability with thymidine phosphorylase deficiency**. *Annals of Neurology* 2004, **56**(6):881-886.
24. Shaibani A, Shchelochkov OA, Zhang S, Katsonis P, Lichtarge O, Wong LJ, Shinawi M: **Mitochondrial neurogastrointestinal encephalopathy due to mutations in RRM2B**. *Arch Neurol* 2009, **66**(8):1028-1032.
25. Tang S, Dimberg EL, Milone M, Wong LJ: **Mitochondrial neurogastrointestinal encephalomyopathy (MNGIE)-like phenotype: an expanded clinical spectrum of POLG1 mutations**. *J Neurol*, **259**(5):862-868.
26. Libernini L, Lupis C, Mastrangelo M, Carrozzo R, Santorelli FM, Inghilleri M, Leuzzi V: **Mitochondrial neurogastrointestinal encephalomyopathy: novel pathogenic mutations in thymidine phosphorylase gene in two Italian brothers**. *Neuropediatrics*, **43**(4):201-208.
27. Marti R, Verschuuren JJ, Buchman A, Hirano I, Tadesse S, van Kuilenburg AB, van Gennip AH, Poorthuis BJ, Hirano M: **Late-onset MNGIE due to partial loss of thymidine phosphorylase activity**. *Ann Neurol* 2005, **58**(4):649-652.
28. Massa R, Tessa A, Margollicci M, Micheli V, Romigi A, Tozzi G, Terracciano C, Piemonte F, Bernardi G, Santorelli FM: **Late-onset MNGIE without peripheral neuropathy due to incomplete loss of thymidine phosphorylase activity**. *Neuromuscul Disord* 2009, **19**(12):837-840.
29. Halter JP, Michael W, Schupbach M, Mandel H, Casali C, Orchard K, Collin M, Valcarcel D, Rovelli A, Filosto M *et al*: **Allogeneic haematopoietic stem cell transplantation for mitochondrial**

neurogastrointestinal encephalomyopathy. *Brain*, **138**(Pt 10):2847-2858.

30. Roeben B, Marquetand J, Bender B, Billing H, Haack TB, Sanchez-Albisua I, Schols L, Blom HJ, Synofzik M: **Hemodialysis in MNGIE transiently reduces serum and urine levels of thymidine and deoxyuridine, but not CSF levels and neurological function**. *Orphanet J Rare Dis*, **12**(1):135.
31. De Giorgio R, Pironi L, Rinaldi R, Boschetti E, Caporali L, Capristo M, Casali C, Cenacchi G, Contin M, D'Angelo R *et al*: **Liver transplantation for mitochondrial neurogastrointestinal encephalomyopathy**. *Ann Neurol*, **80**(3):448-455.
32. Gamez J, Ferreiro C, Accarino ML, Guarner L, Tadesse S, Marti RA, Andreu AL, Raguer N, Cervera C, Hirano M: **Phenotypic variability in a Spanish family with MNGIE**. *Neurology* 2002, **59**(3):455-457.
33. Garone C, Tadesse S, Hirano M: **Clinical and genetic spectrum of mitochondrial neurogastrointestinal encephalomyopathy**. *Brain*, **134**(Pt 11):3326-3332.
34. Slama A, Lacroix C, Plante-Bordeneuve V, Lombes A, Conti M, Reimund JM, Auxenfants E, Crenn P, Laforet P, Joannard A *et al*: **Thymidine phosphorylase gene mutations in patients with mitochondrial neurogastrointestinal encephalomyopathy syndrome**. *Mol Genet Metab* 2005, **84**(4):326-331.

Tables

Table 1

Domain source	Start	End	Description
PANTHER	29	478	THYMIDINE PHOSPHORYLASE
Pfam	38	99	Glycos_trans_3N
SuperFamily	36	102	Nucleoside phosphorylase/phosphoribosyltransferase N-terminal domain
Pfam	110	340	Glycos_transf_3
Gene3D	102	371	Nucleoside phosphorylase/phosphoribosyltransferase catalytic domain superfamily
SuperFamily	106	357	Nucleoside phosphorylase/phosphoribosyltransferase catalytic domain
Smart	388	462	PYNP_C_2
Pfam	390	446	PYNP_C
SuperFamily	375	471	Pyrimidine nucleoside phosphorylase C-terminal domain
TIGRfam	38	453	TIGR02644
Prosite_patterns	144	159	THYMID_PHOSPHORYLASE
PANTHER	29	478	THYMIDINE PHOSPHORYLASE
PIRSF	33	477	TP_PyNP

Figures

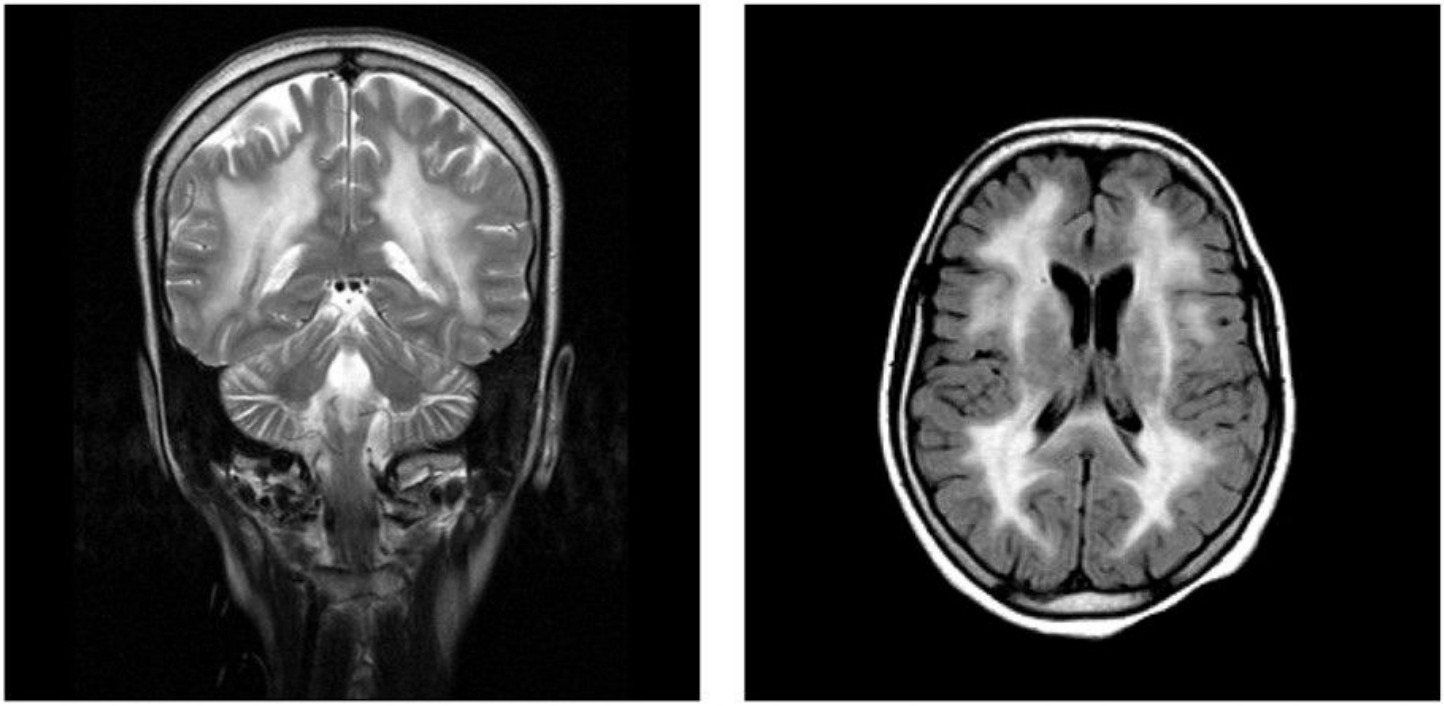

Figure 1

Brain MRI revealing diffuse white matter signal changes.

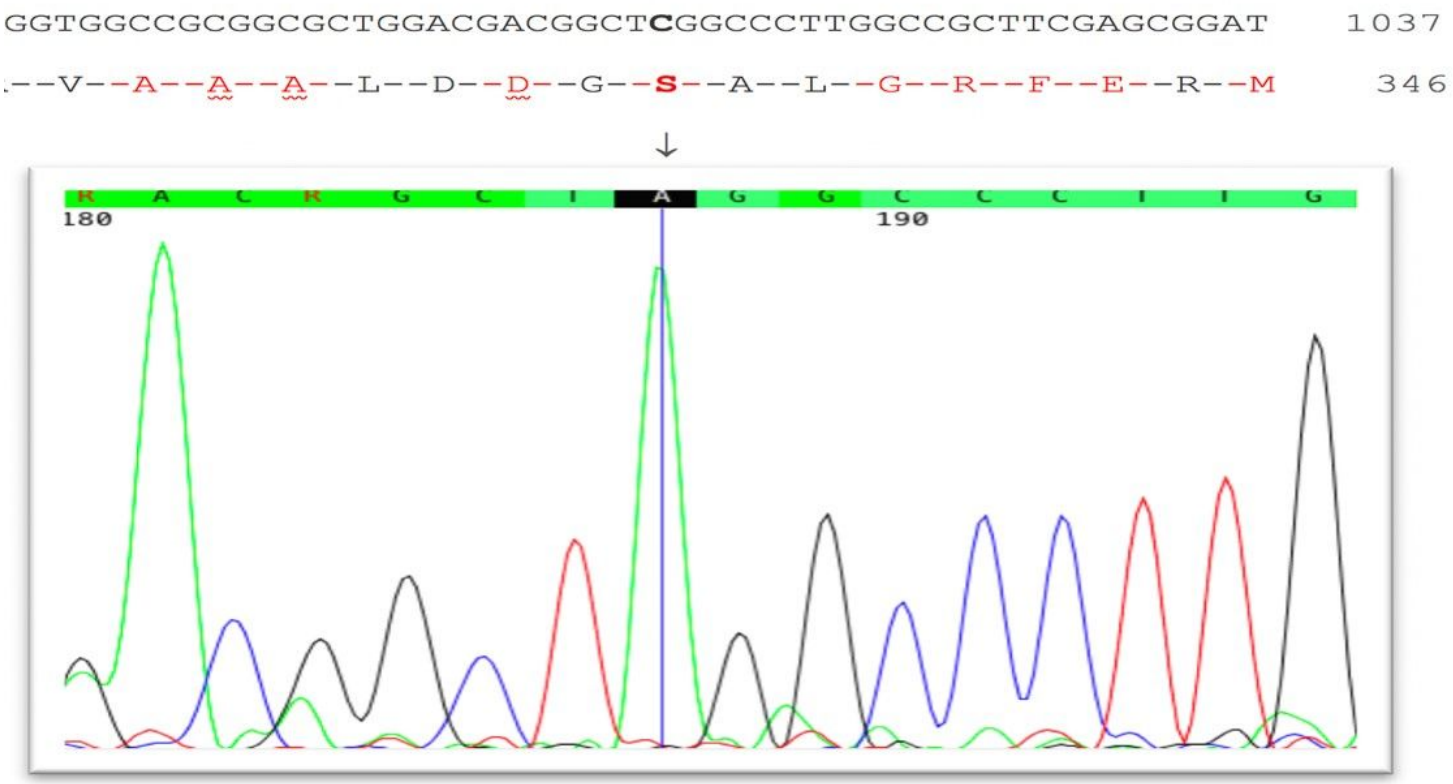

Figure 2

Sanger sequencing chromatogram of the novel identified variant in this study. The arrow shows the position of homozygous mutation in TYMP (c. 1013 C>A, p.S338X).

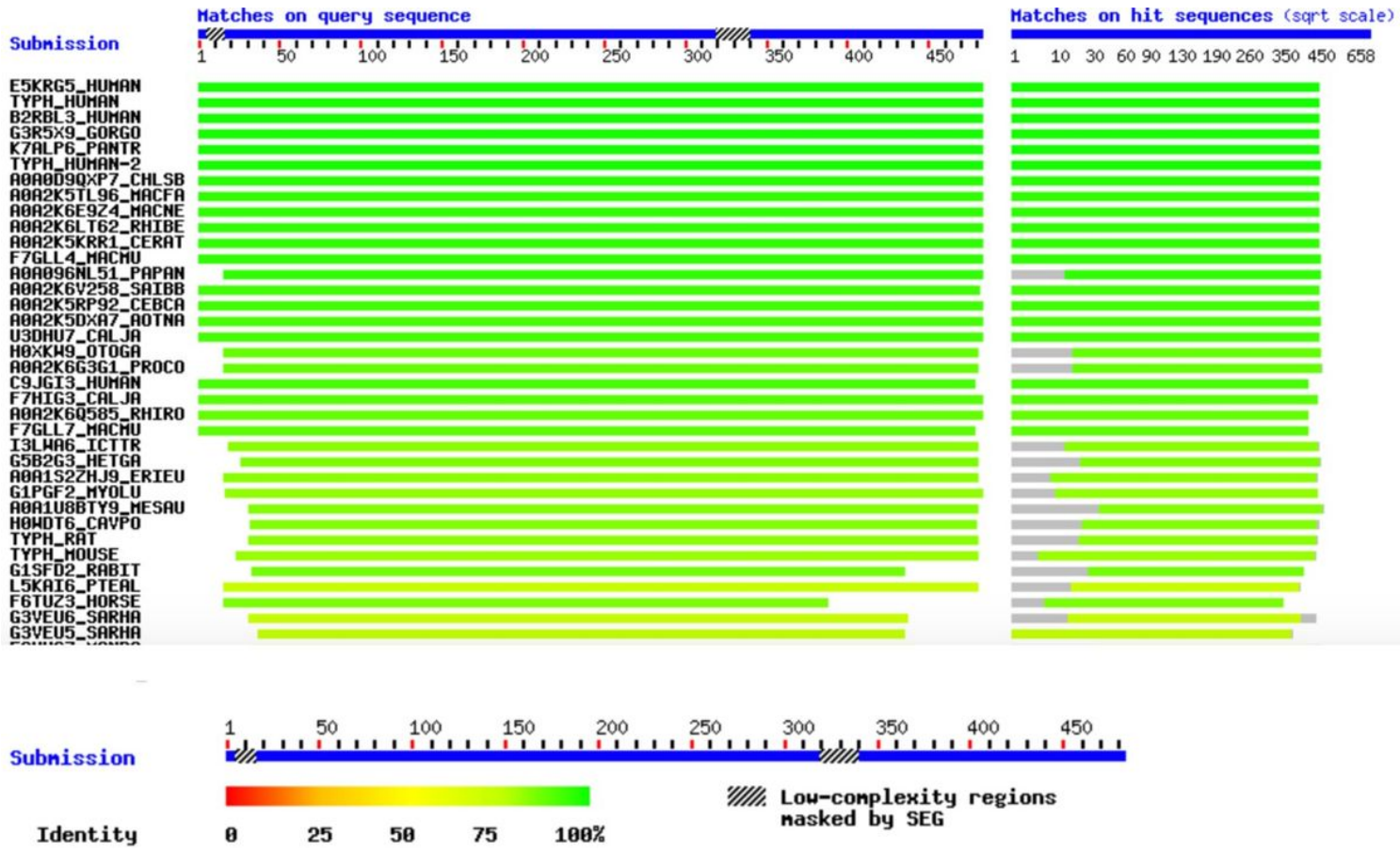

Figure 3

Multiple sequence alignment of the whole amino acid sequence of TYMP with other TYMP orthologs across different species.

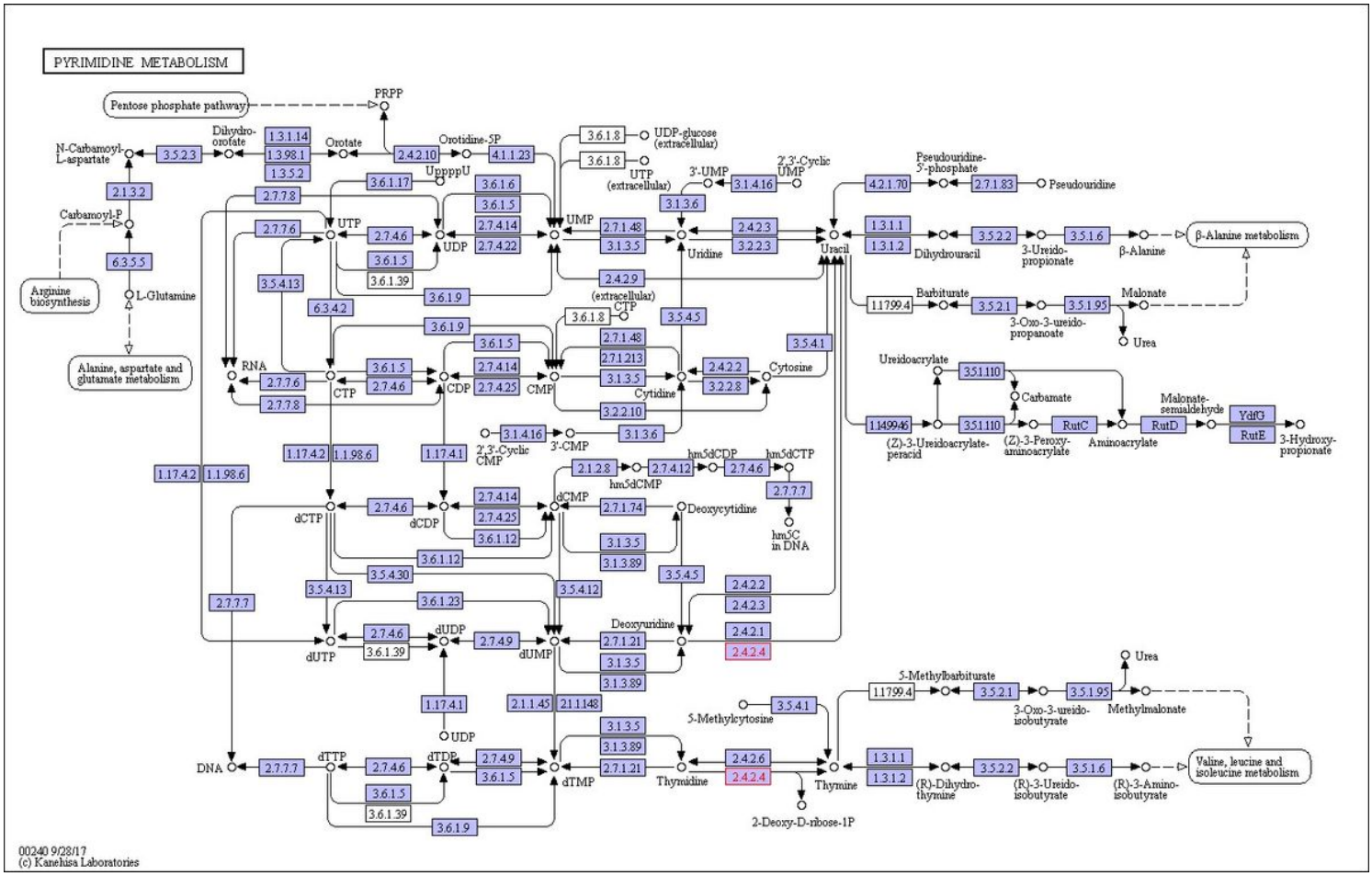

Figure 4

The main roles of TYMP in pyrimidine metabolism extracted from the KEGG database (ko00240). The red numbers (2.4.2.4) shows the positions of the enzyme functions in pyrimidine metabolism. As depicted, this enzyme with its phosphorolysis activity on thymidine (TdR) is able to catalyze TdR to thymine and 2-deoxy-D-ribose 1-phosphate (dR-1-P). TYMP also catalyze the thymidine formation from thymine and dR-1-P. TP also in another catalectic activity can phosphorolyze the deoxyuridine to uracil and dR-1-P, indicating its vital roles in biological metabolism.

Supplementary Files

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