

Novel PANK2 mutation in the first Greek compound heterozygote patient with pantothenate-kinase-associated neurodegeneration

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Abstract

Pantothenate-kinase-associated neurodegeneration is the most common autosomal recessive form of neurodegeneration with brain iron accumulation. Less than 100 mutations in *PANK2* gene (20p13) are responsible for classic and atypical cases. We report here the first Greek case of atypical pantothenate-kinase-associated neurodegeneration, confirmed by molecular analysis that revealed two trans-acting mutations. Our findings highlight the possible role of rare variants contributing to disease risk and possibly to variable clinical phenotype.

Keywords

Pantothenate-kinase-associated neurodegeneration, neurodegeneration with brain iron accumulation, atypical parkinsonism, focal dystonia

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Introduction

Pantothenate-kinase-associated neurodegeneration (PKAN) is the most common cause of neurodegeneration with brain iron accumulation (NBIA).¹ It is caused by mutations in the pantothenate kinase-2 gene (*PANK2*) and it is inherited in an autosomal recessive manner.² The classical form of the disease is characterized by childhood onset and relatively rapid progression; however, approximately 25% of patients have an atypical form, with onset in the second or third decade, a variable rate of progression and the presence of psychiatric symptoms, seizures and focal dystonia.^{1,2} Here, we report the first Greek compound heterozygote patient with atypical PKAN, carrying a novel mutation.

Clinical report

The patient is a 25-year-old male, offspring of a non-consanguineous marriage. At age 22, he had to interrupt his studies on physics due to dystonic right arm movements resulting in writing difficulties. He gradually developed orobuccal dystonia and dysarthria. The patient's Burke–Fahn–Marsden

Dystonia Rating Scale (BFMDRS) score was 12.³ He exhibited mild cogwheel rigidity, reflexes were brisk and plantar responses were indifferent, but neurological examination was otherwise unremarkable. Motor function was evaluated with the Movement Disorder Society–Sponsored Revision of the Unified Parkinson's Disease Rating Scale (MDS-UPDRS), yielding a total score of 6 points.⁴ Ophthalmologic evaluation showed absence of Kayser–Fleischer rings, optic atrophy or pigmentary retinopathy. Peripheral blood acanthocytosis was not observed. Routine laboratory evaluation was normal.

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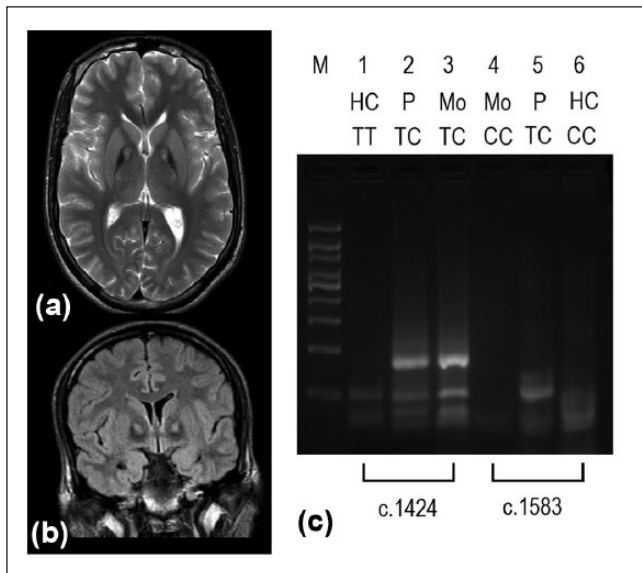


Figure 1. (a, b) Magnetic resonance imaging showing bilateral peripheral hypointensity in the globi pallidi with a central focus of gliosis, called as “eye-of-the-tiger” sign; (c) restriction fragment patterns of PCR products illustrating the alleles observed in loci c.1424 and c.1583.

P: patient; Mo: patient’s mother; HC: healthy control; M: molecular weight marker.

Neuropsychological evaluation revealed no cognitive decline or behavioral/psychiatric symptoms (see Supplementary Material). Brain imaging revealed the “eye-of-the-tiger” sign (Figure 1(a) and (b)). There is no family history. His mother (58 years) is asymptomatic and his father died of lung cancer at age 54. A trial with L-dopa, up to 750 mg/day, had no effect. Orphenadrin (150 mg daily) had an effect that was modest at best and the patient preferred discontinuation due to significant anticholinergic side effects.

Genetic analysis

Blood samples for total DNA extraction were obtained from the patient and his mother after informed consent. The seven exons of *PANK2* gene, including the exon/intron boundaries, were sequenced in the patient’s DNA sample in GENetic DIAgnostic Network (GENDIA, Emiel Vloorsstraat 9, B 2020 Antwerp, Belgium). Two combined heterozygous mutations were detected, namely, p.Met475Thr (c.1424T>C) in exon 5, and p.Thr528Met (c.1583C>T) in exon 6 of *PANK2*.

Molecular detection of either mutation in the *PANK2* gene of the patient, his mother and 100 healthy controls of Greek origin, aged 35–57 years, was performed by restriction fragment length polymorphism typing. This involved a combination of polymerase chain reaction (PCR) amplification and digestion with a specific restriction endonuclease that distinguished the normal and the mutant alleles followed by agarose gel electrophoresis analysis. For c.1424T>C in exon 5, primers

5A (forward): 5′-GTTCTGTTGGGCTTTGTTGCTG-3′ and 5B (reverse): 5′-CACACATTCTTGCTATTGAGCC-3′ were used. The generated PCR product of 172bp was cleaved by restriction enzyme *Nla*III into two fragments of 100 and 72bp only if the normal T allele was present. For c.1583C>T in exon 6, primers 6A (forward): 5′-CATTAACCAGGTGGTATTTGTTGG-3′ and 6B (reverse): 5′-AGTGCTTTCAACTGCCCC TTG-3′ were used. The generated PCR product of 108bp was cleaved by restriction enzyme *Pvu*I into two fragments of 60 and 48bp only if the normal C allele was present.

Restriction enzyme analysis verified that the patient had two mutations in compound heterozygosity: c.1424T>C (p.M475T) and c.1583C>T (p.T528M), at exons 5 and 6, respectively (Figure 1(c)). Mutation c.1583T>C has been previously reported in PKAN patients and was absent in the patient’s mother and 100 healthy Greeks (n=200 *PANK2* genes). However, the mother was a heterozygous carrier of the novel variant c.1424T>C, which was absent in 100 healthy Greeks (n=200 *PANK2* genes), therefore, it seems that it is not a single nucleotide polymorphism but a novel mutation not previously described in *PANK2* patients.

This novel c.1424T>C mutation is not listed in the NHLBI Exome Variant database that contains many thousands of alleles.⁵ It is a missense mutation that results in the substitution of a methionine by a threonine on position 475 of the resulting protein (p.Met475Thr). In silico prediction by SIFT (“Sorting Tolerant From Intolerant,” <http://sift.jevl.org/>),⁶ PolyPhen-2 (Polymorphism Phenotyping version 2, <http://genetics.bwh.harvard.edu/pph2/>)⁷ and Mutation Taster (<http://www.mutationtaster.org/>)⁸ suggests pathogenicity (deleterious, probably damaging and disease-causing, respectively). Based on all the above-mentioned findings, the patient with the atypical PKAN had two disease-causing mutations in *PANK2* gene in combined heterozygosity.

Discussion

One of the mutations observed in our patient (p.T528M) is already known as pathogenic, and it has been suggested to be the second most common *PANK2* mutation.⁹ The other (p.M475T), to the best of our knowledge, has never been described before. It was absent in 100 healthy Greeks (n=200 *PANK2* genes), therefore, it is not a single nucleotide polymorphism, but a novel missense mutation and in silico prediction suggests pathogenicity. Although we have not determined the activity of pantothenate kinase 2 in our patient, the atypical form of the disease is usually associated with missense mutations preserving residual enzymatic activity.¹⁰ Furthermore, a catalytic defect is not obligatory for pathogenesis, since in the p.T528M and some other mutations, enzymatic activity is comparable to this of the wild type protein.⁹

In conclusion, we report a novel *PANK2* mutation in a compound heterozygote patient with atypical PKAN. Compound heterozygotes have been described with atypical PKAN.^{11–13} Interestingly, an atypical Cypriot PKAN case

had a more severe extrapyramidal phenotype with respect to this one seen in our case.¹⁴ However, since genotype–phenotype correlation is limited,¹⁰ further studies are required to elucidate whether such a genotype tends to be associated with the atypical form of the disease.

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Declaration of conflicting interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship and/or publication of this article.

Ethical approval

Our institution does not require ethical approval for reporting individual cases or case series.

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Informed consent

The patient and his mother gave informed consent for inclusion in the study and for genetic testing of PANK2.

References

- Schneider SA, Hardy J and Bhatia KP. Syndromes of neurodegeneration with brain iron accumulation (NBIA): an update on clinical presentations, histological and genetic underpinnings, and treatment considerations. *Mov Disord* 2012; 27: 42–53.
- Hayflick SJ, Westaway SK, Levinson B, et al. Genetic, clinical, and radiographic delineation of Hallervorden-Spatz syndrome. *N Engl J Med* 2003; 348: 33–40.
- Burke RE, Fahn S, Marsden CD, et al. Validity and reliability of a rating scale for the primary torsion dystonias. *Neurology* 1985; 35: 73–77.
- Goetz CG, Tilley BC, Shaftman SR, et al. Movement Disorder Society-sponsored revision of the Unified Parkinson's Disease Rating Scale (MDS-UPDRS): scale presentation and clinimetric testing results. *Mov Disord* 2008; 23: 2129–2170.
- Exome Variant Server. *NHLBI GO Exome Sequencing Project (ESP)*. Seattle, WA, April 2016, <http://evs.gs.washington.edu/EVS/>
- Kumar P, Henikoff S and Ng PC. Predicting the effects of coding non-synonymous variants on protein function using the SIFT algorithm. *Nat Protoc* 2009; 4: 1073–1081.
- Adzhubei IA, Schmidt S, Peshkin L, et al. A method and server for predicting damaging missense mutations. *Nat Methods* 2010; 7: 248–249.
- Schwarz JM, Cooper DN, Schuelke M, et al. MutationTaster2: mutation prediction for the deep-sequencing age. *Nat Methods* 2014; 11: 361–362.
- Zhang YM, Rock CO and Jackowski S. Biochemical properties of human pantothenate kinase 2 isoforms and mutations linked to pantothenate kinase associated neurodegeneration. *J Biol Chem* 2006; 281: 107–114.
- Hartig MB, Hörtnagel K, Garavaglia B, et al. Genotypic and phenotypic spectrum of PANK2 mutations in patients with neurodegeneration with brain iron accumulation. *Ann Neurol* 2006; 59: 248–256.
- Seo JH, Song SK and Lee PH. A novel PANK2 mutation in a patient with atypical pantothenate-kinase-associated neurodegeneration presenting with adult-onset parkinsonism. *J Clin Neurol* 2009; 5: 192–194.
- Pérez-González EA, Chacón-Camacho OF, Arteaga-Vázquez J, et al. A novel gene mutation in PANK2 in a patient with an atypical form of pantothenate kinase-associated neurodegeneration. *Eur J Med Genet* 2013; 56: 606–608.
- Shan J, Wen B, Zhu J, et al. Novel PANK2 gene mutations in two Chinese siblings with atypical pantothenate kinase-associated neurodegeneration. *Neurol Sci* 2013; 34: 561–563.
- Tanteles GA, Spanou-Aristidou E, Antoniou C, et al. Novel homozygous PANK2 mutation causing atypical pantothenate kinase-associated neurodegeneration (PKAN) in a Cypriot family. *J Neurol Sci* 2014; 340(1–2): 233–236.