Journal of the Neurological Sciences 357 (2015) 332–334

Contents lists available at ScienceDirect

Journal of the Neurological Sciences

journal homepage: www.elsevier.com/locate/jns

Letter to the Editor

Novel FA2H mutation in a girl with familial spastic paraplegia

Keywords:
Database
FA2H
New mutation
Nonsense
Spastic paraplegias
SPG35

To the Editor,

Autosomal recessive Hereditary spastic paraplegias (HSPs) are more frequent in consanguineous populations with a prevalence of 0.6/100,000 in Norway and up to 5.75/100,000 in Tunisia. The prevalence of all hereditary spastic paraplegia (HSPs, SPGs) ranges between 4.3 and 9.8/100,000 [1].

The key symptom of the HSP of lower limbs can be complicated by a variety of signs and symptoms including cognitive impairment, optic atrophy, cerebellar ataxia or peripheral nerve involvement. Most of the HSP (about 70%) are caused by mutations in SPAST (SPG4), ATL1 (SPG3A) and REEP1 (SPG31) genes [2,3]. Nineteen of the HSPs follow an autosomal dominant (most in SPG4 and SPG3 genes), 27 an autosomal recessive (most in SPG11 gene), 3 X-linked mode of transmission ((L1CAM (SPG1), PLP1 (SPG2), and SLC16A2 (SPG22) genes); and one a maternal trait of inheritance. Mutations in genes like (NIPA1), (SPG6), KIAA0196 (SPG8), KIF5A (SPG10), RNT2 (SPG12), SPGD1 (SPG13), BSCL2, (SPG17), REEP1 (SPG31), ZFYVE27 (SPG33), and SLC33A1 (SPG42) are reported like autosomal dominant inheritances of the HSPs. Furthermore genes (CYP7B1 (SPG5), SPG7 (SPG7), ZFYVE26 (SPG15), ERLIN2, (SPG18), SPG20 (SPG20), ACP33 (SPG21), KIF1A (SPG30), FA2H (SPG35), NTE (SPG39), GJA12/GJC2 (SPG44), KIAA0415 (SPG48) and 4 more genes encoding for the AP4-complex (SPG47) are associated with rarely autosomal recessive causes of SPG [1].

HSP type 35 (MIM# 612319) is an autosomal recessive form of the HSP caused by mutations in the FA2H gene (MIM# 611026) at 16q21–q23 chromosome, that encodes fatty acid 2-hydroxylase, spanning 372 amino acids and 7 exons; it contains a cytochrome b5 heme-binding domain, and a fatty acid hydroxylase domain encoded by exons 5 through 7. FA2H is involved in the synthesis of 2-hydroxy fatty acid galactolipids, major component of the myelin sheath [4]. Hereditary spastic paraplegias are clinically and genetically highly heterogeneous group of neurodegenerative disorders. The autosomal recessive HSPs are often associated with complex phenotype, and one of the most frequent forms is caused by mutations in the SPG11/KIAA1840 gene [3].

1. Case presentation

We report the clinical, neurophysiological, and molecular findings of a patient with a new FA2H variant, detected by NGS (Next Generation Sequencing) technologies.

Since age 3y, the patient presented alterations in psychomotor development. Lower limb spasticity and optic atrophy was noted. This patient has one brother with similar clinical characteristics and disease progression. Her parents were Pakistani cousins but have no symptoms of HSP.

The main clinical features were exposed at neurological and psychomotor development. There was no evidence of cranial, pulmonary or abdominal alterations. Brain MRI only showed atrophy of the cerebellar vermis. The patient presented mild mental retardation and a mildly decreased muscle tone in upper limbs. No more clinical data were available.

2. Materials and methods

After receiving parents' informed consent, DNA was extracted from peripheral blood by organic Phenol:Chloroform:Isoamyl Alcohol procedure. Extracted DNA was quantified by a 0,8% agarose gel. In a first analysis Sanger sequencing was performed in PLP1 gene, secondly NGS in all genes related to autosomal dominant, recessive and X-linked patterns of HSPs were analysed. NGS was performed following protocols of HaloPlex Target Enrichment System for Illumina sequencing (ultrasequencing in MiSeq (Illumina) platform) and analysed by DNAnexus software. Reference sequences were used from data of HGMD (The Human Gene Mutation Database) for each of the analysed genes.

3. Results

PLP1 gene has been included after doctor's recommendations but no relevant data has been found in the analysis of this gene. For that reason we performed a NGS analysis. However, there were no point mutations results in most of the genes analysed by NGS (SPG3A, BSCL2, HSPD1, KIF5A, NIPA1, REEP1, RTN2, SLC33A1, SNTB1, SPAST, ZFYVE27, AP4B1, AP4E1, AP4M1, AP4S1, AP5Z1, C120RF65, CYP2U1, CYP7B1, DDHD2, ERLIN2, GJC2, KIF1A, PNPLA6, ALS2, ZFYVE27, SPG11, SPG20, SPG21, SPG7, TECPR2, VPS37A, L1CAM, PLP1, SLC16A2 and KDM5C). There was only relevant data of NGS at FA2H gene.

The sequence analysis of the FA2H gene revealed the novel nonsense mutation c.565 (c.565 C > T) in homozygosis; this mutation generates an Arginine to Stop Codon (p.R189X) exchange (Fig. 1 and Table 1). These kinds of mutations are classified as pathogenic because they encode a truncated protein by a premature stop codon of the protein; so they are considered good clinical tools for diagnosis and genetic counseling [5].

http://dx.doi.org/10.1016/j.jns.2015.07.042
0022-510X/

Fig. 1. Illustration of mutation visualization of NGS procedure. At the top of the figure we can see the exact chromosomal location, its cytogenetics location and exact position in the chromosome. Just below we see the reference sequence. In blue and green colour we observe the coverage in reads measurements. In this position it is read for more than 200 reads. In black we see the variant that the program detected and is highlighted in black. In this case both are A so the variant is in homozygosis (show at the bottom in grey).

4. Discussion

We identified a novel nonsense mutation in the FA2H gene in a patient with severe case of HSP. We are the first describing the homozygous effect of c.565 C > T mutation as the cause of HSP. A recent whole exome sequencing analysis reported 2 variants in fatty acid 2-hydroxylase (FA2H) gene: c.169_170insGCGGGCCAGG (p.Asp57Glyfs*66), leading, if translated, to a truncated protein, and the variant c.117C > G (p.Phe39Leu). It was described in a 21 years old woman presented with progressive spastic paraplegia, dysarthria, and strabismus since 7 years of age. Brain MRI disclosed white matter changes and iron accumulation [6]. Others latest articles have reported new missense variants in analysis of family cases like c.353G > A (p.Arg118Gln) in ATL1 gene; and, c.1837G > C (p.Asp613His) in SPAST gene; but there are not many updates at nonsense ones [7,8].

Nonsense mutations in the FA2H gene (fatty acid 2-hydroxylase) have been recently shown to be associated with leukodystrophy with spastic paraparesis and dystonia, complicated spastic paraparesis SPG35 and neurodegeneration with brain iron accumulation [9]. Furthermore, nonsense mutations in FA2H gene cause the absence of the protein product and lead to clinical severe manifestations some of them like dysphagia, optic atrophy, seizures, severe cognitive decline and severe leukodystrophy (e.g., c.509_510delAC and p.Y170X), whereas missense mutations may cause milder phenotypic manifestations [9].

One problem of HSPs is that the clinical spectrum and the onset age vary widely. Increasing awareness about this group of clinically and genetically diverse inherited disorders is especially important for patients and researchers nowadays, since there is rapid progression on the field. Detection of new causative genes, proteins and its mechanisms may bring treatment perspectives in the near future as well as provide clues about other similar neurodegenerative diseases of the motor system [10]. In conclusion, having an updated database in nonsense variants related to HSPs will offer more clinical advantages to carriers, who are considered as having a more aggressive development of HSP.

Conflict of Interest

The authors declare no financial or other conflicts of interest.

References

[1] J. Finsterer, W. Löscher, S. Quasthoff, J. Wanschitz, M. Auer-Grumbach, G. Stevanin, Hereditary spastic paraplegias with autosomal dominant, recessive, X-linked, or maternal trait of inheritance, J. Neurol. Sci. 318 (1) (2012) 1–18.

Table 1
Summary of NGS details in FAH2 gene.

Chr	Pos	Rf	Alle	zyg	Qual	Cov	Allele count	Allele_freq	Gene	Effect	HGVS-cdna	HGVS-prot	Codon	ENSP	ENSG	ENST	Exon
16	74760171	G	A	Hom	25074.10	805	797	0.99	FA2H	stop_gained	c.565C > T	p.Arg189Ter	Cga/Tga	ENSP00000219368	ENSG00000103089	ENST00000219368	4/7

Abbreviations: Chr: chromosome; pos: position; Rf: reference; Alle: allele; cov: coverage; qual: quality.

[2] M. Muglia, L. Citrigno, E. D'Errico, A. Magariello, E. Distaso, A. Gasparro, et al., A novel KIF5A mutation in an Italian family marked by spastic paraparesis and congenital deafness, J. Neurol. Sci. 343 (1) (2014) 218–220.
[3] V. Pensato, B. Castellotti, C. Gellera, D. Pareyson, C. Ciano, L. Nanetti, et al., Overlapping phenotypes in complex spastic paraplegias SPG11, SPG15, SPG35 and SPG48, Brain 137 (Pt. 7) (2014 Jul) 1907–1920.
[4] R. Rupps, J. Hukin, M. Balicki, S. Mercimek-Mahmutoglu, A. Rolfs, C. Dias, Novel mutations in FA2H-associated neurodegeneration: an Underrecognized Condition? J. Child Neurol. 28 (11) (2013 Nov) 1500–1504.
[5] E. Borràs, M. Pineda, A. Brieger, I. Hinrichsen, C. Gómez, M. Navarro, et al., Comprehensive functional assessment of MLH1 variants of unknown significance, Hum. Mutat. 33 (11) (2012) 1576–1588.
[6] J.L. Pedroso, B.W. Handfas, A. Abrahao, F. Kok, O.G. Barsottini, A.S. Oliveira, Fatty acid 2-hydroxylase deficiency: clinical features and brain iron accumulation, Neurology 84 (9) (2015 Mar 3) 960–961.
[7] A. Aulitzky, K. Friedrich, D. Gläser, R. Gastl, C. Kubisch, A.C. Ludolph, et al., A complex form of hereditary spastic paraplegia in three siblings due to somatic mosaicism for a novel SPAST mutation in the mother, J. Neurol. Sci. 347 (1) (2014) 352–355.
[8] T.N. Khan, J. Klar, M. Tariq, S.A. Baig, N.A. Malik, R. Yousaf, et al., Evidence for autosomal recessive inheritance in SPG3A caused by homozygosity for a novel ATL1 missense mutation, Eur. J. Hum. Genet. 22 (2014) 1180–1184.
[9] C. Garone, T. Pippucci, D.M. Cordelli, R. Zuntini, G. Castegnaro, C. Marconi, et al., FA2H-related disorders: a novel c. 270 3A > T splice-site mutation leads to a complex neurodegenerative phenotype, Dev. Med. Child Neurol. 53 (10) (2011) 958–961.
[10] I. Faber, K.R. Servelhere, A.R. Martinez, A. DAbreu, I. Lopes-Cendes, M.C. Franca Jr., Clinical features and management of hereditary spastic paraplegia, Arq. Neuropsiquiatr. 72 (3) (2014) 219–226.

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25 March 2015