Case Report
J Genet Med 2022;19(1):38-41
https://doi.org/10.5734/JGM.2022.19.1.38
ISSN 1226-1769 (Print) 2383-8442 (Online)

JGM Journal of Genetic Medicine

Check for updates

A case of mild CADASIL patient with a novel heterozygous *NOTCH3* variant

WooChan Choi[1], Yang-Ha Hwang[1,2], and Jong-Mok Lee[1,2,*]
[1]Department of Neurology, Kyungpook National University Hospital, Daegu, Korea
[2]Department of Neurology, School of Medicine, Kyungpook National University, Daegu, Korea

Cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL) is a single-gene disease caused by mutations in the neurogenic locus notch homolog protein 3 (*NOTCH3*) gene. The spectrum of clinical manifestations is broad, ranging from asymptomatic to typical ischemic stroke, and mainly depends on the location of the mutations. We describe the case of a 76-year-old female without apparent neurological deficits. However, brain magnetic resonance imaging revealed confluent lesions in the white matter. Direct sequencing of the *NOTCH3* gene revealed a novel pathogenic mutation, c.811T>A, which results in a mild phenotype. Therefore, this report will expand the current knowledge in regards to the mutations that can cause CADASIL.

Key words: Leukoencephalopathy, Ischemic stroke, CADASIL.

Introduction

Cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL, OMIM #125310) is a well-known single-gene disease associated with ischemic stroke and is caused by autosomal dominant mutations in the neurogenic locus notch homolog protein 3 (*NOTCH3*) gene [1,2]. Clinical manifestations of CADASIL include cerebral ischemic episodes, cognitive deficits, migraines with aura, and psychiatric disturbances [1]. Since the first identification of *NOTCH3* mutations in 1996, more than 200 mutations have been ascertained [1,3,4]. The *NOTCH3* gene comprises 33 exons that are translated into a 2321-amino acid protein [2]. Most pathogenic variants are located in epidermal growth factor-like repeats (EGFRs), which can be categorized into two groups, EGFR domains 1-6 and 7-34. The two domains are associated with different phenotypic severity [1]. Specifically, pathogenic mutations in EGFR domains 1-6 are linked to severe phenotypes [1]. Here, we report a mild CADASIL case with a novel mutation in EGFR domain 6 of the *NOTCH3* gene.

Case

The proband, a 76-year-old female, visited our neurology department with complaints of recurrent dizziness. Her medical history included hyperlipidemia, and she was not a smoker or alcohol consumer. She did not present with hypertension, cognitive function impairment, or psychiatric symptoms. In addition, neurological examination revealed no obvious abnormalities, with normal cerebellar and extraocular movements. Blood tests revealed hyperlipidemia, with high serum levels of total cholesterol (222 mg/dL, normal <200 mg/dL) and triglycerides (253

Received: 5 November 2021, Revised: 5 December 2021, Accepted: 5 December 2021, Published: 30 June 2022
*Corresponding author: Jong-Mok Lee, M.D., Ph.D. https://orcid.org/0000-0002-2918-6166
Department of Neurology, School of Medicine, Kyungpook National University, 680 Gukchaebosang-ro, Jung-gu, Daegu 41944, Korea.
Tel: +82-53-200-5765, Fax: +82-53-422-4265, E-mail: azulmar@gmail.com
Conflict of interest: The authors declare that they do not have any conflicts of interest.

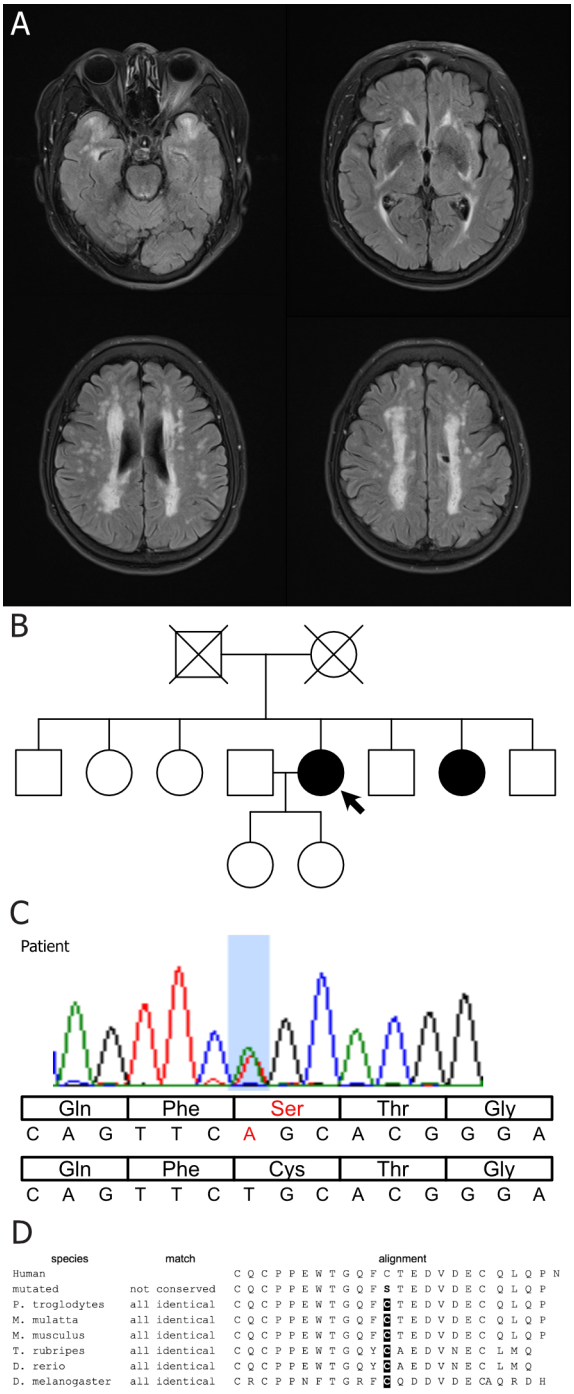

Fig. 1. Brain magnetic resonance imaging (MRI), pedigree, and results of genetic analysis from the proband. (A) Brain MRI revealed bilateral hyperintensities in the periventricular white matter, anterior temporal lobe, as well as the internal and external capsules. Multiple lacunar infarctions are noted in the deep white matter, basal ganglia, and thalamus on fluid-attenuated inversion recovery imaging. (B) Other family members did not complain of cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy symptoms except for the proband and her younger sister. (C) The Sanger sequencing chromatogram reveals a nucleotide substitution from thymine to adenine at position 811 of the *NOTCH3* gene (c.811T>A, p.Cys271Ser, NM_000435.2). (D) Cysteine at position 271 in the NOTCH3 protein is highly conserved throughout species.

mg/dL, normal <200 mg/dL). To investigate the causes of dizziness, brain magnetic resonance imaging (MRI) was performed. T2-weighted and fluid-attenuated inversion recovery images revealed a large confluent area of hyperintensity throughout the periventricular area, deep white matter, and subcortical white matter. Furthermore, the lesions also involved the anterior temporal lobes as well as the internal and external capsules in both hemispheres (Fig. 1A). Asymptomatic lacunar infarcts were observed in the deep white matter, basal ganglia, and thalamus (Fig. 1A). However, because abnormal MRI signal patterns might not be indicative of a specific pathology, the patient was not diagnosed. At a later stage, the patient's younger sister was diagnosed with CADASIL, while other family members, including two of the patient's daughters, did not complain of symptoms of CADASIL (Fig. 1B).

Based on the MRI findings and family history, we decided to perform direct sequencing of the *NOTCH3* gene. A novel heterozygous missense mutation, c.811T>A (p.Cys271Ser, exon 6, NM_000435.2, Fig. 1C) was detected, which was not present in the Genome Aggregation Database, the 1000 Genomes Projects database, or the Leiden Open Variation Database (https://databases.lovd.nl/shared/genes/NOTCH3). Furthermore, this variant was predicted to be disease-causing by the MutationTaster system (http://www.mutationtaster.org) and categorized as "likely pathogenic", satisfying two moderate and two supporting pieces of evidence based on the American College of Medical Genetics and Genomics/Association for Molecular Pathology guidelines [5]. In addition, cysteine at position 271 of the *NOTCH3* protein is highly conserved across species (Fig. 1D). The same mutation in the *NOTCH3* gene was detected in the patient's younger sister, who complained of headaches.

Discussion

For the patient herein described, CADASIL was initially suspected based on the abnormal MRI signal findings and family history and was confirmed by direct sequencing, which detected a novel mutation in the *NOTCH3* gene, c.811T>A (p.Cys271Ser). This variation was predicted to be pathogenic by *in silico* testing. Different amino acid substitutions at the same position (c.812G>T, p.Cys271Phe) have also been reported to be pathogenic [6].

NOTCH3, a large gene containing 33 exons, encodes a single-pass transmembrane receptor which comprises 34 EGFRs and an intracellular domain. The NOTCH3 protein is mainly located

in vascular smooth muscle cells and pericytes [1]. All CADASIL pathogenic variants are reported to be located in EGFRs, and most of them are missense variants that lead to an amino acid substitution at cysteine residues located at odd-number positions [2,7]. Such mutations may lead to disruption of disulfide bonds between cysteines which, in turn, may result in a conformational change that could alter the NOTCH3 protein aggregation pattern [2,7]. Specifically, the pathological NOTCH3 protein accumulation observed in systemic vessels seems to support this prediction [2]. The c.811T>A variant detected in our patient was also predicted to cause the loss of disulfide bonds located at the corresponding cysteine residue at position 262.

Concerning the genotype-phenotype correlation, the determinants of clinical severity in CADASIL remain unknown [1,6-8]. However, several factors seem to be associated with clinical severity, including the location of the mutation, whether the variant affects cysteine residues, as well as other cardiovascular risk factors [1,6-8]. Specifically, a severe disease course is associated with variants in EGFR domains 1-6, whereas mutations located in the EGFR domains 7-34 are associated with milder phenotypes [1]. This is reflected at the neuroanatomical level, as patients with pathogenic variants in EGFR domains 1-6 showed more severe white matter hyperintensity on brain MRI and an earlier onset of stroke than those with mutations in EGFR domains 7-34 [1]. In addition, patients carrying cysteine-affecting variants reported to show more severe clinical severity than those carrying cysteine-sparing variants [7,8]. Patients with the cystine-sparing p.Arg75Pro mutation showed a significantly lower stroke frequency and white matter hyperintensity than patients with p.Arg141Cys or p.Arg182Cys, although all three such mutations were located in EGFR domains 1-6 [7]. According to these findings, our patient, who presented with a cystine-affecting mutation, was expected to show a severe phenotype but only had mild symptoms.

Other factors involved in the clinical presentation of CADASIL are hypertension and smoking [9]. The presence of hypertension and longer duration of smoking years were associated with an increased risk of stroke [9]. However, other factors besides mutations or cardiovascular risk factors may be at play and thus need to be discovered [10].

Concerning the radiological findings of CADASIL, white matter hyperintensities are one of the radiological signs of the disease. Recent studies have recognized that the involvement of the anterior temporal lobe and external capsules may be useful in differentiating CADASIL from other forms of small vessel disease [11]. In particular, the anterior temporal lobe has a much higher specificity (86%) than the external capsules (45%) in identifying CADASIL, while the sensitivity of these two brain regions is approximately the same (89% vs. 93%, respectively) [12]. This characteristic hyperintensity was also observed in both the anterior temporal lobe and external capsules of our patient.

In conclusion, we report a novel mutation in *NOTCH3* associated with a mild CADASIL phenotype that includes dizziness or headaches. Because of such mild presentation, CADASIL was initially not suspected, and the patient diagnosis was delayed. Therefore, in the cases where an unknown white matter disease is detected, physicians should be aware of CADASIL and perform direct sequencing to ensure timely treatment administration.

Authors' Contributions

Conception and design: JML. Acquisition of data: WCC, YHH. Analysis and interpretation of data: WCC, JML. Drafting the article: WCC, JML. Critical revision of the article: JML. Final approval of the version to be published: all authors.

References

1. Rutten JW, Van Eijsden BJ, Duering M, Jouvent E, Opherk C, Pantoni L, et al. Correction: The effect of NOTCH3 pathogenic variant position on CADASIL disease severity: NOTCH3 EGFr 1-6 pathogenic variant are associated with a more severe phenotype and lower survival compared with EGFr 7-34 pathogenic variant. Genet Med 2019;21:1895.
2. Wang T, Baron M, Trump D. An overview of Notch3 function in vascular smooth muscle cells. Prog Biophys Mol Biol 2008;96:499-509.
3. Tikka S, Baumann M, Siitonen M, Pasanen P, Pöyhönen M, Myllykangas L, et al. CADASIL and CARASIL. Brain Pathol 2014;24:525-44.
4. Joutel A, Vahedi K, Corpechot C, Troesch A, Chabriat H, Vayssière C, et al. Strong clustering and stereotyped nature of Notch3 mutations in CADASIL patients. Lancet 1997;350:1511-5.
5. Richards S, Aziz N, Bale S, Bick D, Das S, Gastier-Foster J, et al.; ACMG Laboratory Quality Assurance Committee. 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. Genet Med 2015;17:405-24.
6. Au KM, Li HL, Sheng B, Chow TC, Chen ML, Lee KC, et al. A novel mutation (C271F) in the Notch3 gene in a Chinese man with cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy. Clin Chim Acta 2007;376:229-32.

7. Mukai M, Mizuta I, Watanabe-Hosomi A, Koizumi T, Matsuura J, Hamano A, et al. Genotype-phenotype correlations and effect of mutation location in Japanese CADASIL patients. J Hum Genet 2020;65:637-46.
8. Hu Y, Sun Q, Zhou Y, Yi F, Tang H, Yao L, et al. *NOTCH3* variants and genotype-phenotype features in Chinese CADASIL patients. Front Genet 2021;12:705284.
9. Adib-Samii P, Brice G, Martin RJ, Markus HS. Clinical spectrum of CADASIL and the effect of cardiovascular risk factors on phenotype: study in 200 consecutively recruited individuals. Stroke 2010;41:630-4.
10. Gravesteijn G, Hack RJ, Opstal AMV, van Eijsden BJ, Middelkoop HAM, Rodriguez Girondo MDM, et al. Eighteen-year disease progression and survival in CADASIL. J Stroke 2021;23:132-4.
11. O'Sullivan M, Jarosz JM, Martin RJ, Deasy N, Powell JF, Markus HS. MRI hyperintensities of the temporal lobe and external capsule in patients with CADASIL. Neurology 2001;56:628-34.
12. Markus HS, Martin RJ, Simpson MA, Dong YB, Ali N, Crosby AH, et al. Diagnostic strategies in CADASIL. Neurology 2002;59:1134-8.