BRIEF COMMUNICATION

DEPDC5 mutations are not a frequent cause of familial temporal lobe epilepsy

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Epilepsia, 56(10):e168–e171, 2015
doi: 10.1111/epi.13094

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Summary

Mutations in the *DEPDC5* (DEP domain–containing protein 5) gene are a major cause of familial focal epilepsy with variable foci (FFEVF) and are predicted to account for 12–37% of families with inherited focal epilepsies. To assess the clinical impact of *DEPDC5* mutations in familial temporal lobe epilepsy, we screened a collection of Italian families with either autosomal dominant lateral temporal epilepsy (ADLTE) or familial mesial temporal lobe epilepsy (FMTLE). The probands of 28 families classified as ADLTE and 17 families as FMTLE were screened for *DEPDC5* mutations by whole exome or targeted massive parallel sequencing. Putative mutations were validated by Sanger sequencing. We identified a *DEPDC5* nonsense mutation (c.918C>G; p.Tyr306*) in a family with two affected members, clinically classified as FMTLE. The proband had temporal lobe seizures with prominent psychic symptoms (déjà vu, derealization, and forced thoughts); her mother had temporal lobe seizures, mainly featuring visceral epigastric auras and anxiety. In total, we found a single *DEPDC5* mutation in one of (2.2%) 45 families with genetic temporal lobe epilepsy, a proportion much lower than that reported in other inherited focal epilepsies.

KEY WORDS: Temporal lobe seizures, Genetics, Mutation, Familial focal epilepsy with variable foci.

Familial temporal lobe epilepsy (FTLE) comprises two genetic epilepsy syndromes: autosomal dominant lateral temporal epilepsy (ADLTE; MIM 600512) and familial mesial temporal lobe epilepsy (FMTLE). ADLTE is characterized by focal seizures with auditory or aphasic auras, no brain structural abnormalities, and usually good outcome.[1] Mutations in the leucine-rich glioma-inactivated 1 (*LGI1*) gene are found in about 30% of the ADLTE families.[1] FMTLE, first described by Berkovic et al.,[2] is characterized by auras with prominent psychic and autonomic features, suggesting a mesial temporal origin. Usually, it is not associated with hippocampal sclerosis (HS) or febrile seizures (FS), and has a good prognosis. In addition to the benign condition, a more severe FMTLE form with prolonged FS in infancy and/or hippocampal sclerosis has been described.[3] The genetics of this clinically heterogeneous syndrome remains largely elusive. Two loci on chromosomes 4q13.2-q21.3 and 3q26 have been linked to benign FMTLE,[4–6] whereas other loci on chromosomes 18qter, 1q25-31, and 12q22-23.3 have been linked to FMTLE associated with FS.[7,8]

Recently, mutations in the disheveled, Egl-10, and pleckstrin domain–containing protein 5 (*DEPDC5*) gene have been identified as a major cause of familial focal epilepsy

Accepted June 30, 2015; Early View publication July 27, 2015.

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with variable foci (FFEVF).[9,10] *DEPDC5* mutations have also been found in other inherited focal epilepsies, including autosomal dominant nocturnal frontal lobe epilepsy (ADNFLE) and FTLE.[9,10] The frequency of *DEPDC5* mutations in families with focal epilepsies has been estimated between 12%[9] and 37%.[10] To determine whether *DEPDC5* is a frequent cause of genetic temporal lobe epilepsies, we screened a collection of Italian ADLTE and FMTLE families.

Methods

Study families

The index cases of 45 Italian families with ADLTE (n = 28) or FMTLE (n = 17) were screened for mutations in *DEPDC5*. All participants signed informed consent approved by the local ethics committee. Clinical data of these families, most of which have been described previously,[1,3] are summarized in Table S1. ADLTE families were defined as having two or more affected members with a history of focal seizures characterized by auditory and/or aphasic symptoms, absence of structural or metabolic insults to the central nervous system, and relatively benign evolution.[1] ADLTE families with *LGI1* mutations were excluded. FMTLE families were defined as having two or more affected members with focal seizures characterized by psychic or autonomic symptoms and absence of brain structural abnormalities.[2,3] The clinical phenotype of FMTLE families was generally benign, as that described by Berkovic et al.[2] 1.5 Tesla brain magnetic resonance imaging (MRI) was unremarkable in the affected members of both ADTLE and FMTLE families, except for hippocampal sclerosis, which was observed in three affected members of one FMTLE family (MAT), and in two patients from other FMTLE families (see *Striano et al.* [3]).

Exome and *DEPDC5* exon sequencing

Exome sequencing was performed in the probands of 17 ADLTE families using the SureSelect 50 Mb v. 2.0 capture kit (Agilent Technologies). Enriched libraries were sequenced on Illumina HiSeq2000. Reads were aligned on the hg19 reference sequence with the Burrows-Wheeler Aligner software, and variant calling and genotyping was performed with Genome Analysis Toolkit. An average of 4.545 billion bases of sequence were generated per affected individual, and 80% of the total bases were mapped to the targeted exons. Massive parallel resequencing of *DEPDC5* exons was performed in the probands of the remaining families. Primers required for amplification of targeted regions were designed with online DesignStudio (Illumina, San Diego, CA, U.S.A.). TruSeq Custom Amplicon libraries were prepared and sequenced in pool on Illumina MiSeq System using the MiSeq Reagent Kit v3 (Illumina), at a 2 × 250 bp read length configuration with dual indexing. The total yield per sample was on average 0.23 Gb (min 0.08, max 0.6 Gb). The minimum coverage of individual DEPDC5 exons was 20× in exome sequencing and 140× in massive parallel sequencing analyses. Four small exons (7, 11, 19, and 20, altogether spanning a small portion [278 bp; 5%] of the *DEPDC5* coding region) were poorly or not covered by the latter sequencing method, likely due to inappropriate primer design. *DEPDC5* variants (heterozygous nonsynonymous, splice site, and small insertion-deletion variants) identified by either method that occurred in 1,000 Genomes Project (http://www.1000genomes.org/) and Exome Variant Server (EVS; http://evs.gs.washington.edu/EVS/) databases with frequencies >1% and those classified as benign/tolerated/not damaging by different prediction software tools such as SIFT (http://sift.jcvi.org/) and Polyphen-2 (http://genetics.bwh.harvard.edu/pph2/) were filtered out. Putative mutations were validated by Sanger sequencing with the Big Dye Terminator Cycle sequencing kit (ABI PRISM; Applied Biosystems-Life Technologies, Carlsbad, Germany).

Results

Genetic findings

We screened the index cases of 45 families with FTLE (Table S1) for *DEPDC5* mutations and found a single nonsense mutation (c.918C>G; p.Tyr306*) in the pedigree shown in Figure 1. The mutation, validated by Sanger sequencing, was found in the proband (individual III:1) and in her mother (individual II:2), both affected with mesial temporal lobe epilepsy, but not in 110 geographically matched controls, or in dbSNP138 (http://www.ncbi.nlm.nih.gov/projects/SNP/), 1,000 genomes, ExAc (http://exac.broadinstitute.org/), and EVS databases. This mutation introduces a stop codon, likely resulting in degradation of the mutated transcript by the nonsense-mediated messenger RNA (mRNA) decay system or protein truncation. In either case, *DEPDC5* haploinsufficiency is likely to cause the disease, as reported.[10]

Family description

The proband (patient III:1) of the *DEPDC5*-mutated family (Fig. 1) is a 29-year-old woman having a single, brief (<5 min) febrile seizure at age 2 years and experiencing rare, dyscognitive seizures followed by right arm tonic posturing and joyfulness attitude, started at age 8 years. The patient remained seizure-free on phenobarbital for 3 years. However, at age 11 years, she presented monthly tonic–clonic seizures during sleep and daily episodes of déjà-vu, derealization, and forced thoughts, sometimes with secondary generalization. 1.5 Tesla brain MRI was normal and interictal electroencephalography (EEG) studies revealed only rare, isolated, bitemporal sharp waves. Treatment with valproate and topiramate was poorly effective, whereas she responded to lamotrigine. She has been seizure-free since age 19 years. Her mother (patient II:2) had rare dyscognitive seizures from

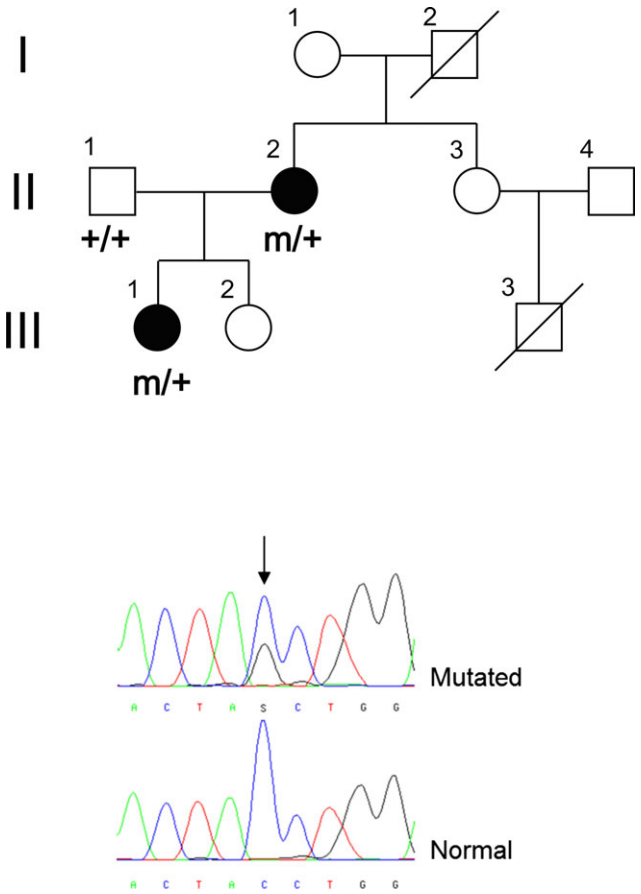

Figure 1.
Family pedigree and mutation in *DEPDC5*. (Top) Pedigree of the *DEPDC5*-mutated family. Circles denote females; squares denote males. Blackened symbols denote subjects with mesial temporal lobe epilepsy. Individuals carrying one mutant and one normal allele are denoted by m/+, whereas the one with no mutations by +/+. (Bottom) Sanger sequence tracing detecting the disease allele. Arrow indicates the variant allele.

age 12 years up to her first pregnancy (31 years). Her seizures were characterized by visceral epigastric auras and anxiety; sometimes automatic walking behaviors also occurred. Brain MRI was normal and interictal EEG studies showed rare right temporal sharp waves. She was seizure-free on carbamazepine (400 mg per day).

Discussion

DEPDC5 mutations have been suggested to be a common cause of genetic focal epilepsy, accounting for 12–37% of all familial focal epilepsy syndromes.[9,10] However, the impact of *DEPDC5* on FTLE has not been determined. We found one *DEPDC5* pathogenic mutation in 17 FMTLE families analyzed, whereas no mutations were detected in 28 ADLTE families. In our family collection, the overall frequency of *DEPDC5* mutations in FTLE is estimated at one (2.2%) of 45, a frequency much lower than that predicted for other genetic focal epilepsies.[9,10] Although mainly linked to nonlesional familial focal epilepsies, *DEPDC5* mutations have recently been associated with focal cortical dysplasia and other brain malformation–associated focal epileptic syndromes, and the proportion of *DEPDC5* mutation–associated cortical malformations is thought to be underestimated (for review, see Poduri[11]). The absence of brain structural alterations in our ADLTE and benign FMTLE familial cases might, at least in part, account for the low frequency of *DEPDC5* mutations in our family cohort. Thus, our data suggest that *DEPDC5* may have a low impact on familial temporal lobe–related epilepsies and, particularly, that *DEPDC5* mutations may not be involved in ADLTE.

The *DEPDC5*-mutated nuclear family described here was diagnosed as FMTLE based on the loose definition: "at least two affected members with mesial temporal lobe epilepsy." It cannot be excluded, however, that other unknown relatives may have developed other types of focal epilepsy (e.g., frontal or parietal), which would change the familial diagnosis to FFEVF. Therefore, identification of *DEPDC5* mutations in small families initially classified as FMTLE should be regarded as a diagnostic criterion for possible FFEVF. In general, the group of families classified as FTLE on the basis of clinical criteria may in fact comprise a proportion of FFEVF families, which could be disclosed by searching for mutations in *DEPDC5*. Based on our results, the proportion of FFEVF families misdiagnosed as FTLE should not be high.

Acknowledgments

Supported by Telethon-Italy (grant no. GGP12078 to C.N.) and the Genetics Commission of the Italian League Against Epilepsy (to R.M. and C.N.). We thank the following colleagues who contributed part of the study patients: U. Aguglia, E. Ferlazzo (Magna Graecia University of Catanzaro, Catanzaro, Italy); P. Banfi (Circolo Hospital, Varese, Italy); S. Binelli (C. Besta Foundation Neurological Institute, Milano, Italy); F. Bisulli, P. Tinuper (IRRCS-Institute of Neurological Sciences, Bologna, Italy); C. Boniver (University of Padua, Padova, Italy); G. Coppola (Faculty of Medicine and Surgery, University of Salerno, Salerno, Italy); A.T. Giallonardo, C. Di Bonaventura, O. Mecarelli, P. Pulitano (Sapienza University of Rome, Roma, Italy); M. Elia (OASI Institute for Research on Mental Retardation and Brain Aging, Troina [Enna], Italy); A. La Neve (University of Bari, Bari, Italy); A. Magaudda (University of Messina, Messina, Italy); M. Marchini (C. Poma Hospital, Mantova, Italy); V. Sofia (University of Catania, Catania, Italy).

Disclosure

None of the authors has any conflict of interest to disclose. We confirm that we have read the Journal's position on issues involved in ethical publication and affirm that this report is consistent with those guidelines.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Table S1. Clinical data of FTLE families.