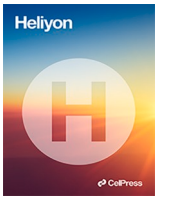
Heliyon 10 (2024) e32693

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon



Research article

Clinical features and genetic analysis of developmental and epileptic encephalopathy caused by biallelic variants of *CACNA1B*

Xin-you Yu [a,1], Qing-mei Sun [b,1], Rui-ping Lu [a], Bo Wei [a], Xiao-yan Wang [a], Li-hua Pan [a,*]

[a] *Gerneral Hospital of Ningxia Medical University, Yin'chuan, Ningxia, 750004, China*
[b] *Gansu Province Maternal and Child Health Care Hospital, Lan'zhou, Gansu, 730000, China*

ARTICLE INFO

Keywords:
CACNA1B gene
Epilepsy
Developmental and epileptic encephalopathy
Neurodevelopmental disorders
Whole exome sequencing

ABSTRACT

Objective: To analyze the clinical features and genetic etiology of a patient with developmental and epileptic encephalopathy.
Methods: The clinical information and peripheral blood of the patient and their family members were collected before the whole exome sequencing analysis was performed and Sanger sequencing was employed to verify the potential variant.
Results: The patient presented with epilepsy and cerebral palsy with his parents, brother, and sister being all healthy. Whole exome sequencing analysis revealed that the child carried the paternal c.823del (p. R275Gfs*31) heterozygous variant and the maternal c.2456del (p. V819Gfs*190) heterozygous variant of the *CACNA1B* gene. Pedigree verification found that the elder brother and amniotic fluid of fetus in womb carried the paternal c.823del heterozygous variant, and the elder sister carried the maternal c.2456del heterozygous variant, which conformed to the law of autosomal recessive inheritance. Neither of these two variants has been reported in the literature and has not been included in the Genomic Mutation Frequency Database (gnomAD); according to the American Academy of Medical Genetics and Genomics Variation Grading Guidelines (ACMG), both variants are classified as pathogenic variants (PVS1+PM2-Supporting + PM3).
Conclusion: This study reported the first case of a child with neurodevelopmental disorder and epilepsy caused by a new compound heterozygous variant of the *CACNA1B* gene in China, clarified its genetic etiology, enriched the mutation spectrum and disease spectrum of *CACNA1B* gene, and provided a basis for prenatal diagnosis of the family.

1. Introduction

Developmental and epileptic encephalopathy (DEEs) are a group of severe early-onset epilepsy characterized by intractable seizures, developmental delays, or developmental regression associated with persistent epileptic activity, usually with early childhood onset and a poor prognosis that places a heavy burden on families and society. According to statistics, the prevalence of epilepsy in children under 2 years of age is nearly 70/100,000, and hereditary epilepsy accounts for more than 0.4 % of the general population and

* Corresponding author.
E-mail address: Panlihuanxmu@163.com (L.-h. Pan).
[1] Xin-you Yu and Qing-mei Sun contributed equally to this work as Co-first author.

https://doi.org/10.1016/j.heliyon.2024.e32693
Received 29 August 2023; Received in revised form 26 May 2024; Accepted 6 June 2024
Available online 7 June 2024

30 % of all epilepsy patients [1]. Rapid advances in genetic testing technology have elucidated the genetic etiology of many DEEs and exhibited high phenotypic and genetic heterogeneity [2,3]. More than 150 single genes have been reported to be associated with DEEs, despite analysis using comprehensive genetic tests, 50 % of patients remain genetically undiagnosed [4,5].

In recent years, with increased disease awareness and the widespread clinical use of genetic testing technologies, genes and diseases related to DEEs have been continuously discovered, many of which are components of the gene-encoded neuronal ion channels that cause epilepsy and play a central role in the occurrence and remission [6,7]. Among these genes associated with DEEs, the most common genetic variants are sequence alterations (30–40 %) and chromosomal deletions or duplications (5–10 %) [8]. *CACNA1B* (OMIM:601012), encoding an N-type voltage-gated calcium channel (Cav2.2), which are essential for neurotransmitter release from cerebral neurons [9–11]. Common variants in CACNA1B were the newly reported gene associated with DEE with only 6 patients being reported worldwide, from Pakistani and European ancestry, and all carrying loss-of-function type variants of the *CACNA1B* double allele, including two frameshift variants p.Leu1222Argfs*29 and p.Gly1192Cysfs*5, a nonsense mutation p.Arg383* and a shear variant c.4857þ1G > C. No related cases and variant loci have been reported in the Chinese population.

This study described a consanguineous family from northwest China, with a pre-documented individual presenting with neurodevelopmental disorder, epilepsy, and cerebral palsy. The purpose of this study was to identify the genetic etiology through whole-exome sequencing analysis of the family, to provide a molecular basis for clinical diagnosis and precise treatment of the affected child, and to provide timely genetic counseling for prenatal diagnosis of the next child in the family line to prevent birth defects.

2. Methods

2.1. Patient and sample collection

The present work was based on a 5 years old male children patient who has epileptic encephalopathy, cerebral palsy, severe neurodevelopmental delay (often accompanied by degeneration) and hyperkinetic movement disorder. His sleep electroencephalogram was showed in (Fig. 1). His family members were including parents, older brother, and older sister who were all healthy, and the mother was pregnant with her 4th child. 3 mL of peripheral blood was collected from the patient and his family members. The mother of the child underwent amniocentesis at 20 weeks of gestation to extract fetal DNA for preservation. Berry Genomics was commissioned to conduct a whole-exome sequencing analysis of the family. This study was approved by the Ethics Committee of the General Hospital of Ningxia Medical University with the informed consent of the child's guardian (2020-664).

2.2. Whole exome sequencing

Genomic DNA was extractd from 200 μL of peripheral blood, 1 μg was of which was taken to interrupt and construct a whole genome library using PCR-free technology with a whole exon capture kit (NanoWES Human Exome, Berry Genomics). After capture, sequencing was performed on an Illumina NovaSeq 6000 high-throughput sequencer with an average sequencing depth of 80X and aligned with the human reference genome (hg38/GRCh38). The data were analyzed using the Verita Trekker® Variant Site Detection System and Enliven® Variant Site Annotation Interpretation System to remove 1000G, ExAC, gnomAD, dbSNP and variant sites with mutation frequencies greater than 0.5 % in the internal database, remove non-functional variant sites (synonymous mutations, non-coding region mutations, etc.), and retain potentially Synonymous mutations that may affect shearing and known variants included in

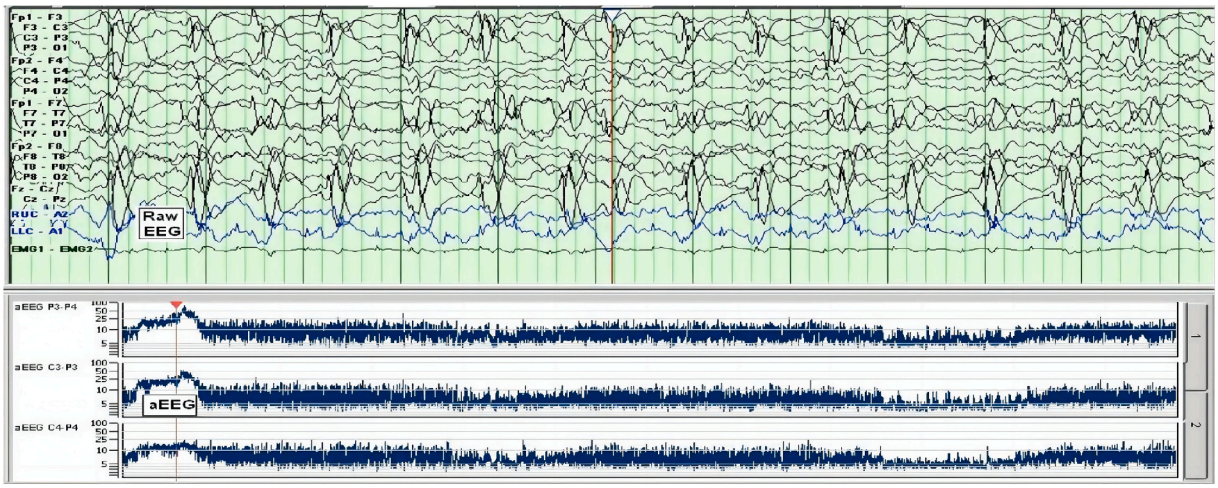


Fig. 1. Sleep EEG analysis: persistent, diffuse moderate-to-high amplitude 2–3 Hz delta waves and 4–7 Hz theta waves in each lead on both sides, with obvious NREM-REM periodic changes. The former is greater than the latter in time, and Symmetrical apex waves can be seen in the anterior head, slightly lower to medium amplitude 12–14 Hz spindle wave, and a small amount of low to medium amplitude 14–28 Hz beta wave rhythm, bilateral amplitude and frequency symmetry.

HGMD and ClinVar databases; SIFT, Polyphen2, CADD and other software were used to predict the pathogenicity of variant loci.

2.3. Pathogenicity rating of variant loci

The pathogenicity rating of variant loci was in accordance with the American College of Medical Genetics and Genomics (ACMG) guidelines and the ClinGen Sequence Variation Interpretation Panel of Experts on Recommendations for the Application of Guidelines Standards and Changes in Ratings. The ACMG ratings are categorized into five levels: "Pathogenic", "Possibly Pathogenic", "Of Uncertain Significance", "Possibly Benign" and "benign". Criteria for pathogenic variants can be classified as very strong (PVS1), strong (PS1～4); moderate (PM1～6), or supporting (PP1～5). Criteria for benign variants can be classified as stand-alone (BA1), strong (BS1～4), or supporting (BP1～6).

2.4. Sanger sequencing

Sanger sequencing was used for lineage verification of the suspected variant loci, and the following two pairs of primer sequences were used for the two variant loci: *CACNA1B*–1F: CGCCATTACTCCATTGCTG; *CACNA1B*-1R: CCTCTGCTCGGTCCTGGT. *CACNA1B*–2F: CTCGGGGTCAGCAAGAGG; *CACNA1B*-2R: GCCAAGACGCCACTCACAT. Gradient PCR was used, and the reaction conditions were: 95 °C for 5 min; 95 °C for 30 s, 62 °C for 30 s, 72 °C for 1 min, a total of 14 cycles; 95 °C for 30 s, 55 °C for 30 s, 72 °C for 1 min, 25 cycles; 72 °C extension for 5 min. PCR amplification products were detected by 1 % agarose gel electrophoresis and then sequenced on an ABI 3500DX sequencer.

3. Results

3.1. Whole exome sequencing analysis

The patient was found to carry two compound heterozygous variants of the *CACNA1B* gene, c.823del (p.R275Gfs*31) in the paternal source and c.2456del (p.V819Gfs*190) in the maternal source, respectively, as detected by whole exome sequencing (Trio-

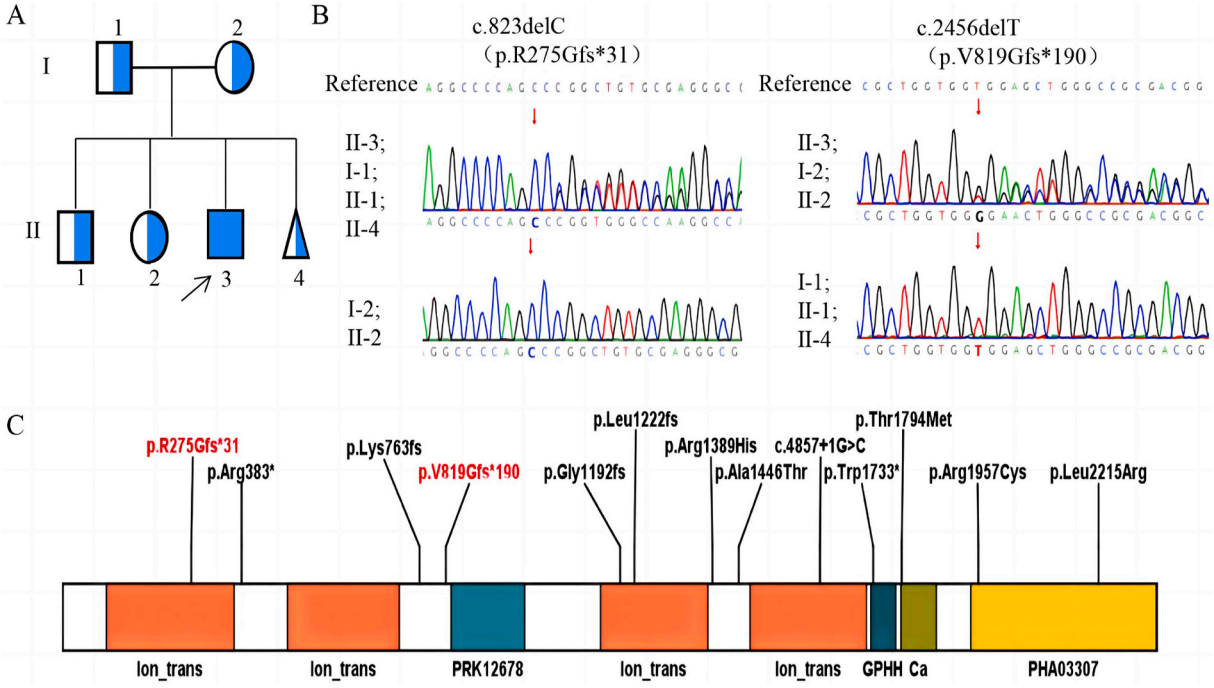


Fig. 2. Whole exome sequencing analysis. A: Pedigrees and genetic analysis in the present family. The pedigree chart typically uses squares to represent male family members and circles to represent female family members. The black-filled symbols indicate individuals who are affected by the condition, while the half-filled symbols show carriers of the mutation. B: The Sanger sequencing of c.823delC(p.R275Gfs*31) and c.2456delT(p.V819Gfs*190) variant of *CACNA1B* in this family. The electropherograms are the graphical outputs from an automated DNA sequencing machine, where each peak represents one of the four nucleobases (adenine, thymine, cytosine, guanine) that make up the DNA sequence. The colors in the electropherogram correspond to the nucleotide bases: adenine (green), thymine (red), cytosine (blue), and guanine (black). C: A schematic diagram of the distribution of *CACNA1B* mutant loci in different domains. The different colored boxes represent various functional domains of the protein encoded by the CACNA1B gene. The red fonts indicate reported variant in the present study.

WES) of the family. Sanger sequencing verification results were consistent with Trio-WES. Both the older brother and the amniotic fluid cells carried the c.823del (p.R275Gfs*31) heterozygous variant, and the older sister carried the c.2456del (p.V819Gfs*190) heterozygous variant of maternal origin (Fig. 2A–C), which was in line with the law of autosomal recessive inheritance.

3.2. Pathogenicity analysis of variant loci

CACNA1B gene c.823del and c.2456 del variants have not been reported in the literature and have not been included in the human exon database (ExAC), reference population thousand genomes (1000 G), and population genome mutation frequency database (gnomAD); Both variants caused changes in the gene's open reading frame, resulting in altered protein function; both variants constitute compound heterozygosity in this family; both variants were classified as pathogenic according to ACMG guidelines (PVS1+PM2+ PM3).

4. Discussion

This study reported the first patient with developmental and epileptic encephalopathy caused by biallelic loss-of-function variants in *CACNA1B* in the Chinese population, which provides a strong molecular basis for the clinical diagnosis of patients and prenatal diagnosis of the next child in the family. The findings also contribute to strengthening our in-depth understanding of the molecular genetics of developmental epileptic encephalopathy and laying the foundation for the study of the molecular mechanism of the disease and the study of genotype-phenotype correlation.

CACNA1B gene is the latest reported gene reported by Gorman et al., which associated with DEEs with a biallelic loss-of-function mutation linked to progressive epileptic dyskinesia [4]. They described 6 cases from 3 unrelated families with similar neurodevelopmental disorders. 3 of the 6 patients had normal early development and were able to sit and talk before the age of 1 year, while the other child had a slightly delayed development, and the remaining two patients never sat or spoke independently. All patients developed refractory focal, generalized, or myoclonic seizures, consistent with epileptic encephalopathy between 9 and 30 months of age, after which all showed developmental decline. Electroencephalography revealed cardiac arrhythmias in some patients. All patients also presented with non-epileptic hyperkinetic movements, including myoclonic dystonia, dyskinesia, oral dyskinesia, and choreoathetosis. Other features included axial hypotonia, peripheral hypertonia with hyperactive reflexes, postnatal microcephaly, tube feeding, strabismus, nystagmus, poor visual annotation or cortical visual disturbances, and recurrent respiratory infections. A minority of patients had mild, nonspecific deformity features such as anteversion of the nostrils, pointed feet, hip dislocation, and/or small testes. Brain imaging showed cerebral atrophy in three patients, normal in one, and no imaging in one patient. The clinical manifestations of the patients in this study family were similar to those reported in the literature, including postnatal microcephaly, seizures, hypotonia, developmental regression, small testis.

Voltage-gated calcium channels (VGCCs) are the main mediators of depolarization-induced calcium entry into neurons, which can initiate action potentials, neurotransmission, excitation-contraction coupling, and other physiological processes [12,13], thereby causing various related channelopathies. Currently, 10 VGCC family members have been identified in mammals and can be divided into two categories: high-voltage-activated (HVA) channels and low-voltage-activated (LVA) channels. HVA channels are heterogeneous protein complexes formed by the co-assembly of the pore-forming Cavα1 subunit plus auxiliary Cavb and Cava2d subunits, whereas LVA channels lack these auxiliary subunits [14]. The Cavα1 subunit is a key determinant of calcium channel subtypes and contains three major subunits (called Cav1, Cav2, and Cav3), each of which is composed of several members [15,16]. Presynaptic neuron Cav2 channel family, including 3 isoforms of Cav2.1, Cav2.2, and Cav2.3 (respectively called P/Q-type, n-type and r-type calcium channels), is encoded by the *CACNA1A*, *CACNA1B*, and *CACNA1E*. In the Lennox-Gastaut syndrome spectrum, both gain-of-function and loss-of-function de novo variants in the *CACNA1A* gene lead to severe developmental epileptic encephalopathy [17]. De novo pathogenic variants in *CACNA1E* cause developmental and epileptic encephalopathy with spasticity, macrocephaly, and dyskinesia [18]. For the *CACNA1B* gene, a 2015 study reported that its missense mutation was associated with the myoclonus-dystonia syndrome [19], but this conclusion was not consistently replicated in subsequent studies [20]. In addition, *CACNA1B* plays an important role in spinal pain signaling and has emerged as an important drug target for chronic pain treatment [21]. Until recently, Gorman et al. found that biallelic loss-of-function variants in *CACNA1B* were associated with neurodevelopmental disorders of epilepsy and non-epileptic hyperkinetic movements [4].

CACNA1B is widely expressed throughout the central nervous system, including the cerebral cortex and white matter, hippocampus, basal ganglia, and cerebellum [22]. It is mainly located at presynaptic terminals and mediates Ca2+ influx, triggering the rapid synaptic release of neurotransmitters [4]. *CACNA1B* is an evolutionarily conserved transmembrane protein with a full length of 2339 residues (Uniprot Q00975) consisting of four homologous repeats (DI-DIV), each repeat containing five hydrophobic transmembrane α-helices (S1, S2, S3, S5, S6), a positively charged transmembrane α-helix (S4), and a P-loop between S5 and S6, where the S1 to S4 segments form a voltage-sensitive region (VSD) and the four VSDs (VSDI-VSDIV) surround the central pore formed by the S5 and S6 helices of the four regions [23]. In addition, the N- and C-termini and intracellular linkers between domains I and II and II and III play important roles in channel expression and gating and protein interactions [24,25]. The *CACNA1B* gene p.R275Gfs*31 variant carried by this patient was located in the DI domain and the p.V819Gfs*190 variant in the junction region of DI and DIII. Both variants are predicted to trigger nonsense-mediated mRNA degradation (NMD), resulting in loss of protein function, which may have an impact on the generation and coordinated transmission of action potentials throughout the nervous system. Further functional studies will contribute to the molecular mechanism of the disease. and genotype-phenotype severity correlation analysis.

There are some limitations in this study. Firstly, we did not check if the patient carried risk alleles in other DEE risk genes in WES.

Secondly, this study lack the experimental validation for these newly discovered variants and the exploration into genotype-phenotype correlations. Thirdly, there was no direct evidence to establish the causal relationship between the two variants and DEE, further functional validation of the two variants is needed. Future study would offer a more comprehensive description and exploration of the functional aspects within the genomic region where these variants are situated.

In conclusion, combined with the clinical information and genetic variation analysis results of the patient and his family members, this study identified two new loss-of-function variants in the *CACNA1B* gene as the genetic etiology of the patient, and at the same time helped the prenatal diagnosis of the next child in this family to prevent birth defects. It enriched the mutation spectrum and phenotype spectrum of the disease and laid a foundation for subsequent research on the molecular mechanism of the disease. Second, individuals with developmental and epileptic encephalopathy (DEE) may benefit from precision therapy in comparison to those with developmental encephalopathy since patients with epilepsy can get advantages from strong antiepileptic medicines [26]. In order to properly diagnose and treat individuals with DEEs and improve their prognosis, early genetic testing will help to lower the overall burden of epilepsy.

Funding

This research was supported by Key Research and Development Program of Ningxia (No.2021BEG03043)

Data availability statement

All data generated and analyzed during this current study are available from the corresponding author on request.

Ethics approval and consent to participate

This study was approved by the Ethics Committee of the General Hospital of Ningxia Medical University with the informed consent of the child's guardian (2020-664).

CRediT authorship contribution statement

Xin-you Yu: Writing – review & editing, Writing – original draft, Formal analysis, Data curation, Conceptualization. **Qing-mei Sun:** Resources, Investigation, Funding acquisition, Formal analysis, Data curation. **Rui-ping Lu:** Software, Project administration, Investigation, Formal analysis, Data curation. **Bo Wei:** Validation, Supervision, Methodology, Funding acquisition. **Xiao-yan Wang:** Validation, Software, Project administration, Investigation, Data curation. **Li-hua Pan:** Writing – review & editing, Writing – original draft, Visualization, Funding acquisition, Formal analysis, Data curation, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

Not applicable.

References

[1] e.-a.u.e.a, International League against Epilepsy Consortium on Complex Epilepsies. Electronic address, Genetic determinants of common epilepsies: a meta-analysis of genome-wide association studies, Lancet Neurol. 13 (2014) 893–903.
[2] C. Hu, D. Liu, T. Luo, Y. Wang, Z. Liu, Phenotypic spectrum and long-term outcome of children with genetic early-infantile-onset developmental and epileptic encephalopathy, Epileptic Disord. 24 (2022) 343–352.
[3] A. Takata, M. Nakashima, H. Saitsu, T. Mizuguchi, S. Mitsuhashi, Y. Takahashi, N. Okamoto, H. Osaka, K. Nakamura, J. Tohyama, K. Haginoya, S. Takeshita, I. Kuki, T. Okanishi, T. Goto, M. Sasaki, Y. Sakai, N. Miyake, S. Miyatake, N. Tsuchida, K. Iwama, G. Minase, F. Sekiguchi, A. Fujita, E. Imagawa, E. Koshimizu, Y. Uchiyama, K. Hamanaka, C. Ohba, T. Itai, H. Aoi, K. Saida, T. Sakaguchi, K. Den, R. Takahashi, H. Ikeda, T. Yamaguchi, K. Tsukamoto, S. Yoshitomi, T. Oboshi, K. Imai, T. Kimizu, Y. Kobayashi, M. Kubota, H. Kashii, S. Baba, M. Iai, R. Kira, M. Hara, M. Ohta, Y. Miyata, R. Miyata, J.I. Takanashi, J. Matsui, K. Yokochi, M. Shimono, M. Amamoto, R. Takayama, S. Hirabayashi, K. Aiba, H. Matsumoto, S. Nabatame, T. Shiihara, M. Kato, N. Matsumoto, Comprehensive analysis of coding variants highlights genetic complexity in developmental and epileptic encephalopathy, Nat. Commun. 10 (2019) 2506.
[4] K.M. Gorman, E. Meyer, D. Grozeva, E. Spinelli, A. McTague, A. Sanchis-Juan, K.J. Carss, E. Bryant, A. Reich, A.L. Schneider, R.M. Pressler, M.A. Simpson, G. D. Debelle, E. Wassmer, J. Morton, D. Sieciechowicz, E. Jan-Kamsteeg, A.R. Paciorkowski, M.D. King, J.H. Cross, A. Poduri, H.C. Mefford, I.E. Scheffer, T. B. Haack, G. McCullagh, S. Deciphering Developmental Disorders, U.K. Consortium, N. BioResource, J.J. Millichap, G.L. Carvill, J. Clayton-Smith, E.R. Maher, F. L. Raymond, M.A. Kurian, Bi-Allelic loss-of-function CACNA1B mutations in progressive epilepsy-dyskinesia, Am. J. Hum. Genet. 104 (2019) 948–956.
[5] M. Hebbar, H.C. Mefford, Recent advances in epilepsy genomics and genetic testing, F1000Res 9 (2020).
[6] D.I. Kaplan, L.L. Isom, S. Petrou, Role of sodium channels in epilepsy, Cold Spring Harb Perspect Med. 6 (2016).
[7] C. Villa, R. Combi, Potassium channels and human epileptic phenotypes: an updated overview, Front. Cell. Neurosci. 10 (2016) 81.
[8] K.L. Helbig, K.D. Farwell Hagman, D.N. Shinde, C. Mroske, Z. Powis, S. Li, S. Tang, I. Helbig, Diagnostic exome sequencing provides a molecular diagnosis for a significant proportion of patients with epilepsy, Genet. Med. 18 (2016) 898–905.
[9] H.A. Jinnah, Y.V. Sun, Dystonia genes and their biological pathways, Neurobiol. Dis. 129 (2019) 159–168.

[10] M. Yamaguchi, T. Nakayama, Z. Fu, N. Sato, M. Soma, A. Morita, S. Hinohara, N. Doba, T. Mizutani, The haplotype of the CACNA1B gene associated with cerebral infarction in a Japanese population, Hereditas 147 (2010) 313–319.
[11] D. Curtis, A.E. Vine, A. McQuillin, N.J. Bass, A. Pereira, R. Kandaswamy, J. Lawrence, A. Anjorin, K. Choudhury, S.R. Datta, V. Puri, R. Krasucki, J. Pimm, S. Thirumalai, D. Quested, H.M. Gurling, Case-case genome-wide association analysis shows markers differentially associated with schizophrenia and bipolar disorder and implicates calcium channel genes, Psychiatr. Genet. 21 (2011) 1–4.
[12] W.A. Catterall, M.J. Lenaeus, T.M. Gamal El-Din, Structure and pharmacology of voltage-gated sodium and calcium channels, Annu. Rev. Pharmacol. Toxicol. 60 (2020) 133–154.
[13] B.A. Simms, G.W. Zamponi, Neuronal voltage-gated calcium channels: structure, function, and dysfunction, Neuron 82 (2014) 24–45.
[14] C.M. Armstrong, D.R. Matteson, Two distinct populations of calcium channels in a clonal line of pituitary cells, Science 227 (1985) 65–67.
[15] W.A. Catterall, Voltage-gated calcium channels, Cold Spring Harbor Perspect. Biol. 3 (2011) a003947.
[16] W.A. Catterall, E. Perez-Reyes, T.P. Snutch, J. Striessnig, International Union of Pharmacology. XLVIII. Nomenclature and structure-function relationships of voltage-gated calcium channels, Pharmacol. Rev. 57 (2005) 411–425.
[17] X. Jiang, P.K. Raju, N. D'Avanzo, M. Lachance, J. Pepin, F. Dubeau, W.G. Mitchell, L.E. Bello-Espinosa, T.M. Pierson, B.A. Minassian, J.C. Lacaille, E. Rossignol, Both gain-of-function and loss-of-function de novo CACNA1A mutations cause severe developmental epileptic encephalopathies in the spectrum of Lennox-Gastaut syndrome, Epilepsia 60 (2019) 1881–1894.
[18] K.L. Helbig, R.J. Lauerer, J.C. Bahr, I.A. Souza, C.T. Myers, B. Uysal, N. Schwarz, M.A. Gandini, S. Huang, B. Keren, C. Mignot, A. Afenjar, T. Billette de Villemeur, D. Heron, C. Nava, S. Valence, J. Buratti, C.R. Fagerberg, K.P. Soerensen, M. Kibaek, E.J. Kamsteeg, D.A. Koolen, B. Gunning, H.J. Schelhaas, M. C. Kruer, J. Fox, S. Bakhtiari, R. Jarrar, S. Padilla-Lopez, K. Lindstrom, S.C. Jin, X. Zeng, K. Bilguvar, A. Papavasileiou, Q. Xing, C. Zhu, K. Boysen, F. Vairo, B. C. Lanpher, E.W. Klee, J.M. Tillema, E.T. Payne, M.A. Cousin, T.M. Kruisselbrink, M.J. Wick, J. Baker, E. Haan, N. Smith, A. Sadeghpour, E.E. Davis, N. Katsanis, G. Task Force for Neonatal, M.A. Corbett, A.H. MacLennan, J. Gecz, S. Biskup, E. Goldmann, L.H. Rodan, E. Kichula, E. Segal, K.E. Jackson, A. Asamoah, D. Dimmock, J. McCarrier, L.D. Botto, F. Filloux, T. Tvrdik, G.D. Cascino, S. Klingerman, C. Neumann, R. Wang, J.C. Jacobsen, M.A. Nolan, R.G. Snell, K. Lehnert, L.G. Sadleir, B.M. Anderlid, M. Kvarnung, R. Guerrini, M.J. Friez, M.J. Lyons, J. Leonhard, G. Kringlen, K. Casas, C.M. El Achkar, L.A. Smith, A. Rotenberg, A. Poduri, A. Sanchis-Juan, K.J. Carss, J. Rankin, A. Zeman, F.L. Raymond, M. Blyth, B. Kerr, K. Ruiz, J. Urquhart, I. Hughes, S. Banka, S. Deciphering Developmental Disorders, U.B.S. Hedrich, I.E. Scheffer, I. Helbig, G.W. Zamponi, H. Lerche, H.C. Mefford, De novo pathogenic variants in CACNA1E cause developmental and epileptic encephalopathy with contractures, macrocephaly, and dyskinesias, Am. J. Hum. Genet. 103 (2018) 666–678.
[19] J.L. Groen, A. Andrade, K. Ritz, H. Jalalzadeh, M. Haagmans, T.E. Bradley, A. Jongejan, D.S. Verbeek, P. Nurnberg, S. Denome, R.C. Hennekam, D. Lipscombe, F. Baas, M.A. Tijssen, CACNA1B mutation is linked to unique myoclonus-dystonia syndrome, Hum. Mol. Genet. 24 (2015) 987–993.
[20] N.E. Mencacci, L. R'Bibo, S. Bandres-Ciga, M. Carecchio, G. Zorzi, N. Nardocci, B. Garavaglia, A. Batla, K.P. Bhatia, A.M. Pittman, J. Hardy, A. Weissbach, C. Klein, T. Gasser, E. Lohmann, N.W. Wood, The CACNA1B R1389H variant is not associated with myoclonus-dystonia in a large European multicentric cohort, Hum. Mol. Genet. 24 (2015) 5326–5329.
[21] R. Patel, C. Montagut-Bordas, A.H. Dickenson, Calcium channel modulation as a target in chronic pain control, Br. J. Pharmacol. 175 (2018) 2173–2184.
[22] M.E. Williams, P.F. Brust, D.H. Feldman, S. Patthi, S. Simerson, A. Maroufi, A.F. McCue, G. Velicelebi, S.B. Ellis, M.M. Harpold, Structure and functional expression of an omega-conotoxin-sensitive human N-type calcium channel, Science 257 (1992) 389–395.
[23] S. Gao, X. Yao, N. Yan, Structure of human Ca(v)2.2 channel blocked by the painkiller ziconotide, Nature 596 (2021) 143–147.
[24] A.J. Castiglioni, J. Raingo, D. Lipscombe, Alternative splicing in the C-terminus of CaV2.2 controls expression and gating of N-type calcium channels, J. Physiol. 576 (2006) 119–134.
[25] Y. Dong, Y. Gao, S. Xu, Y. Wang, Z. Yu, Y. Li, B. Li, T. Yuan, B. Yang, X.C. Zhang, D. Jiang, Z. Huang, Y. Zhao, Closed-state inactivation and pore-blocker modulation mechanisms of human Ca(V)2.2, Cell Rep. 37 (2021) 109931.
[26] S. Raga, N. Specchio, S. Rheims, J.M. Wilmshurst, Developmental and epileptic encephalopathies: recognition and approaches to care, Epileptic Disord. 23 (2021) 40–52.