CLINICAL/SCIENTIFIC NOTES OPEN ACCESS

Missense mutations in DYT-TOR1A dystonia

Zafar Iqbal, PhD, Jeanette Koht, MD, PhD,* Lasse Pihlstrøm, MD, PhD, Sandra P. Henriksen, MSc, Chiara Cappelletti, MSc, Michael Bjørn Russel, MD, PhD, Osmar Norberto de Souza, PhD, Inger Marie Skogseid, MD, PhD,* and Mathias Toft, MD, PhD*

Correspondence
Dr. Iqbal
iqbal.z.phd@gmail.com

Neurol Genet 2019;5:e343. doi:10.1212/NXG.0000000000000343

DYT-TOR1A dystonia is caused by dominant mutations in the *TOR1A* gene, most frequently a heterozygous in-frame deletion in exon 5 (c.904_906delGAG; p.302/303delE).[1] The most frequent phenotype has childhood onset in a limb, spreading to generalized dystonia within a few years. However, also mild focal forms and onset in the cervical and even cranial region have been described. Age at onset varies from 3 to 64 years,[2] and penetrance is only 30%. Other in-frame deletions and point mutations in *TOR1A* have been associated with dystonia in a limited number of patients.[3,4] Here, we report 2 new patients with missense mutations in *TOR1A*.

Patient 1

Patient 1 (IV-1; figure, A) was born from nonconsanguineous Caucasian parents. Focal dystonia started at age 40 years with painful dystonic writer's cramp affecting the right wrist and finger flexors. She was treated with botulinum toxin injections, but after 10 years, she discontinued treatment because of very mild symptoms. At age 60 years, only increased blink rate was noted. Two of her children were identified with hyperkinetic movements from adolescence (figure, A). A daughter (V-2; figure, A) had an increased blink rate and a mild head tremor. Her son (V-3; figure, A) was treated with botulinum toxin for a dystonic head tremor and mild torticollis. Gene panel sequencing was performed as described previously,[5] revealing a likely pathogenic mutation, c.934A>G; p.R312G in *TOR1A* (figure, B), which is conserved down to zebrafish (figure, C), has a Combined Annotation Dependent Depletion (cadd.gs.washington.edu) score of 19.5, and is found only once in 246266 alleles (0.0004061%) in the gnomAD database (gnomad.broadinstitute.org). Sanger sequencing revealed segregation of the mutation with the phenotype (figure, A). Using homology modeling, a possible deleterious effect of p.R312G was assessed.[6] The basic torsinA structure should be unaffected by the variant. In the wild type, however, the highly flexible arginine allows R312 to come as close as 2.7 Å to one of the adenosine triphosphate ribose hydroxyls and make hydrogen bonds. In the R312G mutant, this interaction is lost, thus possibly causing protein malfunctioning (figure, D).

Patient 2

Patient 2 (II-1; figure, E) was born from nonconsanguineous Caucasian, healthy parents. Pre- and postnatal periods and psychomotor development were normal. Dystonia started cervically at age 15 years, with phasic torti- and retrocollis, which within 2 years generalized to involve the trunk and upper extremities. Cervical botulinum toxin injections provided some relief until age 23 years. Oral trihexyphenidyl, and levodopa and clonazepam added 2 years later, improved his severe axial dystonia only slightly. At age 26 years, brain and spine MRI and neuropsychological testing were normal. Testing for the classic 3-basepair deletion in *TOR1A* was negative. He had no impairment of voice, speech, or swallowing, but pathologic face grimacing and moderate

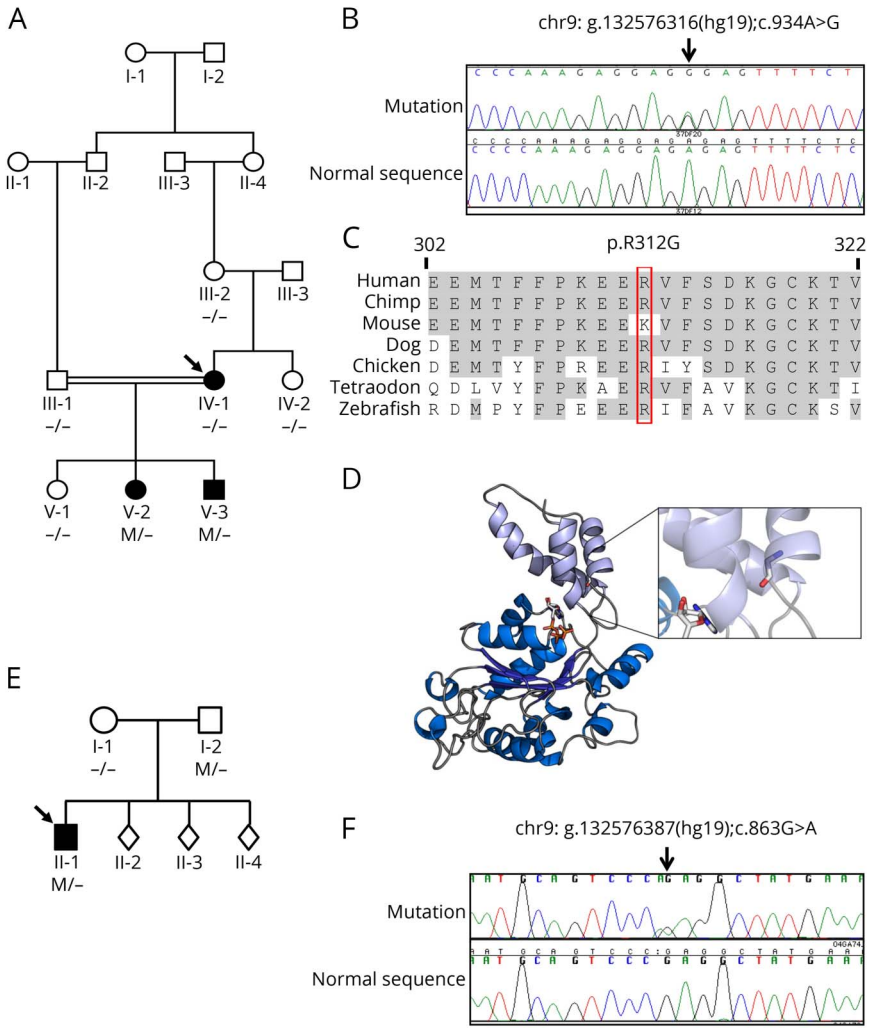
*These authors contributed equally to this study as senior author.

From the Department of Neurology (Z.I., L.P., S.P.H., C.C,. I.M.S,. M.T), Oslo University Hospital; Institute of Clinical Medicine (J.K., M.T), University of Oslo; Department of Neurology (J.K), Drammen Hospital, Vestre Viken Hospital Trust; Head and Neck Research Group, Research Center (M.B.R), Akershus University Hospital; Campus Akershus University Hospital (M.B.R), University of Oslo, Norway; and Faculty of Informatics, Laboratory for Bioinformatics, Modelling & Simulation of Biosystems (O.N.D.S), Pontifical Catholic University of Rio Grande do Sul (PUCRS), Porto Alegre, RS, Brazil.

Go to Neurology.org/NG for full disclosures. Funding information is provided at the end of the article.

The Article Processing Charge was funded by the authors.

Figure Pedigrees of patient 1 and 2, partial Sanger chromatograms, amino acid conservation, and homology modeling



(A) Pedigree structure of the family of patient 1 (IV-1) with the c.934A>G (M) mutation in *TOR1A,* demonstrating segregation of the mutation. The proband is indicated by the sign of arrow. (B) Partial chromatogram indicating the c.934A>G mutation in *TOR1A* in patient 1. (C) Evolutionary conservation of amino acid at position 312, revealing high conservation down to zebrafish. (D) Homology model of the human torsinA p.R312G mutant. The α-helices and β-sheet of the AAA+ domain are colored in blue and purple, respectively. The C-terminal domain is colored in light blue. The ATP molecule and G312 are represented as sticks and colored by CPK. The mutation region is highlighted at the top right of this figure. Image generated with PyMOL (pymol.org/2/). (E) Pedigree structure of the family of patient 2 (II-1) with the c.863G>A (M) mutation in *TOR1A*. The proband is indicated by the sign of arrow. Three siblings are indicated by diamond (undisclosed sex) symbols. The siblings have not been tested for the mutation. (F) Partial chromatogram indicating the c.863G>A mutation in *TOR1A* in patient 2. ATP = adenosine triphosphate.

distal extremity dystonia. Since age 27 years, his generalized dystonia has been treated successfully with deep brain stimulation (DBS) of the internal globus pallidus bilaterally, with a marked and sustained effect, allowing medication to be tapered off. At age 42 years, he has residual mild retrocollis and thoracic kyphoscoliotic posture. Gene panel sequencing revealed a previously reported missense mutation, c.863G>A; p.R288Q in *TOR1A,*[7] with paternal inheritance and reduced penetrance, the latter also witnessed by Zirn et al.[7] (figure, E and F).

Herein, we report a novel *TOR1A* missense mutation, p.R312G, which segregated with mild isolated segmental dystonia in a small family. Multiple lines of bioinformatic predictions indicate possible deleterious effects on protein function. However, functional analyses and identification of genetic recurrence are warranted to confirm its pathogenicity.

Of the published missense mutations in *TOR1A* (mdsgene.org), genetic recurrence has so far only been reported for p.T321M. We report a patient carrying p.R288Q, with adolescent-onset, isolated generalized dystonia and marked axial and little cranial involvement. This mutation was previously reported in a patient with very early childhood-onset lower limb dystonia and severe generalization, who at age 18 years had dysphagia, dysarthria, joint contractures, pyramidal signs, and cerebellar atrophy.[7] Treatment with DBS was not mentioned. Additional experiments strongly indicated pathogenicity of the p.R288Q variant,[7] which is further confirmed by our present finding.

Most reported patients with *TOR1A* missense mutations presented with adult onset, including the p.R312G index patient in our study. This may indicate that missense mutations have a less profound effect on torsinA function than the common deletion. However, the phenotypic spectrum of *TOR1A* mutations is very broad, ranging from nonpenetrance to isolated focal, segmental, or generalized dystonia in carriers of different types of mutations, which is highlighted in our and previous reports.[3,4,7] The causes of this large phenotypic

variation in *TOR1A* mutation carriers still largely remain elusive.

Author contributions

Z. Iqbal: bioinformatic analysis and interpretation of data, wet laboratory work, and drafting and revision of the manuscript. J. Koht: ascertaining the patients and clinical data, study concept and design, and drafting and revision of the manuscript. S.P. Henriksen: preparation of the samples for sequencing and arrangement of the samples. C. Cappelletti: wet laboratory work. M.B. Russell: ascertaining the patients. O. Norberto de Souza: homology modeling and drafting the manuscript. L. Pihlstrøm: bioinformatic analysis, interpretation of data, and revision of the manuscript. I.M. Skogseid: ascertaining the patients and clinical data, study concept and design, and drafting and revision of the manuscript. M. Toft: study concept and design, obtained funding, study supervision, and revision of the manuscript.

Study funding

The study was funded by an internal grant from Oslo University Hospital. Dr. Skogseid was supported by a research grant from The Norwegian Research Council (grant number 229654).

Disclosure

Disclosures available: Neurology.org/NG.

Publication history

Received by *Neurology: Genetics* November 28, 2018. Accepted in final form May 9, 2019.

References

1. Ozelius LJ, Hewett JW, Page CE, et al. The early-onset torsion dystonia gene (DYT1) encodes an ATP-binding protein. Nat Genet 1997;17:40–48.
2. Ozelius LJ, Bressman SB. Genetic and clinical features of primary torsion dystonia. Neurobiol Dis 2011;42:127–135.
3. Calakos N, Patel VD, Gottron M, et al. Functional evidence implicating a novel TOR1A mutation in idiopathic, late-onset focal dystonia. J Med Genet 2010;47:646–650.
4. Leung JC, Klein C, Friedman J, et al. Novel mutation in the TOR1A (DYT1) gene in atypical early onset dystonia and polymorphisms in dystonia and early onset parkinsonism. Neurogenetics 2001;3:133–143.
5. Iqbal Z, Rydning SL, Wedding IM, et al. Targeted high throughput sequencing in hereditary ataxia and spastic paraplegia. PLoS One 2017;12:e0174667.
6. Hettich J, Ryan SD, de Souza ON, et al. Biochemical and cellular analysis of human variants of the DYT1 dystonia protein, TorsinA/TOR1A. Hum Mutat 2014;35:1101–1113.
7. Zirn B, Grundmann K, Huppke P, et al. Novel TOR1A mutation p.Arg288Gln in early-onset dystonia (DYT1). J Neurol Neurosurg Psychiatry 2008;79:1327–1330.