A Second Case With the V374A KCND3 Pathogenic Variant in an Italian Patient With Early-Onset Spinocerebellar Ataxia

Flavia Palombo, PhD, Chiara La Morgia, MD, PhD, Claudio Fiorini, PhD, Leonardo Caporali, PhD, Maria Lucia Valentino, MD, Vincenzo Donadio, MD, PhD, Rocco Liguori, MD, and Valerio Carelli, MD, PhD

Neurol Genet 2022;8:e200004. doi:10.1212/NXG.0000000000200004

Abstract

Background and Objectives
To date, approximately 20 heterozygous mainly loss-of-function variants in KCND3 have been associated with spinocerebellar ataxia (SCA) type 19 and 22, a clinically heterogeneous group of neurodegenerative disorders. We aimed at reporting the second patients with the V374A KCND3 mutation from an independent family, confirming its pathogenic role.

Methods
We describe the clinical history of a patient with SCA and conducted genetic investigations including mitochondrial DNA analysis and exome sequencing.

Results
This male patient was reported to have unstable gait with tremors at the lower limbs and dysarthric speech since childhood. A neurologic examination also showed dysarthria, nystagmus, action tremor, dysmetria, and weak deep tendon reflexes. He had marked cerebellar atrophy at brain MRI, more evident at vermis. Molecular analysis, including exome sequencing and an in silico panel analysis of genes associated with SCA, revealed the c.1121T>C [p.V374A] mutation in KCND3.

Discussion
This report consolidates the pathogenicity of the V374A KCND3 mutation and suggests that the ataxic paroxysmal exacerbations are not a key phenotypic feature of this mutation.
**Case Report**

This male patient, now aged 50 years, with a negative family history, had unstable gait with tremors at the lower limbs and dysarthric speech since childhood. Our first evaluation was at age 40 years. A neurologic examination showed dysarthria, nystagmus in all directions of gaze, action tremor, dystmetria, weak deep tendon reflexes, truncal ataxia, ataxic gait, and instability in upright position with a positive Romberg sign. He also had bilateral pes cavus. He had marked cerebellar atrophy at brain MRI, more evident at vermis (Figure, A–C). Somatosensory-evoked potential displayed slightly increased time in cortical responses. At the last cognitive evaluation in 2017, the Mini-Mental Status Exam score and the score at the Brief Test of Cognition (IQ at the lower range of the normal values) were normal. At the Brief Neuropsychological Examination test, the patient presented a mild intellectual disability with abnormalities in attentive/executive functions, unchanged compared with the cognitive evaluation in 2010. The abnormalities in executive/praxic functions are likely due to a deficit in planning strategies. EMG, EEG, and ECG findings and audiometry and ophthalmologic examination findings were normal. Polysomnography revealed the presence of moderate sleep apnea syndrome. The patient is overweight (weight: 125...
kg). Initially investigated for mitochondrial disease, lactic acid was normal, whereas 3 cytochrome c oxidase (COX)-negative fibers were noted at muscle biopsy (Figure, D and E). A complete sequencing of mitochondrial DNA (mtDNA) extracted from skeletal muscle did not show any pathogenic variant (haplogroup U2e2a1c) and pathologic accumulation of macrodeletions. Finally, the mtDNA copy number assessment was normal. The only noticeable result was a relative increase of 7S DNA. Assessment of coenzyme Q in muscle biopsy was also normal.

Genetic investigation was then expanded to exome sequencing, and an in silico panel analysis of genes associated with spinocerebellar ataxia (SCA) revealed the presence of a heterozygous missense variant, c.1121T>C [p.V374A], in KCND3 (NM_004980.5). This variant has not been reported in the gnomAD database and was predicted to be damaging with a 24.4 CADD-PHRED score. According to the American College of Medical Genetics classification, the c.1121T>C variant was classified as likely pathogenic with the PM1, PM2, PP2, and PP3 criteria. Segregation analysis was consistent with a possible de novo origin of the V374A variant in the patient because available relatives tested were negative and neurologic disturbances were never reported in the parents and siblings (Figure, F).

**Data Availability**

Anonymized data not published here will be made available by request from any qualified investigator.

**Discussion**

We reported an independent case of SCA associated with the V374A variant in KCND3 gene. The cases originally described were a mother and a son (index case) diagnosed with adult-onset slowly progressive cerebellar ataxia, bradyphrenia, and normal general intellectual ability despite low results in cognitive tests. Both presented cerebellar atrophy, more severe in the index case, with moderate-to-severe cerebellar hypometabolism (Table 1). Notably, the index case experienced paroxysmal ataxia exacerbations responsive to acetazolamide when exposed to accelerations/decelerations. The V374A pathogenicity was confirmed in vitro: electrophysiology studies showed that the V374A variant was nonfunctional and caused a conductance reduction predicted to generate an increased Purkinje neuron firing frequency. This family presented an additional A671V variant in the KCNC3 gene (SCA13), for which a potential synergistic effect was excluded in vitro.

Our patient, differently from those described by Paucar et al., was affected by early-onset cerebellar ataxia, which progressively worsened, apparently without paroxysmal exacerbations. Notably, mild abnormalities in cognitive testing were also observed. The finding of rare COX-negative fibers and increased 7S DNA may be envisaged as secondary reflection on mitochondrial metabolism due to dysfunctional energy-consuming ion channeling. To date, approximately 20 mutations SCA19/22 have been described in patients with heterogeneous clinical presentations, mainly including cerebellar ataxia, cognitive dysfunction, and movement disorders such as parkinsonism (Table 2).

In conclusion, our case consolidates the pathogenicity of this mutation, with a substantially overlapping phenotype except for the paroxysmal exacerbations, for which a synergistic effect of the A671V in KCNC3 gene cannot be completely excluded, and the onset of the disease.
Study Funding
Supported by the "Ricerca Corrente" funding (F.P., C.F., L.C. and V.C.), from the Italian Ministry of Health.

Disclosure
F. Palombo reports no disclosures. C. La Morgia reports Consultancies for Chiesi Farmaceutici, Regulatory Pharma Net, and Thenewway srl; speaker honoraria from Santhera Pharmaceuticals, Chiesi Farmaceutici, Regulatory Pharma Net, Thenewway srl, First Class srl, and Biologix; and PI/SI for clinical trials sponsored by GenSight Biologics and Santhera. C. Fiorini, L. Caporali, M.L. Valentino, and V. Donadio report no disclosures. R. Ligori acts as a scientific consultant in boards of Argenx BV, Alexion Pharma Italy s.r.l., and UCB Pharma S.p.A. and received speaker honoraria from Amicus Therapeutics s.r.l. and Editree s.r.l. V. Carelli acts as a scientific consultant in boards of GenSight Biologics, Stealth BioTherapeutics, Santhera Pharmaceuticals, and Chiesi and received speaker honoraria from Chiesi and an unrestricted research grant from Stealth BioTherapeutics. Go to Neurology.org/NG for full disclosure.

Publication History
Received by Neurology: Genetics January 24, 2022. Accepted in final form April 13, 2022. Submitted and externally peer reviewed. The handling editor was Stefan M. Pulst, MD, Dr med.

Table 2 Clinical Features and Inheritance of Patients With KCND3 Mutations

| KCND3 variant | Clinical feature | Inheritance |
|---------------|-----------------|-------------|
| p.K214R       | Episodic gait disorder, vertigo, paraesthesia, pyramidal signs, abnormal ocular movement | AD with incomplete penetrance |
| p.F227 deletion | Slowly progressive cerebellar ataxia, onset from teenage to middle age; oculomotor abnormalities, pyramidal signs parkinsonism, epilepsy, or cognitive impairment have been reported in some cases | AD, recurrent mutation |
| p.R293_F295 duplication | Early-onset cerebellar ataxia, intellectual disability, oral apraxia, and epilepsy | De novo mutation |
| p.S301P       | Early onset forms with neurodevelopmental disorder, epilepsy, parkinsonism-dystonia, and ataxia in adulthood | Apparently de novo mutation |
| p.C317V       | Cerebellar ataxia onset at teenage, developmental delay, intellectual disability, myoclonus, and dystonia | De novo mutation |
| p.V338E       | Adult-onset cerebellar ataxia; cognitive dysfunction | AD |
| p.G345V       | Adult-onset cerebellar ataxia; variable pyramidal signs and oculomotor abnormalities | AD with incomplete penetrance |
| p.S347W       | Adult-onset slowly progressive cerebellar ataxia | Undetermined |
| p.T352P       | Mild cerebellar ataxia, cognitive impairment; variable degree of oculomotor disturbance, neuropathy, tremor, and myoclonus | AD |
| p.I362M       | Cerebellar ataxia | AD |
| p.M365T       | Cerebellar ataxia | AD |
| p.M373L       | Adult-onset pure cerebellar ataxia | AD |
| p.V374A       | Progressive cerebellar ataxia and bradyphrenia, cognitive impairment, paroxysmal ataxia exacerbations Cerebellar ataxia, dysarthria, and mild cognitive impairment | AD Apparently de novo mutation |
| p.P375S       | Teenage-onset or adult-onset cerebellar ataxia; cognitive dysfunction, dystonia, and bradykinesia | AD |
| p.T377M       | Adolescent-onset or adult-onset cerebellar ataxia; cognitive impairment in some patients | Recurrent mutation |
| p.G384S       | Cerebellar ataxia, intellectual disability, dystonia, and myoclonus | De novo mutation |
| p.S390N       | Teenage-onset or adult-onset cerebellar ataxia; cognitive dysfunction in some patients | Recurrent mutation |
| p.V392I       | Cerebellar ataxia, intellectual disability, epilepsy, early repolarization syndrome, and paroxysmal atrial fibrillation | Undetermined |
| p.R419H       | Slowly progressive cerebellar ataxia, parkinsonism, and cognitive dysfunction | Sporadic case |
| p.R431C       | Episodic ataxia | Sporadic case |
| p.L450F       | Late-onset cerebellar ataxia and pyramidal signs | AD |
| p.P633S       | Late-onset cerebellar ataxia, decreased reflexes, and vibration sense | Sporadic case |

Abbreviation: AD, autosomal dominant.
Adapted from Hsiao et al.3
Appendix Authors

| Name                  | Location                                                                 | Contribution                                                                 |
|-----------------------|---------------------------------------------------------------------------|------------------------------------------------------------------------------|
| Flavia Palombo, PhD   | IRCCS Istituto delle Scienze Neurologiche di Bologna, Programma di Neurogenetica, Italy | Analyzed NGS data, performed molecular studies, and drafted the article |
| Chiara La Morgia, MD, PhD | IRCCS Istituto delle Scienze Neurologiche di Bologna, UOC Clinica Neurologica, Italy | Performed clinical assessment and drafted the article |
| Claudio Fiorini, PhD  | IRCCS Istituto delle Scienze Neurologiche di Bologna, Programma di Neurogenetica, Italy | Performed wet phase of NGS |
| Leonardo Caporali, PhD | IRCCS Istituto delle Scienze Neurologiche di Bologna, Programma di Neurogenetica, Italy | Performed mitochondrial studies |
| Maria Lucia Valentino, MD | IRCCS Istituto delle Scienze Neurologiche di Bologna, UOC Clinica Neurologica, Italy, Department of Biomedical and NeuroMotor Sciences (DIBINEM), University of Bologna, Italy | Performed and analyzed muscle biopsy and critical revision of the article |
| Vincenzo Donadio, MD, PhD | IRCCS Istituto delle Scienze Neurologiche di Bologna, UOC Clinica Neurologica, Italy | Performed clinical assessment and critical revision of the article |

Appendix (continued)

| Name                  | Location                                                                 | Contribution                                                                 |
|-----------------------|---------------------------------------------------------------------------|------------------------------------------------------------------------------|
| Rocco Liguori, MD     | IRCCS Istituto delle Scienze Neurologiche di Bologna, UOC Clinica Neurologica, Italy, Department of Biomedical and NeuroMotor Sciences (DIBINEM), University of Bologna, Italy | Supervised the study and critical revision of the article |
| Valerio Carelli, MD, PhD | IRCCS Istituto delle Scienze Neurologiche di Bologna, Programma di Neurogenetica, Italy, Department of Biomedical and NeuroMotor Sciences (DIBINEM), University of Bologna, Italy | Study design, supervised the study, and critical revision of the article |

References
1. Lee YC, Durr A, Majczenko K, et al. Mutations in KCND3 cause spinocerebellar ataxia type 22. *Ann Neurol* 2012;72(6):859-869.
2. Duarri A, Jezierska J, Fokkens M, et al. Mutations in potassium channel KCND3 cause spinocerebellar ataxia type 19. *Ann Neurol* 2012;72(6):870-880.
3. Hsiao CT, Tropea TF, Fu SJ, et al. Rare gain-of-function KCND3 variant associated with cerebellar ataxia, parkinsonism, cognitive dysfunction, and brain iron accumulation. *Int J Mol Sci* 2021;22(15):8247.
4. Paucar M, Ågren R, Li T, et al. V374A KCND3 pathogenic variant associated with paroxysmal ataxia exacerbations. *Neurol Genet* 2021;7(1):e546. doi: 10.1212/NXG.000000000000546.