

CASE REPORT

A Novel Disease-Causing ASPA Gene Mutation (c.432+1 G>C) in an Iranian Patient with Canavan Disease: A Case Report*

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Abstract

Canavan disease is an autosomal recessive genetic disease and rare fatal childhood neurological disorder caused by mutations in the *ASPA* gene, which resulted in a catalytic deficiency of the *ASPA* enzyme that catalyzes the hydrolysis of N-acetylaspartic acid into aspartate and acetate. Herein, we report an Iranian patient diagnosed with Canavan disease with a novel splice-site mutation in the *ASPA* gene (NM_000049.4; c.432+1 G>C). This report is based on a homozygous c.432+1 G>C mutation in the *ASPA* gene identified from an Iranian patient. As a result, a novel homozygous pathogenic mutation on *ASPA* is the cause of disease in the patient. (**International Journal of Biomedicine. 2021;11(4):594-597.**)

Key Words: Canavan disease • novel mutation • ASPA gene • aspartoacylase • N-acetylaspartic acid

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Abbreviations

NAA, N-acetylaspartic acid; MRS, magnetic resonance spectroscopy; MRI, magnetic resonance imaging; gDNA, genomic DNA; WES, whole-exome sequencing; ACMG, American College of Medical Genetics and Genomics.

Introduction

Canavan disease is an autosomal-recessive leukodystrophy and fatal neurological disease which is characterized by developmental delay, neurologic deterioration with severe intellectual disability, and early death.⁽¹⁾ The underlying cause of this disease is the deficiency in the enzyme aspartoacylase, which leads to high levels of N-acetylaspartic acid (NAA) in

the urine, brain, and body fluids. The *ASPA* gene mutations are responsible for this deficiency (RefSeq NM_000049.4).⁽²⁾ *ASPA* is a catabolic enzyme that is primarily in oligodendrocytes in the central nervous system.⁽³⁾ *ASPA* catalyzes the hydrolysis of NAA to generate aspartate and acetate, it is a homodimer and essential in the synthesis of myelin. Patients who are deficient in the *ASPA* enzyme activity have an abnormal elevation of NAA in the brain. This can be identified by applying magnetic resonance spectroscopy (MRS) even before increasing its concentration in the urine, which is suitable for the early diagnosis of Canavan disease.⁽⁴⁾ Clinical symptoms are not manifested at the time of birth; however, the clinical triad of hypotonia, macrocephaly, and head lag often in association with macrocephaly and

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P181L, 244delAT, 923delT, C152W, V14G, D249V, and E214X with a severe phenotype. A stop codon or frameshift occurs in many of these mutations that is related to the onset during the first few months after birth (2 or 3 months of age). In Jewish populations, E285A and Y231X mutations are correlated with a severe phenotype as well.⁽²⁰⁾ The mutation that was found in our patient also cause a severe phenotype.

Deficiency of the *ASPA* activity caused by the nonsense tyr231ter, the missense ala305glu mutation, or the glu285ala mutation establishes that the three coding-sequence mutations are the cause of Canavan disease.⁽²¹⁾ The 433-2 A to G transition in intron 2 (in the splice acceptor site) would result in skipping of exon 3. Additionally, skipping of 94-base exon 3, in the final transcript will change the reading frame. A frameshift accompanied by an exon-skipping would result in the aspartoacylase deficiency.⁽²²⁾

In 2012, Durmaz et al. reported a novel heterozygous mutation Y88X (T to A nucleotide change at codon 88 in exon 2) within the aspartoacylase gene in a consanguineous family with an affected child diagnosed as Canavan disease. This mutation converts the codon for tyrosine (TAT) into a premature termination codon (TAA).⁽²⁰⁾ Also, in 2015, Ashrafi et al. indicated a novel homozygous missense mutation (c.202G>A) in the *ASPA* gene in exon 1 which was found in an Iranian patient.⁽²³⁾ In our case, we presented a novel homozygous pathogenic mutation in *ASPA* gene (c.432+1 G>C) related to Canavan disease. This mutation was at the 5' splice-site beginning intron 2, which can cause mis-splicing and alter the reading frame, and consequently, it will probably result in a serious alteration in ASPA protein conformation and leads to the Canavan phenotype. This type of mutation has not been reported in other populations.

Conclusion

In the present study, we report a 2-year-old Iranian boy with severe Canavan disease who harbors a novel pathogenic homozygous mutation (c.432+1 G>C) in the *ASPA* gene. Homozygous mutation as in the intron 2 of *ASPA* gene in the present case is a novel splice-site mutation that was not reported elsewhere. The mutation that leads to the Canavan disease has been defined in the family; it would make prenatal diagnosis possible and suggest parents with such disorder plan for the next pregnancy.

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Competing Interests

The authors declare that they have no competing interests.

Disclaimers

The views expressed in this article are the author's own and do not reflect the official position of the institutions.

References

1. Pleasure D, Guo F, Chechneva O, Bannerman P, McDonough J, Burns T, Wang Y, Hull V. Pathophysiology and Treatment of Canavan Disease. *Neurochem Res.* 2020 Mar;45(3):561-565. doi: 10.1007/s11064-018-2693-6.
2. Zaki OK, Krishnamoorthy N, El Abd HS, Harche SA, Mattar RA, Al Disi RS, Nofal MY, El Bekay R, Ahmed KA, George Priya Doss C, Zayed H. Two patients with Canavan disease and structural modeling of a novel mutation. *Metab Brain Dis.* 2017 Feb;32(1):171-177. doi: 10.1007/s11011-016-9896-9.
3. Madhavarao CN, Moffett JR, Moore RA, Viola RE, Namboodiri MA, Jacobowitz DM. Immunohistochemical localization of aspartoacylase in the rat central nervous system. *J Comp Neurol.* 2004 May 3;472(3):318-29. doi: 10.1002/cne.20080.
4. Gujar SK, Maheshwari S, Björkman-Burtscher I, Sundgren PC. Magnetic resonance spectroscopy. *J Neuroophthalmol.* 2005 Sep;25(3):217-26. doi: 10.1097/01.wno.0000177307.21081.81.
5. Hoshino H, Kubota M. Canavan disease: clinical features and recent advances in research. *Pediatr Int.* 2014 Aug;56(4):477-83. doi: 10.1111/ped.12422.
6. Leone P, Shera D, McPhee SW, Francis JS, Kolodny EH, Bilaniuk LT, Wang DJ, Assadi M, Goldfarb O, Goldman HW, Freese A, Young D, Doring MJ, Samulski RJ, Janson CG. Long-term follow-up after gene therapy for canavan disease. *Sci Transl Med.* 2012 Dec 19;4(165):165ra163. doi: 10.1126/scitranslmed.3003454.
7. Janson CG, McPhee SW, Francis J, Shera D, Assadi M, Freese A, Hurh P, Haselgrove J, Wang DJ, Bilaniuk L, Leone P. Natural history of Canavan disease revealed by proton magnetic resonance spectroscopy (1H-MRS) and diffusion-weighted MRI. *Neuropediatrics.* 2006 Aug;37(4):209-21. doi: 10.1055/s-2006-924734.
8. Janson CG, Kolodny EH, Zeng BJ, Raghavan S, Pastores G, Torres P, Assadi M, McPhee S, Goldfarb O, Saslow B, Freese A, Wang DJ, Bilaniuk L, Shera D, Leone P. Mild-onset presentation of Canavan's disease associated with novel G212A point mutation in aspartoacylase gene. *Ann Neurol.* 2006 Feb;59(2):428-31. doi: 10.1002/ana.20787.
9. Mendes MI, Smith DE, Pop A, Lennertz P, Fernandez Ojeda MR, Kanhai WA, et al. Clinically Distinct Phenotypes of Canavan Disease Correlate with Residual Aspartoacylase Enzyme Activity. *Hum Mutat.* 2017 May;38(5):524-531. doi: 10.1002/humu.23181.
10. Sheikh-Hosseini M, Moarefzadeh M, Alavi-Moghaddam H, Morovvati S. A Novel Mutation in Aicardi-Goutières' Syndrome: A Case Report. *Journal of Pediatric Neurology.* 2021;19(01):050-3.
11. Arjmand B, Larijani B, Sheikh Hosseini M, Payab M, Gilany K, Goodarzi P, Parhizkar Roudsari P, Amanollahi Baharvand M, Hoseini Mohammadi NS. The Horizon of Gene Therapy in Modern Medicine: Advances and Challenges. *Adv Exp Med Biol.* 2020;1247:33-64. doi: 10.1007/5584_2019_463.
12. Baslow MH, Guilfoyle DN. Canavan disease, a rare early-onset human spongiform leukodystrophy: insights into its genesis and possible clinical interventions. *Biochimie.* 2013 Apr;95(4):946-56. doi: 10.1016/j.biochi.2012.10.023.
13. Hershfield JR, Pattabiraman N, Madhavarao CN, Namboodiri MA. Mutational analysis of aspartoacylase:

- implications for Canavan disease. *Brain Res.* 2007 May 7;1148:1-14. doi: 10.1016/j.brainres.2007.02.069.
14. Hussain R, Daud S, Kakar N, Ahmad A, Baloch AH, Tareen AM, Kakar MA, Ahmad J. A missense mutation (p.G274R) in gene ASPA causes Canavan disease in a Pakistani family. *Mol Biol Rep.* 2012 May;39(5):6197-201. doi: 10.1007/s11033-011-1438-2.
15. Wijayasinghe YS, Pavlovsky AG, Viola RE. Aspartoacylase catalytic deficiency as the cause of Canavan disease: a structural perspective. *Biochemistry.* 2014 Aug 5;53(30):4970-8. doi: 10.1021/bi500719k.
16. Zeng BJ, Pastores GM, Leone P, Raghavan S, Wang ZH, Ribeiro LA, Torres P, Ong E, Kolodny EH. Mutation analysis of the aspartoacylase gene in non-Jewish patients with Canavan disease. *Adv Exp Med Biol.* 2006;576:165-73; discussion 361-3. doi: 10.1007/0-387-30172-0_11.
17. Eke GH, Iscan A, Cece H, Calik M. A mutation of aspartoacylase gene in a Turkish patient with Canavan disease. *Genet Couns.* 2012;23(1):9-12.
18. Di Pietro V, Cavallari U, Amorini AM, Lazzarino G, Longo S, Poggiani C, Cavalli P, Tavazzi B. New T530C mutation in the aspartoacylase gene caused Canavan disease with no correlation between severity and N-acetylaspartate excretion. *Clin Biochem.* 2013 Dec;46(18):1902-4. doi: 10.1016/j.clinbiochem.2013.09.004.
19. Bley A, Denecke J, Kohlschütter A, Schön G, Hischke S, Guder P, Bierhals T, Lau H, Hempel M, Eichler FS. The natural history of Canavan disease: 23 new cases and comparison with patients from literature. *Orphanet J Rare Dis.* 2021 May 19;16(1):227. doi: 10.1186/s13023-020-01659-3.
20. Durmaz AA, Akin H, Onay H, Vahabi A, Ozkinay F. A novel aspartoacylase (ASPA) gene mutation in Canavan disease. *Fetal Pediatr Pathol.* 2012 Aug;31(4):236-9. doi: 10.3109/15513815.2011.650292.
21. Kaul R, Balamurugan K, Gao GP, Matalon R. Canavan disease: genomic organization and localization of human ASPA to 17p13-ter and conservation of the ASPA gene during evolution. *Genomics.* 1994 May 15;21(2):364-70. doi: 10.1006/geno.1994.1278.
22. Kaul R, Gao GP, Aloya M, Balamurugan K, Petrosky A, Michals K, Matalon R. Canavan disease: mutations among Jewish and non-Jewish patients. *Am J Hum Genet.* 1994 Jul;55(1):34-41.
23. Ashrafi M, Tavasoli A, Katibeh P, Aryani O, Vafaei-Shahi M. A Novel Mutation in Aspartoacylase Gene; Canavan Disease. *Iran J Child Neurol.* 2015 Fall;9(4):54-7.
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