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Dissecting the phenotypic and genetic spectrum of early childhood-onset generalized epilepsies

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Highlights

- Early childhood-onset generalized epilepsies have an overlapping genetic and phenotypic spectrum.
- Two-thirds of the pathogenic variants were found in *SLC6A1*, *SLC2A1*, and *SYNGAP1*.
- *SLC6A1* mutation is responsible for 15% (3 out of 20) of early onset absence epilepsy.

Abstract

Purpose: Although the genetic and clinical aspects of epilepsy with myoclonic-atonic seizures (MAE) and early onset absence epilepsy (EOAE) have been investigated thoroughly, other early childhood-onset generalized epilepsies that share clinical features with MAE and EOAE have not been characterized. In this study, we aimed to delineate the genetic and phenotypic spectrum of early childhood-onset generalized epilepsies, including MAE and EOAE.

Methods: We recruited 61 patients diagnosed with MAE, EOAE, genetic epilepsy with febrile seizure plus (GEFS+) and unclassified generalized epilepsies that shared seizure onset age and

seizure types. Genetic causes were investigated through targeted gene panel testing, whole exome sequencing, chromosomal microarray, and single-gene Sanger sequencing.

Results: We classified 11 patients with MAE, 20 with EOAE, 9 with GEFS+ spectrum. Epilepsy syndrome was not specified in the remaining 21 patients. The clinical features were comparable across groups. Nevertheless, patients with EOAE tended to show better developmental and seizure outcomes. A total of 23 pathogenic sequences and copy number variants from 12 genes were identified (23/61, 37.7%). Genetic etiologies were confirmed in 36.4% (4/11) of the MAE group, 45% (9/20) of the EOAE group, 22.2% (2/9) of the GEFS+ spectrum, and 38.1% (8/21) of the unclassified group. The most frequently identified genes with pathogenic variants were *SLC6A1* (7 patients), *SLC2A1* (4 patients), and *SYNGAP1* (4 patients).

Conclusion: Early childhood-onset generalized epilepsy appeared to be characterized by an overlapping genetic and phenotypic spectrum. *SLC6A1* and *SLC2A1* appeared to be important genetic causes of early childhood-onset generalized epilepsy.

Keywords

epilepsy with myoclonic-atonic seizures, early onset absence epilepsy, genetic testing

1. INTRODUCTION

After Doose et al. [1] described epilepsy with myoclonic-atonic seizure (MAE), exploration of genetic causes has been ongoing [2]. Recent studies have identified pathogenic variants in *SLC2A1* and *SLC6A1* in 4% and 5% of MAE patients, respectively [3, 4]. *SLC2A1* has also been identified as responsible for up to 10% of early onset absence epilepsy (EOAE) patients [5-7].

Similarly, EOAE and childhood absence epilepsy have also been reported in patients with pathogenic variants in *SLC6A1* [8]. Therefore, although the principal seizure type may characterize MAE and EOAE as distinct epilepsy syndromes, they may share a genetic etiology. In practice, when stringent criteria for epilepsy syndrome are applied, many patients with clinical features that overlap with MAE or EOAE remain unclassified. One research group has proposed a new epilepsy syndrome named early childhood myoclonic epilepsy, including a subset of previously unclassified patients [9]. However, early childhood myoclonic epilepsy could be simultaneously classified as MAE when the broad criteria for MAE are applied [3].
The spectrum of genetic epilepsy with febrile seizure plus (GEFS+) has evolved to include phenotype ranging from febrile seizure to MAE and Dravet syndrome [10]. Therefore, a proportion of patients with early childhood-onset generalized seizures could be categorized into GEFS+ spectrum if associated with an appropriate family history or febrile/afebrile generalized tonic-clonic seizures (GTCs).

As Doose et al. [1] acknowledged the overlapping subtypes in his original cohort, the current nosological issue of early childhood-onset generalized epilepsy classification is such that it consists of a heterogeneous mixture of patients with varying genetic etiologies. Considering the overlapping clinical features of early childhood-onset generalized epilepsy patients, a comprehensive genetic study is needed to better define the genetic and phenotypic spectra of this group. In the present study, we performed genetic testing of patients with early childhood-onset generalized epilepsy. The study cohort included MAE, EOAE, GEFS+ spectrum, and unclassified patients who shared seizure onset age and 1 or more types of generalized seizures. We also performed an in-depth analysis of the cohort's genetic and phenotypic spectra.

2. Methods

2.1. Patients

This study was approved by the institutional review board (IRB) of Seoul National University Hospital (IRB No. 1009-027-331 for genetic testing and 1804-052-936 for retrospective review of medical records). All patients or their legal representatives provided written informed consent.

Inclusion criteria were as follows: (1) onset of generalized seizures (myoclonic, myoclonic-atonic, atonic, typical/atypical absence) between 7 months to 6 years of age; (2) 2–4 Hz generalized spike/polyspike and slow waves on an electroencephalogram (EEG); and (3) absence of brain structural abnormalities. Patients with Dravet syndrome and Lennox–Gastaut syndrome were excluded. For the diagnosis of MAE, either video-EEG-proven myoclonic-atonic seizures or myoclonic seizure plus history of frequent drop attacks was required. For the diagnosis of EOAE, a typical absence seizure with an ictal EEG of 2.5–3 Hz generalized rhythmic spike-wave was required. We used the recently proposed definition of GEFS+ and febrile seizure plus [10] and classified the patients who met the definition as being a group with GEFS+ spectrum. If MAE or EOAE was identified among GEFS+ family, we classified them as GEFS+ spectrum. The proposed upper age limit of seizure onset differs between MAE (6 years of age) and EOAE (4 years of age). We decided to use the upper age limit of inclusion as 6 years of age to include a broader range of patients. Accordingly, 13 patients whose seizure onset was between 4 and 6 years of age were included. When patients could not be classified as having one of MAE, EOAE or GEFS+ spectrumthey were grouped as unclassified. The specific types of generalized seizures

were defined according to the guidelines of the International League Against Epilepsy (ILAE) (www.epilepsydiagnosis.org).

Based on these criteria, we enrolled a total of 61 patients; i.e., 11 with MAE, 20 with EOAE, 9 with GEFS+ spectrum, and 21 were unclassified. The following clinical variables were collected for all enrolled patients: age of seizure onset, follow-up duration, type of generalized seizure, family history, developmental status before and after the seizure onset, comorbidities, EEG, video-EEG, and response to treatment.

2.2. Genetic testing

The various genetic tests were performed for each patient based on their clinical presentation and the availability of testing. Briefly, single-gene testing and chromosomal array testing were used as first-tier genetic testing in 41 patients. If the genetic cause was not identified by these tests, we performed either targeted-gene panel testing or whole-exome sequencing as the final genetic testing. Twenty patients had gene-panel sequencing or whole-exome sequencing as their first-tier test. The detailed genetic testing methods performed for each patient are provided in Supplementary Table 1.

For targeted gene panel sequencing, we used custom-designed SureSelect Target Enrichment System Kits (Agilent Technologies, Santa Clara, CA, USA) that included 127 genes (Supplementary Table 2). Library preparation was performed according to the manufacturer's instructions (Agilent Technologies). The library was paired-end sequenced on a HiSeq 2500 sequencing system (Illumina, San Diego, CA, USA). For variant annotation, we aligned paired-end sequencing reads with lengths of 101 bp to the Genome Reference Consortium Human Build

37 (patch release 13) using the Burrows-Wheeler Aligner alignment tool (version 0.7.13). Picard software (v. 2.1.1), SAM (v. 1.3.1), and the Genome Analysis Toolkit (GATK, v. 3.8) were used in subsequent data analyses. Variant calling was performed using GATK's Haplotype Caller. We employed several databases along with RefSeq using ANNOVAR for variant annotation. The allele frequencies were referenced from the Exome Aggregation Consortium database. We also searched for all variants in the Human Gene Mutation Database. The pathogenicity of novel variants was evaluated according to the American College of Medical Genetics and Genomics (ACMG) standards and guidelines.

For whole exome sequencing, we used the SureSelect Human All Exon V5 capture kit (Agilent Technologies) for target enrichment. Library preparation, sequencing, and sequence analysis followed the same procedures as targeted gene panel sequencing. We only focused on 127 epilepsy-related genes for downstream analysis of the whole exome sequencing data.

CMA analysis was performed using a human genome oligonucleotide comparative genomic hybridization microarray (Agilent Technologies). The detailed protocol was described previously [11].

All putative sequence variants were confirmed by Sanger sequencing. Family member DNA, when available, was also sequenced to confirm familial segregation of the variant.

3. Results

3.1. Phenotypic features

Among the 61 patients enrolled, 34 were male and the average age of seizure onset was 35.6 months (range, 10–58 months). Twenty patients (20/61, 32.8%) had a history of febrile seizure

before their first unprovoked seizure. A family history of epilepsy or febrile seizure was noted in 14 patients (23.0%). Twenty-four patients (24/61, 39.3%) showed developmental delay at seizure onset and an additional 11 patients developed a mild-to-moderate degree of intellectual disability after seizure onset. Eleven patients (11/61, 18.0%) had psychiatric comorbidities including attention deficit–hyperactivity disorder, affective disorder, behavioral disorder, and anxiety disorder. The mean follow-up duration was 4.2 years (range, 1 month–11 years) and the mean age at last follow-up was 8.3 years (range, 2.1–18.3 years). All but 1 patient had been treated with 1 or more antiepileptic drugs (AEDs), and 37 of 61 (60.7%) achieved a seizure-free state. Among the 37 seizure-free patients, 14 discontinued AEDs after a mean seizure-free duration of 33 months (range, 17–51 months). The remaining 23 patients remained seizure-free for 34.9 months (range, 6–96 months) with an average of 1.8 AEDs (range, 1–4 AEDs). Twenty-four patients had recurrent seizures and their mean follow-up period was 3.3 years (range, 3 months–7.4 years).

We classified the 61 patients into 4 subgroups: MAE, EOAE, GEFS+ spectrum and unclassified. The seizure type was determined based on 24-hour continuous video-EEG monitoring in 40 patients (65.6%) and by interictal EEG and parental seizure report in 21 patients (34.4%). Eleven patients were categorized as having MAE (18.0%), 20 as having EOAE (32.8%), 9 as having GEFS+ spectrum (14.8%), and 21 were unclassified (34.4%). The clinical manifestations according to classification are summarized in Table 1. A detailed description of each patient is provided in Supplementary Table 3. Patients in the MAE and unclassified groups showed similar clinical features with respect to seizure types, developmental status, and seizure outcomes. Most of the EOAE patients did not present other generalized seizures except for

typical absence seizures. EOAE patients tended to show less developmental delay before seizure onset and better seizure outcomes than other groups.

3.2. Genetic analysis

A total of 23 pathogenic or likely pathogenic variants from 11 genes were identified (Table 2). Fifteen of the variants were identified from gene panel sequencing, 4 from Sanger sequencing of a single gene, 3 from whole exome sequencing, and 1 from CMA. Family studies were performed in 19 cases and 16 had a *de novo* variant. Two variants (Cases 5 and 33) were inherited from 1 of the asymptomatic parents, but were classified as pathogenic or likely pathogenic according to ACMG guidelines. *SLC6A1* was the most frequently identified causative gene, found in 7 patients (Fig. 1). *SYNGAP1* and *SLC2A1* were each found in 4 patients. On CMA testing, case 43 was found to have a microdeletion of chromosome 3p25 (position 9710458-12316964, 2.54Mb). This included all of *SLC6A1*. Case 13 was a male patient who was supposed to carry a *de novo* mosaic variant in *PCDH19* (Supplementary Fig. 1). We identified 8 variants with unknown significance from 6 genes (Supplementary Table 4). Four variants that met the PM2 criteria of the ACMG guidelines (absence from controls) were classified as variants of unknown significance due to absence of family studies.

3.3. Genetic and phenotypic spectrum

The diagnostic yield for the entire cohort was 37.7% (23/61). When the diagnostic yield was compared according to each subgroup, 4/11 (36.4%) in the MAE group, 9/20 (45.0%) in the EOAE, 2/9 (22.2%) in the GEFS+ spectrum group, and 8/21 (38.1%) in the unclassified group had a confirmatory genetic diagnosis (Fig. 2). Variants in *SLC6A1* and *SLC2A1* were the most

frequently mutated and were distributed among MAE, EOAE, and unclassified group. Mutation in *SYNGAP1* was also identified in 4 patients. The clinical presentations of these patients are displayed in Table 3. All patients with a *SYNGAP1* mutation had atypical absence seizures and were placed in the unclassified group. Three-fourths of *SLC2A1*- or *SYNGAP1*-positive patients showed poor treatment response, whereas 70% of patients with *SLC6A1* achieved a seizure-free state. Compared with the mutation-negative group, the mutation-positive group had a higher proportion of developmental delay (73.9% vs. 47.3%, Table 4). More than two-thirds of the patients with developmental delay in the mutation-positive group showed delay prior to seizure onset. The proportion of seizure-free patients between the 2 groups was comparable (56.5% vs. 63.1%). Nevertheless, fewer seizure-free patients in the mutation-positive group were eventually able to discontinue AEDs (2/23, 8.7%) than in the mutation-negative group (11/38, 28.9%).

4. Discussion

In 1989, ILAE proposed that MAE be defined according to the following electroclinical features: normal development before onset of epilepsy; onset of myoclonic, myoclonic-astatic (atonic), or astatic (atonic) seizures between 7 months and 6 years of age; and the presence of generalized spike or polyspike wave discharges on EEG [12]. However, the use of inclusion criteria has been inconsistent between studies, especially with respect to seizure type and developmental status [13-16]. Because myoclonic-atonic seizures are difficult to confirm without a neurophysiologic study, the presence of either myoclonic or atonic seizure has been regarded as sufficient to fulfill the diagnostic criteria for MAE [3, 13]. The patients with developmental delay before seizure onset were also included for the genetic study of *SLC2A1* and *SLC6A1* [3, 4]. EOAE also faced a

similar nosological issue, although this syndrome is not currently recognized by the ILAE. EOAE was defined as an earlier onset (before 4 years of age) version of childhood absence epilepsy in the original study by Suls et al. that addressed *SLC2A1* mutations in EOAE patients [5]. While studies using stringent criteria failed to identify *SLC2A1* mutations in EOAE patients [15, 17], studies that included patients with developmental delay and generalized seizures other than typical absence seizure replicated the results [6, 7].

According to this evidence, we attempted to recruit a broad spectrum of patients for comprehensive genetic testing to better define the genotype and phenotype spectra of this group. We found that the MAE and unclassified groups showed similar clinical features, especially for seizure and developmental outcome that were consistent with those of a previous MAE study [2]. Diagnostic yield of genetic testing was also comparable between these 2 groups (36.4% vs. 38.1%). The EOAE group tended to show slightly better development and seizure outcome. Diagnostic yield of the EOAE group was slightly higher than that of the other groups (45.0%). We obtained a molecular diagnosis for about one fifth (22.2%) of patients with GEFS+ spectrum, which was lower than for other subgroups. Among the 23 pathogenic variants in 11 genes, pathogenic variants in *SLC2A1* and *SLC6A1* comprised nearly half (47.8%, 11/23) of the genetic etiologies in our cohort. Therefore, our results showed significant overlap among subgroups with respect to genetic etiology, although they are categorized as different epilepsy syndromes. Because the current classification of epilepsy syndrome inevitably includes subtypes with different genetic etiologies, lumping the subgroups into early childhood-onset generalized epilepsy would be a practical way to obtain a better diagnostic yield from genetic tests.

The *SLC6A1* gene encodes a gamma-aminobutyric acid (GABA) transporter (GAT1) and alteration in GAT1 leads to aberrant tonic $GABA_A$ inhibition, which results in absence seizures in GAT-1 knockout mice [18]. We noted that 3 out of 7 patients with pathogenic variants in *SLC6A1* were classified in the EOAE group. Moreover, typical or atypical absence seizures were the most frequent seizure type in the group with pathogenic variants in *SLC6A1*. This result is also consistent with those of a recent study that reported absence seizures as the main type linked with *SLC6A1* mutations [8]. Thus, although the main epilepsy syndrome has been regarded as MAE [4, 8], EOAE might be an underestimated epilepsy syndrome related to *SLC6A1*.

The 4 patients with pathogenic *SYNGAP1* variants showed atypical absence seizures as the main seizure type and all showed developmental delay before seizure onset. The reported seizure types in patients with pathogenic *SYNGAP1* variants consisted of myoclonic, atonic, myoclonic absences or absences [19-21]. Three patients were classified as having MAE in the previous study [21]. Nevertheless, previous studies did not analyze the epilepsy phenotype of the *SYNGAP1* mutation in detail [19-21]. Together with our data, the evidence suggests that *SYNGAP1* also appears to be responsible for a significant proportion of the genetic etiology in early childhood-onset generalized epilepsy.

The pathogenic variants in the other 8 genes were found in only 1 patient. *GABRG2* and *SCN1A* are known to be linked with GEFS+. In our study, the pathogenic variants from these 2 genes were found in GEFS+ spectrum group which confirms a previous finding that these genes are rare causes of MAE and usually occur against the background of GEFS+ [22]. A propensity for generalized seizures in patients with pathogenic variants in *KCNA2*, *IQSEC2*, and *HNRNPU*

has been reported [23-25]. Therefore, early childhood-onset generalized epilepsy might lie within the phenotypic spectrum of these genes and should be further demonstrated in future studies.

Should confirming a genetic etiology be a component of characterizing subgroups? While this question could best be answered on an individual gene basis, we found a trend such that the mutation-positive group showed a higher proportion of patients with developmental delay before seizure onset (56.5% vs. 28.9%). With respect to seizure outcome, fewer patients in the mutation-positive group were in long-term remission after AED discontinuation (8.7% vs. 28.9%). This tendency could be explained partly by the fact that many of the genes identified in our study have also been implicated in neurodevelopmental disorders. Therefore, neurodevelopmental status needs to be weighed equally with seizure-related features to better characterize the genotype and phenotype spectrum.

In conclusion, we provided a detailed description of the genetic and phenotypic spectrum of early childhood-onset generalized epilepsy based on shared seizure onset age and seizure types. Our results suggest overlapping genetic etiologies by demonstrating *SL6A1* and *SLC2A1* mutations across subgroups.

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Disclosure

The authors have no conflict of interest to disclose.

Ethical Publication Statement

We confirm that we have read the journal's position on issues regarding ethical publication, and we affirm that this report is consistent with those guidelines.

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Table 1. Summary of clinical information according to each subgroup.

	MAE (n=11)	EOAE (n=20)	GEFS+ spectrum (n=9)	Others (n=21)
Sex (M:F)	7:4	6:14	7:2	14:7
Seizure onset age (months) (Median, (range))	31.3 (13-59)	42.6 (12-58)	34.9 (27-47)	31.5 (10-59)
Follow-up duration (years) (Median, range)	3.1 (0.1-10.5)	4.8 (0.3-21.1)	4.0 (0.1-7.8)	4.3 (0.1-13.1)
Family history (n, (%))				
Febrile seizure	0 (0.0)	3 (15.0)	2 (22.2)	3 (14.3)
Epilepsy	2 (18.2)	0 (0.0)	3 (33.3)*	2 (9.5)
History of febrile seizure (n, (%))	4 (36.4%)	3 (15.0)	9 (100.0)	4 (19.0)
Seizure types (n, (%))				
Myoclonic	11 (100.0)	1 (5.0)	6 (66.7)	8 (38.1)
Myoclonic-atonic	1 (9.1)	0 (0.0)	0 (0.0)	0 (0.0)
Atonic	10 (91.0)	0 (0.0)	1 (11.1)	2 (9.5)
Absence	2 (18.2)	20 (100.0)	2 (22.2)	2 (9.5)
Atypical absence	2 (18.2)	0 (0.0)	6 (66.7)	17 (81.0)
GTC	4 (36.4)	5 (25.0)	9 (100.0)	5 (23.8)
Developmental delay				
Before seizure onset	6 (54.5)	3 (15.0)	3 (33.3)	12 (57.1)
After seizure onset	1 (9.1)	6 (30.0)	1 (11.1)	3 (14.3)
Psychiatric comorbidity (n, (%))	1 (9.1)	2 (10.0)	0 (0.0)	1 (4.8)
ADHD	0 (0.0)	1 (5.0)	0 (0.0)	1 (4.8)
Behavioral problem	0 (0.0)	3 (15.0)	1 (11.1)	0 (0.0)
Affective/anxiety disorder	0 (0.0)	0 (0.0)	1 (11.1)	0 (0.0)
ASD				
No. of last AEDs (median, range)	2.3 (1-4)	1.5 (1-3)	2.1 (1-3)	1.9 (1-4)
Treatment response (n, (%))				
Seizure-free without AEDs	0 (0.0)	5 (25.0)	3 (33.3)	5 (23.8)
Seizure-free with AEDs	5 (45.5)	10 (50.0)	4 (44.4)	5 (23.8)
Uncontrolled	6 (54.5)	5 (25.0)	2 (22.2)	11 (52.4)

MAE, epilepsy with myoclonic-atonic seizures; EOAE, early onset absence epilepsy; GEFS+, genetic epilepsy with febrile seizure plus; GTC, generalized tonic clonic; ADHD, attention deficit–hyperactivity disorder; ASD, autism spectrum disorder; AEDs, antiepileptic drugs

* A patient has family history of epilepsy as well as febrile seizure

Table 2. Profile of pathogenic or likely pathogenic variants

Patient No.	Test	Gene	Nucleotide change	Amino acid change	Inheritance pattern	ACMG Classification
Case 12	Panel	*ANKRD11*	c.2398_2401delGAAA	p.E800Nfs	De novo	Pathogenic
Case 19	Panel	*CACNA1A*	c.C1900T	p.Q634X	De novo	Pathogenic
Case 33	Panel	*GABRG2*	c.C1273T	p.R425X	Asymptomatic father	Pathogenic
Case 2	Panel	*HNRNPU*	c.C691T	p.R231X	De novo	Pathogenic
Case 3	Panel	*IQSEC2*	c.G136T	p.E46X	De novo	Pathogenic
Case 22	WES	*KCNA2*	c.C466T	p.P156S	De novo	Pathogenic
Case 13	Panel	*PCDH19*	c.A1606T	p.K536X	De novo	Pathogenic
Case 36	Panel	*SCN1A*	c.G937T	p.D313Y	Symptomatic father	Pathogenic
Case 59	Sanger	*SLC2A1*	c.T1300A	p.F434I	De novo	Pathogenic
Case 25	Sanger	*SLC2A1*	c.G458A	p.R153H	De novo	Pathogenic
Case 26	Sanger	*SLC2A1*	c.G904A	p.G312S	De novo	Pathogenic
Case 5	Panel	*SLC2A1*	c.G940C	p.G314R	Asymptomatic mother	Likely pathogenic
Case 46	Panel	*SLC6A1*	c.C130T	p.R44W	NA	Likely pathogenic
Case 16	Panel	*SLC6A1*	c.C456A	p.Y152X	De novo	Pathogenic
Case 21	Panel	*SLC6A1*	c.G1323+1A	-	NA	Pathogenic
Case 27	Panel	*SLC6A1*	c.G913C	p.A305P	De novo	Likely pathogenic
Case 6	WES	*SLC6A1*	c.C1070T	p.A357V	De novo	Likely pathogenic
Case 43	CMA	*SLC6A1*	Whole deletion	Whole deletion	De novo	Pathogenic
Case 47	Panel	*SLC6A1*	c.850-1G>T	-	NA	Pathogenic
Case 50	Panel	*SYNGAP1*	c.2014delA	p.T672Rfs	De novo	Pathogenic
Case 53	WES	*SYNGAP1*	c.531-532delTA	p.F177Lfs	De novo	Pathogenic
Case 58	Panel	*SYNGAP1*	c.G2116-1A	-	De novo	Pathogenic

Case 56	Sanger	*SYNGAP1*	c.1219delC	p.L407Ffs	NA	Pathogenic

WES, whole exome sequencing; CMA, chromosomal microarray; NA, not applicable

Table 3. Phenotypic description of patients with mutations in *SLC6A1*, *SLC2A1*, and *SYNGAP1*

	SLC6A1 (n=7)	*SLC2A1* (n=4)	*SYNGAP1* (n=4)
Epilepsy syndrome (n, (%))			
MAE	1 (14.3)	1 (25.0)	0 (0.0)
EOAE	3 (42.9)	2 (50.0)	0 (0.0)
GEFS+ spectrum	0 (0.0)	0 (0.0)	0 (0.0)
Unclassified	3 (42.9)	1 (25.0)	4 (100.0)
Seizure type (n, (%))			
Myoclonic	1 (14.3)	1 (25.0)	1 (25.0)
Myoclonic-atonic	0 (0.0)	0 (0.0)	0 (0.0)
Atonic	2 (28.6)	1 (25.0)	1 (25.0)
Absence	4 (57.1)	3 (75.0)	0 (0.0)
Atypical absence	3 (42.9)	1 (25.0)	4 (100.0)
GTC	1 (14.3)	1 (25.0)	0 (0.0)
Developmental delay (n, (%))			
Before seizure onset	4 (57.1)	2 (50.0)	4 (100.0)
After seizure onset	2 (28.6)	0 (0.0)	0 (0.0)
Psychiatric comorbidity (n, (%))	1 (14.3)	2 (50.0)	1 (25.0)
Treatment response (n, (%))			
Seizure-free without AEDs	1 (14.3)	0 (0.0)	0 (0.0)
Seizure-free with AEDs	4 (57.1)	1 (25.0)	1 (25.0)
Uncontrolled	2 (28.6)	3 (75.0)	3 (75.0)

MAE, epilepsy with myoclonic-atonic seizures; EOAE, early onset absence epilepsy; GEFS+, genetic epilepsy with febrile seizure plus; GTC, generalized tonic clonic; AEDs, antiepileptic drugs

Table 4. Comparison between mutation positive and negative group

	Mutation-positive (n=23)	*Mutation-negative (n=38)*
Epilepsy syndrome (n, (%))		
MAE	4 (17.4)	)7 (18.4)
EOAE	9 (39.1)	11 (28.9)
GEFS+ spectrum	2 (8.7)	7 (18.4)
Unclassified	8 (34.8)	13 (34.2)
Follow-up duration (years, (range))	5.19 (0.1-13.1)	3.63 (0.1-21.1)
Seizure type (n, (%))		
Myoclonic	6 (26.1)	13 (34.2)
Myoclonic-atonic	1 (4.3)	0 (0.0)
Atonic	5 (21.7)	0 (0.0)
Absence	12 (52.2)	9 (23.7)
Atypical absence	9 (39.1)	14 (36.8)
GTC	7 (30.4)	12 (31.6)
Developmental delay (n, (%))		
Before seizure onset	13 (56.5)	11 (28.9)
After seizure onset	4 (17.4)	7 (18.4)
Psychiatric comorbidity (n, (%))	6 (26.1)	5 (13.2)
Treatment response (n, (%))		
Seizure-free without AEDs	2 (8.7)	11 (28.9)
Seizure-free with AEDs	11 (47.8)	13 (34.2)
Uncontrolled	10 (43.5)	14 (36.8)

MAE, epilepsy with myoclonic-atonic seizures; EOAE, early onset absence epilepsy; GEFS+, genetic epilepsy with febrile seizure plus; GTC, generalized tonic clonic; AEDs, antiepileptic drugs

Figures

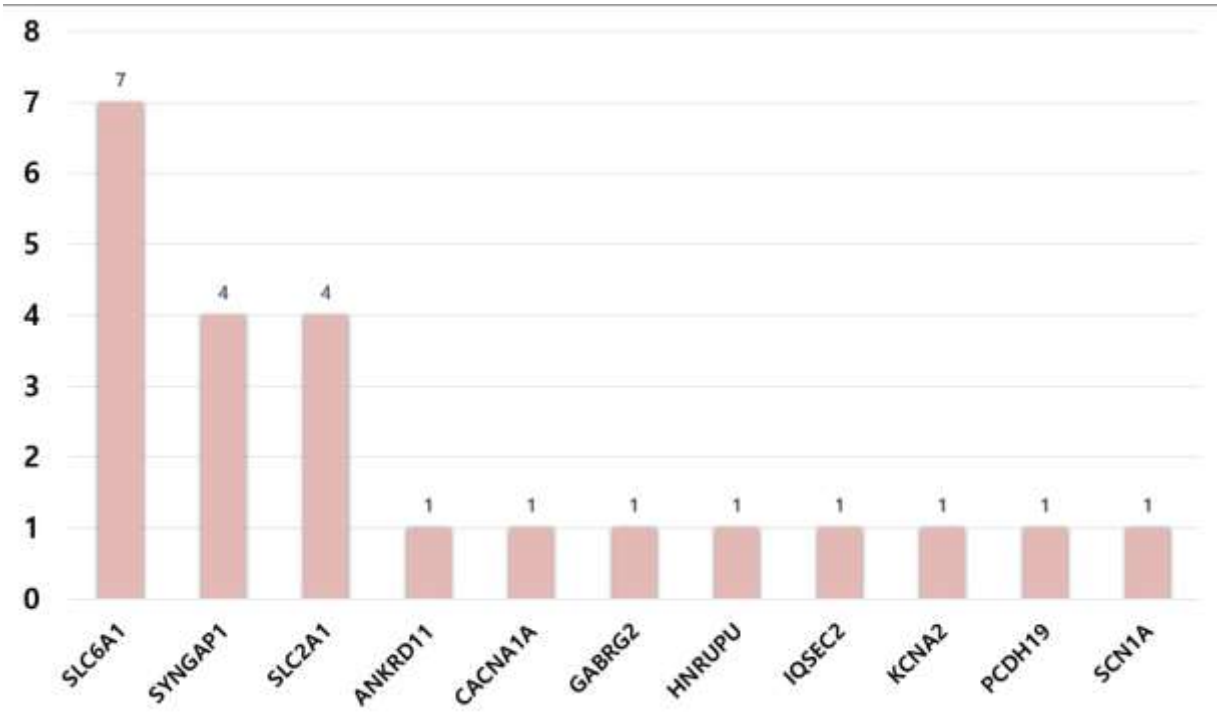
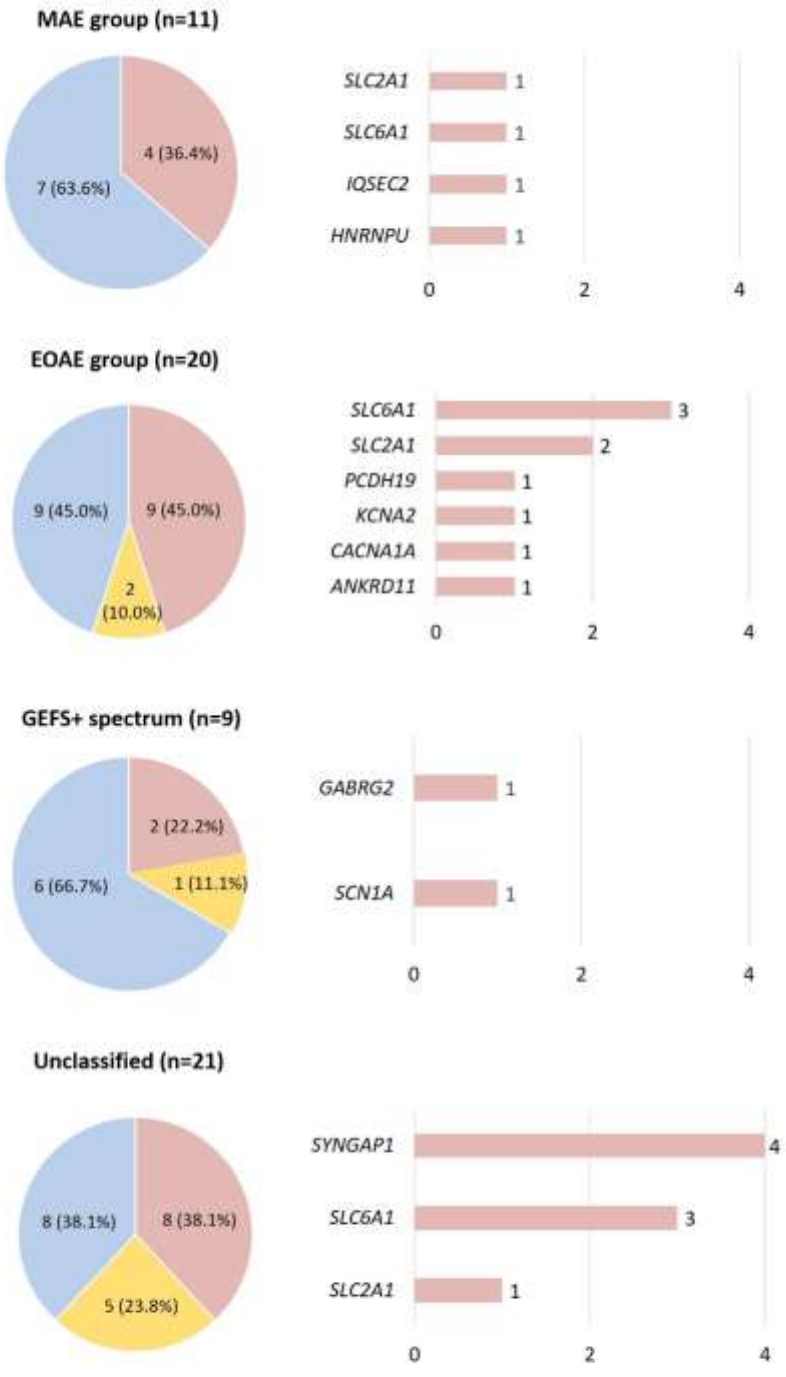


Fig. 1. Frequency of gene with pathogenic or likely pathogenic variants



MAE, epilepsy with myoclonic-atonic seizures; EOAE, early onset absence epilepsy; GEFS+, generalized epilepsy with febrile seizure plus

Fig. 2. Diagram comparing the diagnostic yield of genetic testing between 3 groups. The frequency of genes with pathogenic or likely pathogenic variants in each group is shown as a bar graph