Seizure variables and their relationship to genotype and functional abilities in the CDKL5 disorder

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ABSTRACT

Objective: To investigate seizure outcomes and their relationships to genotype and functional abilities in individuals with the cyclin-dependent kinase-like-5 (CDKL5) disorder.

Methods: Using the International CDKL5 Disorder Database, we identified 172 cases with a pathogenic *CDKL5* mutation. We categorized individual mutations into 4 groups based on predicted structural and functional consequences. Negative binomial regression was used to model the linear association between current seizure rate and mutation group, current level of assistance required to walk 10 steps, and the highest level of expressive communication used to convey refusal or request.

Results: All but 3 (169/172) patients had a history of epilepsy. The median age at seizure onset was 6 weeks (range 1 week–1.5 years) and the median seizure rate at ascertainment was 2 per day (range 0–20 per day). After adjusting for walking ability and confounders including use or otherwise of polytherapy, seizure rate was lower in those with truncating mutations between aa172 and aa781 compared to those with no functional protein (incidence rate ratio [IRR] 0.57; 95% confidence interval [CI] 0.35–0.93). Ability to walk and use of spoken language were associated with lower rates of current seizures when compared to those with the least ability after adjusting for genotype (walking: IRR 0.62; 95% CI 0.39–0.99, communication: IRR 0.48; 95% CI 0.23–1.02). At a median age at questionnaire completion of 5 years, those previously treated with corticosteroids had more frequent seizures than those who have never been treated, whether or not there was a history of infantile spasms.

Conclusions: Epilepsy is pervasive but not mandatory for the CDKL5 disorder. Genotype and functional abilities were related to seizure frequency, which appears refractory to antiepileptic drugs. ***Neurology*® 2016;87:2206–2213**

GLOSSARY

aa = amino acid; **AED** = antiepileptic drug; **CDKL5** = cyclin-dependent kinase-like-5; **CI** = confidence interval; **IRR** = incidence rate ratio; **VNS** = vagal nerve stimulation.

The CDKL5 disorder, caused by mutations in the cyclin-dependent kinase-like-5 (*CDKL5*) gene,[1] originally thought to be an atypical form of Rett syndrome,[2] is now considered a distinct disorder in its own right.[3] Hallmark features include early-onset epilepsy within the first 3 months of life and severe neurodevelopmental problems.[3] Nevertheless, information on seizure outcomes is limited, with reports mainly of refractory seizures but also periods of seizure freedom.[4–12] Most clinical information on the CDKL5 disorder has been derived from case studies with modest numbers of participants (4–12 individuals per study).[1,5,9–12] Adequately sized studies are needed in order to help in prognostication, aid in the allocation of resources for affected individuals, and increase knowledge on *CDKL5* function, leading to development of better treatments. While it is difficult to confirm seizure types using parental information, assessing seizure frequency from parental reporting is standard in clinical practice and is used

Supplemental data at Neurology.org

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Table 1 Characteristics of individuals with pathogenic *CDKL5* mutation by epilepsy status (n = 172)

	Epilepsy group (n = 169)	No epilepsy group (n = 3)
Sex, n (%)		
Female	147 (87.0)	2 (66.7)
Country of residence, n (%)		
United States	87 (51.5)	2 (66.7)
Non-United States	82 (48.5)	1 (33.3)
Age at ascertainment, y, n (%)		
<1.5	25 (14.8)	0
1.5-6	77 (45.6)	1 (33.3)
7-12	45 (26.6)	1 (33.3)
>12	22 (13.0)	1 (33.3)
Mutation group, n (%)		
No functional protein	50 (29.6)	1 (33.3)
Missense/in-frame mutations within catalytic domain	43 (25.4)	1 (33.3)
Truncating mutations between aa172 and aa781	52 (30.8)	0
Truncating mutations after aa781	13 (7.7)	0
Mutation not grouped	11 (6.5)	1 (33.3)
Age at seizure onset (n = 167), wk, median (range)	6 (1-78)	
History of infantile spasms (n = 142), n (%)	48 (33.8)	
Current seizure rate (n = 153), n (%)		
Seizure-free	12 (7.8)	
Monthly	15 (9.8)	
Weekly	17 (11.1)	
Daily, 1-5/d	86 (56.2)	
Daily, >5/d	23 (15.0)	
Honeymoon period (n = 59), mo, median (range)	6 (2.5-72)	
Ever been treated with steroids/ACTH (n = 141), n (%)	43 (30.5)	
No. AEDs currently used (n = 159), n (%)		
0	18 (11.3)	
1	25 (15.7)	
2	44 (27.7)	
3	47 (29.6)	
4	23 (14.5)	
5 or more	2 (1.3)	
Current use of ketogenic diet, n (%)		
No	145 (85.8)	
Yes	24 (14.2)	
Current use of vagal nerve stimulation, n (%)		
No	143 (84.6)	
Yes	26 (15.4)	
Standing for 20 seconds (n = 134[a]), n (%)		
Unable	71 (54.2)	
Assisted	32 (24.4)	

Continued

as a benchmark in randomized clinical trials investigating efficacy of antiepileptic drugs. Using data from an ongoing international CDKL5 patient registry,[13] populated primarily by parents about their children, we have demonstrated variability of attainment of developmental milestones and some relationships with genotype. Building on this work, the current study investigated seizure outcomes in individuals with the CDKL5 disorder and relationships with genotype, walking, and expressive communication skills.

METHODS Established in 2012, the International CDKL5 Disorder Database collects information from families/caregivers of a child with the CDKL5 disorder through completion of online or paper questionnaires.[13] As of January 2016, data on 241 individuals were recorded in the Database. Cases were included in this study if the *CDKL5* variant was considered to be pathogenic and information on epilepsy had been provided.

As previously described,[13] *CDKL5* mutations were grouped according to predicted structural and functional consequences. The groups were (1) variants resulting in no functional protein (including variants causing loss of the functional components in the catalytic domain before amino acid [aa] 172 and full gene deletions), (2) missense/in-frame variants within the catalytic domain (includes missense variants within the protein's kinase active region or in-frame variants), (3) truncating variants located between aa172 and aa781 (includes any variants resulting in a truncation such as nonsense or frameshift variants potentially resulting in maintaining kinase activity but loss of the C-terminal region), and (4) truncating variants occurring after aa781 (maintains kinase activity and the majority of the C-terminal region). Other mutations of unknown significance (e.g., duplication) were not included in the analysis. Based on appropriate developmental milestones for this disorder and the age distribution of our sample, age at ascertainment was categorized as less than 1.5 years, 1.5–6 years, 7–12 years, or more than 12 years.

Families/caregivers were asked to report on concerns during the neonatal period. Age at seizure onset was the age at first observation of seizures. Current seizure rate was the average number of seizures per day at the time of ascertainment and, for descriptive purposes, was categorized as seizure-free (for at least 2 months), monthly, weekly, 1 to 5 a day, or more than 5 a day. Families/caregivers also reported the longest prior seizure-free ("honeymoon") period of more than 2 months including start and end age. Monotherapy was defined as the current use of any single antiepileptic drug (AED) and polytherapy was concurrent use of 2 or more prescribed AEDs at the time of ascertainment. Ever been treated with steroids/ACTH and current use of nonpharmaceutical therapies such as ketogenic diet and vagal nerve stimulation (VNS) were both coded as either yes or no.

Families/carers were asked to indicate the current level of assistance required by their child to stand for 20 seconds or walk for 10 steps, coded as needing no assistance (independent), needing some assistance (assisted), or unable to complete/needing maximal assistance (unable). Responses were then dichotomized into yes (independent and assisted) or no (unable). The highest level of expressive communication was derived from responses to questions about the communication the child used to convey refusal or requesting an object or experience. Responses were

Table 1 Continued

	Epilepsy group (n = 169)	No epilepsy group (n = 3)
Independent	28 (21.4)	3 (100)
Walking for 10 steps (n = 133[a]), n (%)		
Unable	84 (64.6)	
Assisted	20 (15.4)	
Independent	26 (20.0)	3 (100)
Highest level of expressive communication (n = 134[a]), n (%)		
Simple communication	39 (29.8)	
Complex gestures, concrete symbols, and vocalizations	68 (51.9)	2 (66.7)
Spoken language, signs, and abstract symbols	24 (18.3)	1 (33.3)
Prevalence of early concerns, n (%)		
Within 48 hours following birth		
Need assistance to breathe (n = 170)	8 (4.8)	0
Cried more than expected (n = 171)	22 (13.1)	0
Sensitive to noise (n = 169)	19 (11.5)	1 (33.3)
Did not like being touched or held (n = 170)	5 (3.0)	0
Felt floppy when moved or handled (n = 167)	29 (17.7)	1 (33.3)
Had difficulty sleeping (n = 171)	17 (10.1)	2 (66.7)
Difficult to feed (n = 169)	46 (27.7)	2 (66.7)
Slept for long periods of time (n = 168)	32 (19.4)	0
Gave strange stares or looks (n = 169)	16 (9.6)	0
First month following birth		
Need assistance to breathe (n = 168)	3 (1.8)	0
Cried more than expected (n = 169)	45 (27.1)	1 (33.3)
Sensitive to noise (n = 169)	45 (27.1)	1 (33.3)
Did not like being touched or held (n = 167)	9 (5.5)	0
Felt floppy when moved or handled (n = 169)	50 (30.1)	2 (66.7)
Had difficulty sleeping (n = 169)	42 (25.3)	2 (66.7)
Difficult to feed (n = 169)	40 (24.1)	1 (33.3)
Slept for long periods of time (n = 168)	45 (27.3)	0
Gave strange stares or looks (n = 168)	52 (31.5)	0

Abbreviations: aa = amino acid; AED = antiepileptic drug.
Percentages have been rounded and may not total to 100%.
[a] Limited to individuals at least 2 years of age at ascertainment.

categorized as simple communication including body language (e.g., twisting body or kicking), early sounds (e.g., crying, screaming, or grunting), facial expression (e.g., grimacing or frowning), and simple gestures (e.g., pushing away object or holding hands); complex gestures, concrete symbols, and vocalizations (e.g., shaking heads, rejecting/selecting drawings, or pointing at objects); or spoken language and abstract symbols such as signing "yes" or "no." Individuals of age below 2 years were excluded from the description of motor and communication skills to avoid scoring children on tasks too complex for their chronological age.

Standard protocol approvals, registrations, and patient consents. The study was approved by the University of Western Australia Human Research Ethics Committee (reference # RA/4/1/5024).

Statistical analysis. Descriptive statistics were used to summarize the characteristics of individuals in the study. The Kaplan-Meier method was used to estimate the probability of and median age at first seizure and the log-rank test was used to test for differences in age at seizure onset by mutation group. Fisher exact test of independence was used to evaluate whether the prevalence of early neonatal concerns varied with age at seizure onset, and Kruskal-Wallis equality-of-populations rank test to investigate the duration of honeymoon period by mutation group. Negative binomial regression was used to model the relationships between current seizure rate (expressed as count data) and mutation group, ever been treated with steroids/ACTH, an interaction term of both mutation group and ever been treated with steroids/ACTH, walking, and expressive communication skills. Crude estimates, in addition to those adjusted for potential confounders, such as sex, duration of epilepsy, presence of a honeymoon period, antiepileptic polytherapy, and use of ketogenic diet and VNS, were reported. Data were assumed missing at random and only complete data were analyzed in the model. All statistical analyses were conducted using Stata version 14 (StataCorp LP, College Station, TX).

RESULTS By January 2016, 192 individuals registered with the International CDKL5 Disorder Database had a pathogenic *CDKL5* mutation. The median age at ascertainment was 5 years 1 month (range 3 months–33 years 10 months) and 84.9% (n = 163) were female. Of these, 172 families had responded to the questionnaire section related to epilepsy and the characteristics of their child are shown in table 1. Three individuals never developed epilepsy and their clinical details are presented in table 2. For those with reported epilepsy (n = 169), the majority (87.0%) were female and about half (51.5%) were living in the United States. Nearly half (45.6%) were between 1.5 and 6 years of age at ascertainment. The most common mutation groups were truncating mutations between aa172 and aa781 (30.8%), followed by no functional protein (29.6%) and missense/in-frame mutations within catalytic domain (25.4%). In the children who developed epilepsy, difficulty feeding (27.7%, 46/166), prolonged sleeping time (19.4%, 32/165), and floppiness when moved or handled (17.7%, 29/164) were the most commonly reported early concerns within 48 hours of birth. These concerns were also noted by families/carers of the 3 children with no history of epilepsy. By 1 month of age, 31.5% (52/165) of the families whose children developed epilepsy reported that their child gave strange stares or looks. Just over a quarter (27.1%, 45/166) also reported that their children cried more than expected. The prevalence of floppiness had increased to 30.1% (50/166) while difficulty feeding was 24.1% (40/166). At ascertainment, the majority of children with epilepsy were unable to stand (54.2%, 71/131) or walk (64.6%, 84/130), and less than a third (29.8%, 39/131) had the simplest level of communication. On the other

Table 2 Clinical information on individuals who had never developed epilepsy or were seizure-free at time of ascertainment

Sex	Age at ascertainment, y	Nucleotide change	Amino acid change	Duration of seizure-free period, mo	Previous antiepileptic medications	Current antiepileptic medications	Walking for 10 steps	Highest expressive communication level
Never developed epilepsy (n = 3)								
Female	8.8	c.2684C>T	p.Pro895Leu[a]	NA	NA	NA	Independent	Complex gestures, concrete symbols, and vocalizations
Male	4.2	c.514G>A	p.Val172Ile[a]	NA	NA	NA	Independent	Complex gestures, concrete symbols, and vocalizations
Female	29.0	c.183delT	p.Met63Cysfs*13	NA	NA	NA	Independent	Spoken language, signs, and abstract symbols
Seizure-free (n = 12)								
Female	7.2	c.2504delC	p.Pro835Hisfs*2	6	TPM, VPA, others	ZNS	Assisted	Spoken language, signs, and abstract symbols
Female	6.3	c.978-?_2980+?del	p.(?)	65	VGB, TPM	None	Independent	Spoken language, signs, and abstract symbols
Male	1.0	c.2420_2430del	p.Ser807Cysfs*2	5	PB	TPM, LEV	Unable	Simple communication
Female	7.1	c.146-6T>G	p.(Glu49Valfs*2)	35	PB, LEV, TPM	None	Independent	Complex gestures, concrete symbols, and vocalizations
Female	20.4	c.-253-?_*1085del	p.(?)	204	VPA, CBZ, VGB	None	Unable	Spoken language, signs, and abstract symbols
Female	6.9	c.100-9_100-3delCCCTTGCinsGCAGA	p.(Lys33dup)	81	PB	None	Independent	Spoken language, signs, and abstract symbols
Female	1.8	c.602T>C	p.Leu201Pro	11	LEV, PB	VPA, OXC	Unable	Simple communication
Female	2.2	c.1581del	p.Thr528Profs*44	14	LEV, OXC, VGB, TPM	None	Unable	Complex gestures, concrete symbols, and vocalizations
Female	4.8	Deletion in exon 1	n/a	38	PB, VGB	VPA	Independent	Complex gestures, concrete symbols, and vocalizations
Female	19.7	c.1675C>T	p.Arg559*	120	LTG	None	Unable	Complex gestures, concrete symbols, and vocalizations
Female	11.1	c.1791delC	p.Tyr598Thrfs*18	24	PB, LEV, TPM, CBZ, CLN	LTG, DZ, LZ, GBP	Unable	Complex gestures, concrete symbols, and vocalizations
Female	4.3	c.2276+1G>A	p.(Val718_Trp759delinsGly)	44	LEV, PB, TPM, CLN, VPA	None	Assisted	Spoken language, signs, and abstract symbols

Abbreviations: CBZ = carbamazepine; CLN = clonazepam; DZ = diazepam; GBP = gabapentin; LEV = levetiracetam; LTG = lamotrigine; LZ = lorazepam; NA = not applicable; OXC = oxcarbazepine; PB = phenobarbital; TPM = topiramate; VGB = vigabatrin; VPA = valproate; ZNS = zonisamide.

[a] See in silico analyses of the missense variations presented in table e-5.

hand, more children without epilepsy were able to stand and walk independently, and communicate using complex gestures or spoken language (table 1).

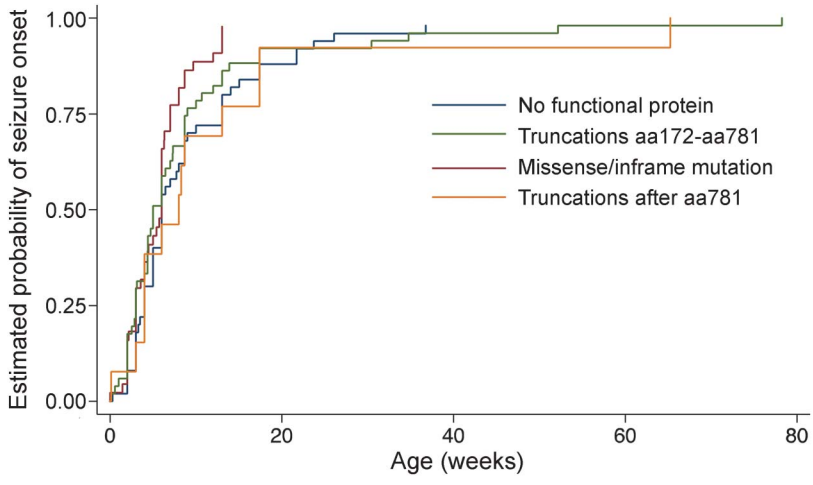
The median age at seizure onset was 6 weeks (range 1 week–1.5 years) and did not vary by mutation group (log-rank test χ^2 [3] = 4.44, p = 0.218) (figure 1). Around one-third (33.8%, n = 48) of the individuals with epilepsy were reported to have had infantile spasms (table 1). Seizures developing within the first month of life appeared to be associated with early concerns such as prolonged sleep and strange stares as reported over the same period (table 3). Of the 169 individuals with epilepsy, 163 (96.4%) families provided information on periods of seizure freedom, and 71 (43.6%) of these families reported their child had ever been seizure-free for more than 2 months. Of these, a honeymoon period with a median onset of 2 years (range 2 months–11 years) and a median duration of 6 months (range 2.5 months–6 years) was recorded for 52/70 (74%) and 59/70 (84%) individuals, respectively. Duration of honeymoon period did not vary by mutation group (Kruskal-Wallis equality-of-populations rank test χ^2 [3] = 3.673, p = 0.299). The age at seizure onset, age at ascertainment, and when available, age at the start and end of seizure-free period of the 169 individuals with epilepsy history are illustrated in figure e-1 at Neurology.org.

The median seizure rate was 2 per day (range 0–20 per day). The majority of children (56.2%) were reported to have, on average, 1 to 5 seizures per day. Around one-fifth (20.9%) had less than one seizure per day and a smaller group (15.0%) had more than 5 seizures a day. Fewer than one-tenth (7.8%, n = 12) of individuals had not experienced any seizures in the 6 months prior to ascertainment and their clinical details are presented in table 2. For the 30 individuals who were younger than 2 years, 10 children had no functional protein mutation and most (80%) had 1 to 5 seizures a day and 2 had monthly seizures. In those with missense/in-frame mutations within catalytic domain (n = 9), 8 individuals had current seizures (weekly n = 2, 1–5/day n = 5, >5/day n = 1) and 1 had no current seizures. All children with truncating mutations between aa172 and aa781 had current seizures (monthly n = 1, weekly n = 2, 1–5/day n = 4, >5/day n = 3). Only one individual had a truncating mutation after aa781 and no current seizure was reported.

In those aged 2 years and over, and after adjusting for walking ability and confounders, the seizure rate was lower in those with missense/in-frame mutations (incidence rate ratio [IRR] 0.60; 95% confidence interval [CI] 0.35–1.02) and truncating mutations between aa172 and aa781 (IRR 0.57; 95% CI 0.35–0.93) compared to individuals with no functional protein (table 4). Similarly, after controlling for communication level and confounders, the rate of seizures was lower (IRR 0.60; 95% CI, 0.36–0.90) in those with a truncating mutations between aa172 and aa781 when compared with those with no functional protein. Regarding functional abilities, there was evidence to suggest that being able to walk assisted or independently was associated with a lower rate of current seizures when compared to those unable to walk (IRR 0.62; 95% CI 0.39–0.99) after adjusting for genotype. Likewise, individuals who were able to use spoken language had a lower rate of seizures when compared to those who used simple communication skills (IRR 0.48; 95% CI 0.23–1.02) (table 4).

The top 10 AEDs, out of the 29 that were reported, are shown in table e-1. Clobazam was the most commonly used AED, followed by valproate, topiramate, levetiracetam, and vigabatrin. Ketogenic diet (24 responses) and VNS (26 responses) were also used. In the United States, topiramate and clonazepam were frequently prescribed, while valproate, vigabatrin, rufinamide, and zonisamide were more commonly used in countries outside the United States. Close to one-third of individuals with epilepsy (30.5%, n = 43) were previously treated with steroids/ACTH. At a median age at questionnaire completion of 5 years (n = 108, range 1–33 years), the seizure rate was higher in those treated with steroids compared with those who never received the treatment irrespective of adjusting or not for confounders (table e-2). The association appeared to be similar between those with and without a history of infantile spasms, and in those with infantile spasms mutation

Figure 1 **Estimated probability of seizure onset by mutation group among individuals with a pathogenic *CDKL5* mutation (n = 158)**



Kaplan-Meier estimates of probability of seizure onset by mutation group among individuals with a pathogenic *CDKL5* mutation. Twelve individuals in the genotype subcategory "mutation not grouped" were excluded from the analysis.

Table 3 Frequency distribution of early parental concerns within the first month following birth by seizure onset age in individuals with pathogenic *CDKL5* mutation and epilepsy (n = 169)

		Age at seizure onset		
	No.	≤1 mo, n (%)	>1 mo, n (%)	*p* Value[a]
Need assistance to breathe	163	1 (1.8)	2 (1.9)	1.000
Cried more than expected	165	13 (23.6)	32 (29.1)	0.578
Sensitive to noise	165	16 (29.1)	28 (25.5)	0.709
Did not like being touched or held	163	1 (1.9)	8 (7.3)	0.274
Felt floppy when moved or handled	164	20 (35.7)	29 (26.9)	0.281
Had difficulty sleeping	165	11 (20.0)	31 (28.2)	0.343
Difficult to feed	164	13 (24.1)	27 (24.6)	1.000
Slept for long periods of time	164	20 (36.4)	24 (22.0)	0.062
Gave strange stares or looks	163	24 (45.3)	27 (24.6)	0.011

[a] Two-sided Fisher exact test of independence.

group also affected the risk (table e-3). Among individuals who were diagnosed with infantile spasms (n = 41, median age at questionnaire completion 6 years, range 1–29 years), past treatment with steroids was associated with a more than 2-fold increase in current rate of seizure in those with missense/in-frame mutations within catalytic domain and truncations between aa172 and aa781 (table e-4). However, such relationship was not observed in those with non-functional protein mutations.

DISCUSSION Seizures are the most common clinical feature in the CDKL5 disorder. This article describes the onset and frequency of seizures in a unique, large sample of affected individuals. The size of our sample has provided novel information on relationships between seizure outcome and both genotype and functional abilities taking into account the influences of confounding variables. This study has characterized the features of the honeymoon period commonly observed in this disorder and demonstrated that, although uncommon, seizure freedom can be attained.

Six weeks was the median age at seizure onset,[3] although age at onset ranged from 1 to 78 weeks. Seizures occurred slightly earlier when there were other neonatal concerns but we could not identify any relationship between age at onset and genotype. However, compared to other mutation groups and after adjustment for functional ability, those individuals with no functional protein were more likely to have frequent seizures, the protective effect being greatest for those with a truncating mutation between aa172 and aa781. The underlying mechanism for these findings remains unclear, possibly representing a different trajectory for this group or perhaps capacity for greater responsiveness biologically to AEDs. We also found a reduction in seizure frequency in those with a truncation after aa781 but this reduction was less evident perhaps because of the smaller case numbers overall.

There has been varying reports of the short-term efficacy of steroids in CDKL5-related epilepsy and for childhood epilepsy in general.[14,15] Overall our results show that refractory epilepsy is not uncommon in CDKL5 disorder, and perhaps use of steroids was an indicator of difficult to treat epilepsy. Alternatively, AEDs can worsen seizures[16] and it is plausible that steroids could worsen seizures in specific CDKL5 genotypes but the modest numbers in our subgroup analyses on use of steroids need to be borne in mind. Further research on the role of steroids in the early and later stages of this disorder is needed, ideally in prospective trials.

Better functional abilities were also associated with less frequent seizures after accounting for mutation group, possibly reflecting better overall neurologic function. These relationships are, however, complex because we previously found that the mutation group with a truncation after aa781 was more likely to acquire developmental skills with better functional abilities.[13] Although the CDKL5 disorder is often labeled as an "epileptic encephalopathy defined by cognitive involvement due to ictal and/or inter-ictal activities," a recent multinational study considered it as a "genetic epilepsy with encephalopathy."[14] The debate on this matter continues. Of interest are 3 patients who never had seizures, which is unusual for the CDKL5 disorder. This raises the question of the pathogenicity of their *CDKL5* variations. Given that the p.Met63Cysfs*13 variation causes a frameshift mutation that completely disrupts the catalytic domain, it would be assumed to be a loss of function mutation. Regarding the 2 missense variations, in silico analyses suggest that they are both likely to be pathogenic (table e-5). Therefore, we reaffirm that epilepsy is not an absolute component of the CDKL5 disorder. We also notice that these individuals had better functional abilities. When these data are combined with the number of patients in our cohort who were seizure-free but with varying functional abilities, it raises the question of whether less severe epilepsy or better control of epilepsy allows for greater function or not.

A honeymoon period with seizure cessation for a period of time has previously been described in the CDKL5 disorder[10] and our study has further characterized its features. As reported by just over a third of families, this could last up to 72 months with a median duration of 6 months and, after adjustment for functional ability, was predictive of reduced seizure frequency in the longer term. We also catalogued the large number of AEDs used and identified

Table 4 Relationship between current seizure rate[a] and genotype, gross motor, and communication skills adjusting for pertinent confounders in individuals with pathogenic *CDKL5* mutation and epilepsy (n = 105[b])

	No.	Median seizure count (range)	Crude IRR (95% CI)	p Value	Adjusted[c] IRR (95% CI)	p Value	Adjusted[d] IRR (95% CI)	p Value
Mutation group								
No functional protein	33	3 (0–12)	Ref		Ref		Ref	
Missense/in-frame mutations	26	1 (0–20)	0.77 (0.45–1.31)	0.335	0.60 (0.35–1.02)	0.057	0.65 (0.38–1.11)	0.113
Truncations between aa172/aa781	36	2 (0–15)	0.70 (0.43–1.16)	0.166	0.57 (0.35–0.93)	0.025	0.60 (0.36–0.99)	0.044
Truncations after aa781	10	2 (0–7)	0.64 (0.30–1.38)	0.258	0.82 (0.39–1.70)	0.591	0.97 (0.45–2.11)	0.943
Walking for 10 steps								
Unable	67	2 (0–20)	Ref		Ref			
Assisted/independent	38	1 (0–12)	0.53 (0.34–0.81)	0.004	0.62 (0.39–0.99)	0.044		
Highest expressive communication								
Simple communication	29	3 (0–10)	Ref				Ref	
Complex communication	58	2 (0–20)	0.65 (0.42–1.01)	0.058			0.73 (0.46–1.15)	0.174
Spoken language	18	1 (0–5)	0.38 (0.20–0.72)	0.003			0.48 (0.23–1.02)	0.055
Sex								
Female	96	2 (0–20)	Ref		Ref		Ref	
Male	9	4 (0–8)	1.51 (0.75–3.02)	0.246	1.22 (0.64–2.34)	0.542	1.12 (0.57–2.17)	0.745
Duration of epilepsy, y	105	2 (0–20)	0.98 (0.94–1.02)	0.272	0.98 (0.94–1.02)	0.277	0.98 (0.94–1.02)	0.293
Ever had a honeymoon period								
No	63	2 (0–20)	Ref		Ref		Ref	
Yes	42	2 (0–8)	0.67 (0.44–1.02)	0.064	0.62 (0.40–0.94)	0.025	0.62 (0.40–0.95)	0.029
Current number of antiepileptic drugs used								
None/monotherapy	29	0 (0–12)	Ref		Ref		Ref	
Polytherapy	76	2 (0–20)	1.68 (1.05–2.69)	0.031	1.63 (0.98–2.72)	0.062	1.68 (1.01–2.79)	0.046
Current use of ketogenic diet								
No	88	2 (0–20)	Ref		Ref		Ref	
Yes	17	3 (0–8)	0.97 (0.55–1.69)	0.903	0.91 (0.54–1.54)	0.724	0.88 (0.51–1.51)	0.648
Current use of VNS								
No	84	2 (0–20)	Ref		Ref		Ref	
Yes	21	2 (0–12)	1.06 (0.64–1.77)	0.815	1.27 (0.77–2.09)	0.358	1.37 (0.81–2.29)	0.237

Abbreviations: aa = amino acid; CI = confidence interval; IRR = incidence rate ratio; Ref = reference level; VNS = vagal nerve stimulation.
[a] Average number of seizures per day.
[b] Excludes individuals younger than 2 years at ascertainment or with ungrouped mutation. Also, only those with data in both independent and dependent variables were included.
[c] Adjusted for all independent variables except highest expressive communication.
[d] Adjusted for all independent variables except walking for 10 steps.

some differences in their use in the United States compared with elsewhere.

The majority of studies to date relating to epilepsy in the CDKL5 disorder have been clinical studies with small case numbers of less than 10 patients.[4–12] The largest 2 have been an early French case series,[10] and a recent multinational study with 39 patient contributions from 27 clinicians in 11 countries.[14] The first described the clinical and EEG trajectory of epilepsy in 12 patients whose median age at onset was 4 weeks, ranging from 1 to 10 weeks.[10] Over half of the 12 patients were seizure-free, in contrast to a much lower percentage in our more statistically powerful study, and any genotype–phenotype correlations were only based on 6 patients. The second study collated the experiences of 39 patients with the CDKL5 disorder with respect to response of seizure frequency to AEDs including the ketogenic diet.[14] In line with our results, only 2 of 39 patients remained seizure-free after 12 months follow-up but no genotype data were presented.

Large sample sizes are mandatory for investigation of genotype and other relationships in rare disorders especially in the case of CDKL5 where, unlike Rett

syndrome,[17] the majority of mutations are unique. Our current categorization of mutations was based on predicted structural and functional consequences,[13] but it may not be optimal. Although our database comprises the largest known data collection on the CDKL5 disorder, there may still be some concerns about parent report, but such data are well-accepted in clinical practice and in clinical trials.[14] However, our study is likely skewed towards families of higher socioeconomic status as genetic testing is required for diagnosis.

This observational study adds to the understanding of epilepsy in the CDKL5 disorder. Investigating factors that may predict either better or worse outcomes is essential in our quest to improve management and quality of life for these patients and their families.

AUTHOR CONTRIBUTIONS

Stephanie Fehr: study concept and design, acquisition of data, critical revision of manuscript for intellectual content. Kingsley Wong: analysis and interpretation of data, statistical analysis, drafting/critical revision of manuscript for intellectual content. Richard Chin: analysis and interpretation of data, drafting/critical revision of manuscript for intellectual content. Simon Williams: critical revision of manuscript for intellectual content. Nick de Klerk: analysis and interpretation of data, critical revision of manuscript for intellectual content. David Forbes: critical revision of manuscript for intellectual content. Rahul Krishnaraj: critical revision of manuscript for intellectual content. John Christodoulou: critical revision of manuscript for intellectual content. Jenny Downs: study concept and design, acquisition of data, analysis and interpretation of data, drafting/critical revision of manuscript for intellectual content, study supervision. Helen Leonard: study concept and design, acquisition of data, analysis and interpretation of data, drafting/critical revision of manuscript for intellectual content, study supervision.

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DISCLOSURE

The authors report no disclosures relevant to the manuscript. Go to Neurology.org for full disclosures.

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