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ARTICLE

CACNA1A haploinsufficiency causes cognitive impairment, autism and epileptic encephalopathy with mild cerebellar symptoms

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***CACNA1A* loss-of-function mutations classically present as episodic ataxia type 2 (EA2), with brief episodes of ataxia and nystagmus, or with progressive spinocerebellar ataxia (SCA6). A minority of patients carrying *CACNA1A* mutations develops epilepsy. Non-motor symptoms associated with these mutations are often overlooked. In this study, we report 16 affected individuals from four unrelated families presenting with a spectrum of cognitive impairment including intellectual deficiency, executive dysfunction, ADHD and/or autism, as well as childhood-onset epileptic encephalopathy with refractory absence epilepsy, febrile seizures, downbeat nystagmus and episodic ataxia. Sequencing revealed one *CACNA1A* gene deletion, two deleterious *CACNA1A* point mutations including one known stop-gain and one new frameshift variant and a new splice-site variant. This report illustrates the phenotypic heterogeneity of *CACNA1A* loss-of-function mutations and stresses the cognitive and epileptic manifestations caused by the loss of $Ca_V2.1$ channels function, presumably affecting cerebellar, cortical and limbic networks.**
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INTRODUCTION

The *CACNA1A* gene on chromosome 19p13 encodes the alpha1 subunit of the $Ca_V2.1$ P/Q-type voltage-gated calcium channel. Mutations in this gene cause three allelic autosomal dominant conditions: episodic ataxia type 2 (EA2, OMIM: 108500), spinocerebellar ataxia type 6 (SCA6, OMIM: 183086) and familial hemiplegic migraine type 1 (FHM1, OMIM:141500).[1–3] These conditions occasionally overlap since patients with FHM1 may have cerebellar symptoms, and 33% of patients with SCA6 display episodic features characteristic of EA2.[4–6] EA2 usually presents during childhood or early adulthood[7] with intermittent episodes of ataxia and nystagmus lasting minutes to days.[8] These episodes are classically triggered by exertion, stress, heat, fever, alcohol, caffeine or drugs such as phenytoin.[9] They tend to respond to acetazolamide or 4-aminopyridine (4-AP). A downbeat nystagmus often persists between episodes.[10] Most patients eventually develop progressive ataxia with cerebellar atrophy.[7] A minority of patients present generalized absence epilepsy[11] and/or learning difficulties,[5] but the extent of neurocognitive impairment associated with these mutations has not been fully described to date.

We describe four French Canadian non-consanguineous families with 16 affected individuals carrying *CACNA1A* loss-of-function mutations and presenting with epileptic encephalopathy or cognitive impairment including intellectual disability (ID), ADHD or autism, as well as downbeat nystagmus and intermittent ataxia, which did not dominate the clinical picture.

METHODS

All patients were investigated on a clinical basis at the CHU Ste-Justine. Informed consent was obtained for genetic testing in accordance with the institution's ethics committee board requirements. Comparative genomic hybridization (CGH) assays were conducted at the CHUSJ using a 135k-feature whole-genome microarray (SignatureChip OS2.0 manufactured for Signature Genomic Laboratories (Spokane, WA, USA) by Roche NimbleGen, Madison, WI, USA; based on UCSC 2006 hg18 assembly), or at the CHUS using a 180k-feature whole-genome microarray (Cytosure ISCAv2, 4x180k, Oxford Gene Technology, Becgbroke, UK), according to the manufacturers' protocols. Genomic coordinates indicate the minimal size of the CNVs. *CACNA1A* sequencing of the entire coding region (47 exons) and flanking exon–intron splice-site junctions was performed at Athena Diagnostics or at Medical Neurogenetics Laboratories. Variants identified in the course of this project were submitted to the ClinVar database (http://www.ncbi.nlm.nih.gov/clinvar/) (#SCV000196749–SCV000196752).

RESULTS

Clinical descriptions and investigations

The clinical presentation and investigations of patients recruited in this study are summarized in Table 1. Family trees are illustrated in Figure 1. Mutations are depicted in Figure 2. Detailed description for each case is provided in the following sections.

Family 1

Patient 4.1 was born at term after an unremarkable pregnancy and had normal early psychomotor development before seizure onset. He developed refractory epilepsy with generalized tonic-clonic seizures and absences starting at 11 months of age. His electroencephalograms (EEGs) were characterized by intermittent generalized theta activity with spikes or with intermittent bi-posterior spike-wave activity. The seizures were successively treated with a combination of valproic acid, topiramate and levetiracetam. He also presented with monthly

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Table 1 Patient clinical description and investigations

				Cerebellar symptoms				
ID	*Age at onset*	*Seizures*	*Neurocognitive symptoms*	*Acute*	*Chronic*	*EEG*	*MRI*	*Genetic*
Family 1								
3.2	26 Y	Typical FS	Learning difficulties ADHD	A+Ny	A+Ny			*Idem*
3.3	Adult	Typical FS	Learning difficulties ADHD	A+Ny	A+Ny			
3.4	4 Y	Typical FS	None	A	None			
4.1	11 Mo	GTCS, absences	GDD moderate-severe ID, global IQ: 47 atypical social interactions	A+Ny	A+Ny	Diffuse slowing, generalized theta bursts interspersed with spikes, bi-occipital spikes	Hippocampal asymmetry (L>R)	Stop-gain mutation in exon 23; c.3832C>T (p.Arg1278Ter; NM_001127221.1; rs121909323)
4.2	18 Mo	Typical FS	GDD ASD ADHD Global IQ: 78	Ny	Ny	N	N	*Idem*
4.3	20 Mo	None	GDD ASD ADHD	Ny	Ny	N	N	*Idem*
Family 2								
1.1	10 Y	None	N/A	A+Ny	A+Ny			
3.1	12 Y	None	None	A+Ny	A+Ny			*Idem*
3.2	11 Y	None	Learning difficulties, mild ID	A+Ny	A+Ny			Frameshift mutation in exon 19; c.2867_2869del (p.Arg957fs; NM_001174080.1)
4.1	18 Mo	Absences	Learning difficulties Mild ID ADHD	A+Ny	A+Ny, dysarthria	3 Hz SW	N	*Idem*
4.2	6 Mo	Atypical FS	ADHD Learning difficulties Memory difficulties Global IQ: 71	A+Ny	A+Ny		N	*Idem*
Family 3								
3.1	20 Mo	Typical FS	GDD ADHD Dyslexia Mild ID Global IQ: 72	A+Ny	A+Ny		T2 Hyperintensity of L globus pallidus	Splice-site mutation exon 4 c.868+5G>A (NG_011569.1)
2.1	6 Y	None	Learning difficulties ADHD	A+Ny	A+Ny	N	N	*Idem*

Table 1 (Continued)

ID	Age at onset	Seizures	Neurocognitive symptoms	Cerebellar symptoms: Acute	Cerebellar symptoms: Chronic	EEG	MRI	Genetic
Family 4								
2.1	6 Y	None	Learning difficulties ADHD Suspected mild ID	A+Ny	A+Ny		N	Chr19.hg18: g.13,380,344_ 13,465,506del (Del19p13.13 within the *CACNA1A* gene)
3.1	8 Mo	GTCS, focal seizures	GDD ASD ADHD	A+Ny	A	N	N	*Idem*
3.2	15 Mo	None	GDD Mild-moderate ID Global IQ: 58 ADHD	A+Ny	Ny	N	N	*Idem*

Abbreviations: A, ataxia; ADHD, attention-deficit hyperactivity disorder; ASD, autism spectrum disorder; EEG, electroencephalogram; FS, febrile seizure; GDD, global developmental delay; GTCS, generalized tonic-clonic seizures; ID, intellectual deficiency; IQ, intellectual quotient; Mo, months; MRI, magnetic resonance imaging; N, normal; N/A, not available; Ny, nystagmus; SW, spike and wave; Y, years of age.

episodes of malaise, altered contact, hypotonia and nystagmus lasting 2–3 min, which did not respond to anticonvulsive drugs. The child's development was globally delayed, with significant language impairment at 18 months of age. At 6 years of age, he displayed a moderate-severe ID (global IQ: 47) with altered socialization skills. Upon examination, he presents a downbeat nystagmus and an unsteady broad-based gait, without frank dysmetria.

The child's father (3.2) presented with febrile seizures and learning disabilities with ADHD during childhood but completed his high school degree and is now employed. Upon questioning, he described intermittent episodes of ataxia, vertigo, vertical oscillopsia and dysarthria lasting 15–60 minutes, occurring every week, and triggered by stress, infections, fatigue and sweet foods, since the age of 26 years. These episodes had been attributed to hormonal imbalances although he had never consulted a neurologist for these symptoms. Interestingly, his twin brother (patient 3.3) presented similar episodic symptoms and suffered from impaired socialization skills, had learning disabilities, required special schooling and is currently unemployed. The twins' father (2.3) presents episodic ataxia and persistent nystagmus. Their brother (patient 3.4) presented febrile seizures during childhood. He displays infrequent (two to three times a year) episodes of ataxia lasting less than an hour and triggered by fatigue or exertion. This man has two children, a girl (patient 4.2) and a boy (patient 4.3), who both presented with global developmental delay (DD), autism and ADHD. Both children had single episodes of behavioral arrest with facial automatisms and had normal EEGs. On examination, both children display a downbeat nystagmus (see Supplementary Video S1) and gaze-evoked nystagmus without ataxia or dysmetria.

CGH and metabolic investigations were normal in all three children (including lactate, pyruvate, amino acid screen, organic acid screen, urinary purines and pyrimidines and urine creatine). Patient 4.3 underwent full ophtalmological workup, with normal electroretinogram. His brain magnetic resonance imaging (MRI) revealed slight size asymmetry of the hippocampi without structural anomalies, and normal cerebellar and brainstem structures. Brain MRIs of the two other children were normal. Targeted sequencing of the *CACNA1A* gene in all three children revealed a variant in exon 23; c.3832C>T (p.(Arg1278Ter; NM_001127221.1; NG_011569.1; rs121909323)) leading to a premature stop codon and previously associated with EA2.[12,13] Additional sequencing of 35 known epileptic encephalopathy genes in patient 4.1 as described by Michaud *et al*,[14] was performed and was negative.

Family 2

Patient 4.1 was born at term after an unremarkable pregnancy. He presented with global DD evolving toward mild ID, ADHD and learning difficulties. At 18 months of age, he developed refractory absences with daily episodes of brief behavioral arrests and facial automatisms. His EEGs reveal generalized spike-wave activity. His absences were refractory to valproic acid and ethosuximide but partially responsive to a combination of valproic acid and levetiracetam. In addition, this boy presents recurrent episodes of nystagmus without ataxia, lasting 5–15 s, occurring two to three times a day, often precipitated by fatigue or febrile illnesses. At 9 years of age, he remains with moderate global delay, delayed fine motor skills and he requires special education. His neurological examination reveals a familial macrocrania (98th percentile), downbeat nystagmus, hypometric saccades, slight dysarthria and unsteady gait without frank ataxia.

Upon questioning, the child's mother (patient 3.1) described episodes of ataxia, vertigo and nausea, without headache, lasting

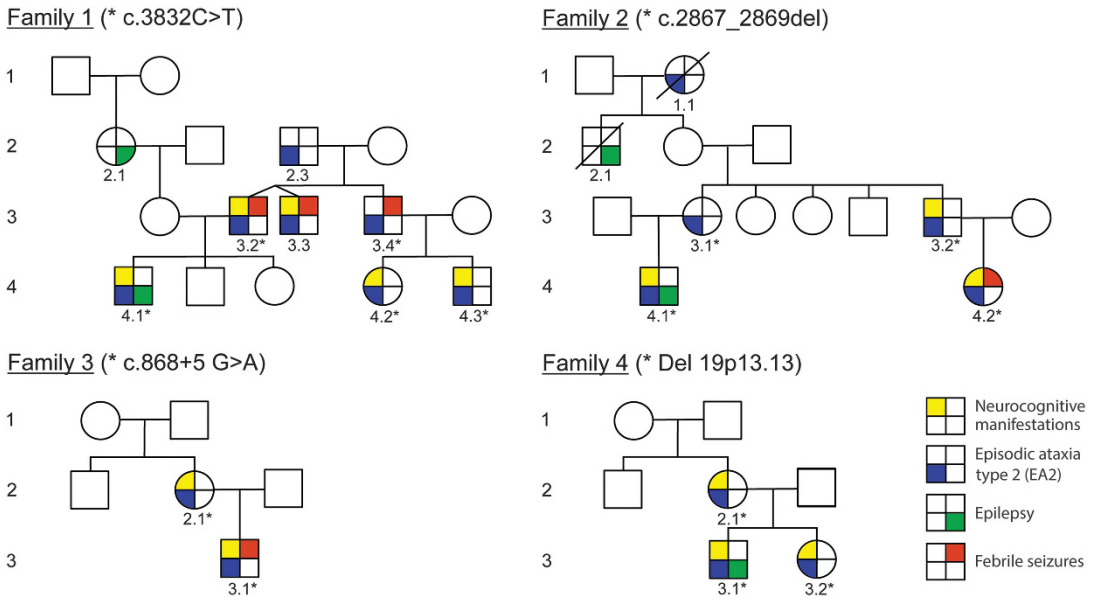

Figure 1 Family trees. The genealogical tree for each family is illustrated, with color-coding for associated symptoms (cf inset). Asterisks denote patients for which *CACNA1A* mutations were confirmed.

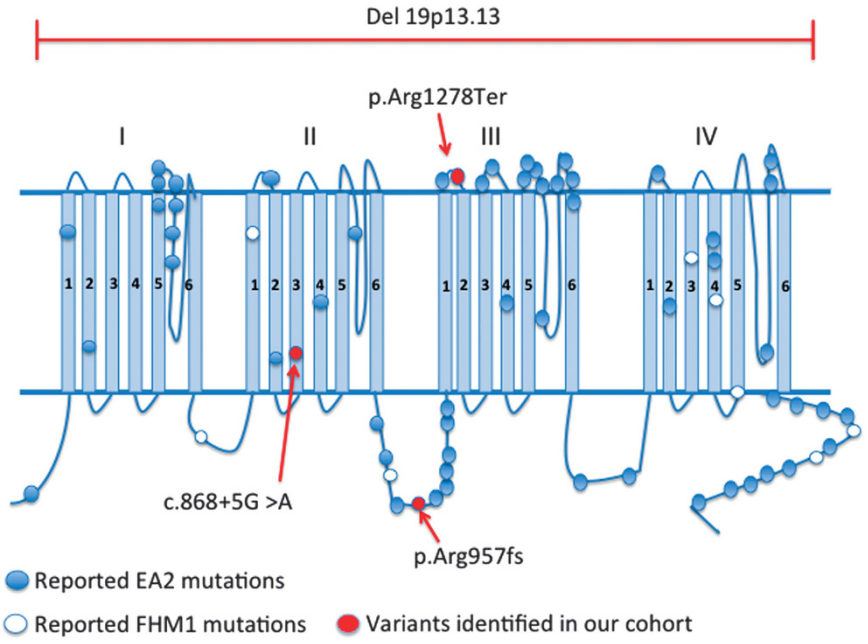

Figure 2 New and previously reported mutations in $Ca_V2.1$. Previously reported mutations in *CACNA1A*, causing either episodic ataxia type II (EA2; blue circles) or familial hemiplegic migraine type I (FHM1; white circles) affect most domains of the alpha1 subunit of the $Ca_V2.1$ calcium channel, as illustrated here (adapted from Mantuano *et al*, 2010). Red circles illustrate loss-of-function mutations reported in this publication.

1–2 h and occurring multiple times a week since the age of 12, for which she had never consulted. Her episodes resolved with acetazolamide. Her examination reveals downbeat nystagmus, without dysmetria or ataxia. Her maternal grandmother (patient 1.1) had presented similar episodes of ataxia with nystagmus for which she never sought medical attention. She had lost a son (patient 2.1) at 18 months of age of severe refractory epilepsy.

The child's maternal uncle (patient 3.2) presents episodes of intermittent ataxia, nystagmus and oscillopsia, lasting 10–15 min, occurring multiple times per week, precipitated by stress, emotion and exertion, since the age of 11. These events respond partially to acetazolamide. He also presented episodes of stress-induced cataplexy, without myokymia or myotonia. He had mild ID and required special schooling. He is currently unemployed. He developed a progressive ataxia with dysarthria in his forties. His neurological examination reveals dysmetria, gait ataxia and diffuse hyporeflexia.

This man has a 13-year-old daughter (patient 4.2), born at term from an uneventful pregnancy. At 6 months of age, she developed episodes of vertigo and ataxia with vomiting. She learned to walk at 16 months of age and was ataxic from the onset. She remained with paroxysmal episodes of ataxia, nytagmus and malaise lasting 1 h, recurring daily, precipitated by exertion or fatigue, sometimes

accompanied by headaches. These episodes are partially responsive to acetazolamide. Between episodes, she had progressive gait ataxia and downbeat nystagmus with vertical oscillopsia. She presented recurrent generalized febrile seizures from 15 months to 9 years of age. In addition, she had cognitive deficits with learning disabilities, ADHD and behavioral difficulties and required special schooling. Her global IQ was evaluated at 71 on WISC-IV testing. Her neurological examination is remarkable for downbeat nystagmus, truncal ataxia, ataxic gait and diffuse hyporeflexia.

The metabolic workup and CGH assay for both children as well as brain MRIs were unremarkable. *CACNA1A* sequencing revealed a new frameshift variant in exon 19, c.2867_2869del (p.(Arg957fs; NM_001174080.1; NG_011569.1)).

Family 3

Patient 3.1, now 16 years old, was born prematurely at 34 weeks of gestation from an unremarkable delivery. He presented with DD and unsteady gait since he learned to walk at 2 years of age. Around 4 years of age, he developed daily episodes of oscillopsia and ataxia leading to falls, lasting 15–30 min, triggered by exertion or stress. His episodes responded completely to acetazolamide. He remained with intercurrent nystagmus and unsteady gait. He had one brief generalized tonic-clonic febrile seizure before age 4. He had mild ID (global IQ: 72), ADHD, dyslexia and required special schooling. His neurological examination revealed downbeat nystagmus, horizontal gaze-evoked nystagmus, saccadic visual pursuit, hypermetric saccades, limb hypotonia, unsteady gait during rapid direction changes and no dysmetria.

The boy's mother (patient 2.1) aged 50 years presented during childhood with learning difficulties requiring special schooling, ADHD and dropped out of school after grade 9. She is currently unemployed. Since 6 years of age, she presented daily episodes of unsteady gait, nystagmus, malaise lasting 2–3 h, precipitated by stress, fatigue or exertion. These had been wrongly attributed to anxiety for which she received benzodiazepines, but responded completely to acetazolamide. She developed a progressive gait ataxia, mild dysarthria and persistent nystagmus, which became symptomatic at the age of 42 years. Her neurological examination revealed downbeat nystagmus and horizontal gaze-evoked nystagmus, hypermetric saccades, mild dysarthria, a gait ataxia and hand clumsiness without dysmetria.

The boy's investigation included a normal karyotype, fragile X screen and metabolic workup. His brain MRI revealed a non-specific millimetric T2 signal hyperintensity in the left globus pallidus. *CACNA1A* sequencing in both individuals revealed a new splice-site variant, c.868+5G>A (NG_011569.1; NM_000068.3), predicted to be pathogenic by abolishing a known splice site for exon 4. Deletion of this conserved exon leads to a clear loss-of-function in mice models of the disease.[15,16]

Family 4

Patient 3.1 was born at 36 weeks after an uneventful pregnancy. At 8 months of age, he presented with recurrent afebrile episodes of altered consciousness with eyeball revulsion lasting 10–20 s. He also presented three brief generalized tonic-clonic seizures. His EEG and brain MRI were unremarkable. He was treated with carbamazepine, which was replaced by lamotrigine when seizures recurred. At 12 months of age, the patient developed recurring episodes of ataxia and nystagmus, lasting 12–24 h, during which he laid on the ground and refused to stand. Acetazolamide treatment reduced the duration of these episodes, although they persist at a frequency of two to three times per month at 2.5 years of age. The episodes are often accompanied by headache and vomiting, and sometimes require hospitalization for analgesia and rehydration. The boy displayed a global DD, walked independently at 24 months and is clumsy on fine motor skills. He spoke his first intelligible words at 26 months. At 2.5 years of age, his vocabulary was restricted to six words and he could not juxtapose words. He was socially awkward, does not point, drags the adults toward objects of interest, has stereotypic and restricted interests, was impulsive and inattentive and his ADOS confirmed ASD. His examination at 2.5 years of age revealed emotional lability, poor eye contact, bilateral epicanthal folds, mild limb hypotonia, truncal ataxia, unsteady gait with wide base and no dysmetria.

His sister (patient 3.2) was born at term after an uneventful pregnancy. She presented with global DD: she walked at 15 months, had poor fine motor skills, spoke her first words at 15 months but sentences only at 4 years of age. She had mild-moderate ID (global IQ: 58 on WISC-IV testing), ADHD and anxiety, and she required special schooling. She developed episodic ataxia at 15 months of age, with episodes lasting 1 h, every 2 weeks, triggered by fever, infections or vestibular stimulation (swing). These events improved with acetazolamide. She also reported tension headaches once a month. Her examination at 11.5 years of age revealed downbeat nystagmus, mild dysarthria, discrete dysmetria, clumsiness on rapidly alternating movements and inability to perform repetitive motor sequences (perseveration and impulsivity), but no ataxia. Her brain MRI and EEG were unremarkable. A complete metabolic workup at 15 months of age was negative.

The mother (patient 2.1), now aged 33, had a normal early development but presented significant learning disabilities with ADHD, required special schooling from the first grade onwards and dropped out of school after 10 years. She is currently unemployed. Her cognitive abilities were not formally tested but she was reported to have mild ID. She developed episodic ataxia at 21 years of age, with episodes lasting 2–3 h, recurring once a month, triggered by stress and exercise. She also developed progressive interictal gait instability since the age of 32 years and falls frequently. She reported migraines on a monthly basis. Her brain MRI was unremarkable. Her physical examination revealed executive slowing, a downbeat nystagmus, mild dysarthria, bilateral clumsiness on rapidly alternating movements, no frank dysmetria, no frank ataxia but gait instability on rapid direction changes and unstable tandem walking.

CGH in both children revealed a maternally inherited 0.085 Mb deletion on chromosome 19: Chr19.hg18:g.13,380,344_13,465,506del (hg18/NCBI36; NG_011569.1; Del19p13.13), within the *CACNA1A* gene and spanning most of the gene.

DISCUSSION

We reported 16 affected individuals from four non-consanguineous families carrying *CACNA1A* loss-of-function variants. Although all of our patients displayed mild intermittent cerebellar symptoms, their most striking features were the cognitive or behavioral impairments and seizure susceptibility that accompanied their disorder. This report therefore stresses the significant non-cerebellar symptoms associated with *CACNA1A* haploinsufficiency.

The initial presentation in our patients were diverse and included epileptic encephalopathy with generalized epilepsy, DD with febrile seizures, DD with autism spectrum disorder (ASD) or learning disabilities with episodic ataxia. The acute cerebellar symptoms displayed by our patients were either an isolated downbeat nystagmus ($n=2/16$, 13%) or episodes of intermittent ataxia, oscillopsia and nystagmus typical of EA2. These episodes vary in intensity from a few seconds to hours, are typically provoked by stress, exertion, fatigue or illness and vary in frequency between individuals, from weekly

episodes to rare annual episodes. Most patients ($n=12/16$, 75%) developed progressive ataxia during adolescence or adulthood and the majority present interictal downbeat nystagmus ($n=14/16$, 88%). This is consistent with the current literature describing progressive ataxia in 80% of patients with EA2[7] and persistent nystagmus in 90% of patients.[10] Nonetheless, in six individuals, the ataxia did not dominate the initial presentation and was only revealed by close questioning of relatives for whom the cerebellar symptoms had been misdiagnosed as anxiety or hormonal imbalance.

A significant proportion of our patients presented with DD ($n=6/16$, 38%), ID ($n=6/16$, 38%) or learning difficulties ($n=4/16$, 25%), often with ADHD and impulsivity ($n=11/16$, 69%). These deficits were apparent from early childhood and had significant impact on the patient's educational path and social integration. In addition, three children carry a diagnosis of ASD and another has altered social skills with stereotypic behaviors suggestive of ASD ($n=4/16$, 25%). Cognitive and behavioral impairments have been reported in a minority of patients with *CACNA1A* mutations, including in two children with sporadic epileptic encephalopathies,[17,18] in six children with EA2 and ID,[19–22] including two siblings with a 19p13.13 deletion.[23] Rare cases of FHM1 with ID[24] and progressive cognitive decline[24–26] have been reported. Therefore, our data together with previous reports suggest that children with global DD, ID or ASD with mild cerebellar symptoms, with or without a family history suggestive of EA2, should be investigated for loss-of-function mutations in the *CACNA1A* gene.

In addition, the patients described here presented a high rate of epilepsy or febrile seizures. Indeed, three children from three unrelated families presented with epileptic encephalopathy with either generalized absence seizures or focal seizures with or without generalized tonic-clonic seizures ($n=3/16$, 19%). In addition, six patients ($n=6/15$, 40%) had febrile seizures during childhood. *CACNA1A* loss-of-function mutations have been associated with rare cases of generalized absence epilepsy[22,27–32] or epileptic encephalopathy.[17,18] Furthermore, up to 7% of patients with EA2 were described to develop absence epilepsy, as reviewed by Rajakulendran *et al*.[11] The current report illustrates the higher rate of febrile seizures or epilepsy in patients with loss-of-function mutations in *CACNA1A*, which might represent only a fraction of EA2 cases. Perhaps most importantly, our report indicates that mutations in *CACNA1A* should be excluded in children with developmental disorders and refractory generalized epilepsy even in the absence of frank cerebellar symptoms. A downbeat nystagmus was observed in most of our patients and might alert clinicians to this genetic condition in children with refractory epilepsy.[33–39]

Mechanistically, $Ca_V2.1$ channels are voltage-gated calcium channels expressed at the pre-synaptic and somatodendritic level of a variety of cerebral and spinal neuronal populations.[15,40,41] $Ca_V2.1$ channels have been shown to mediate synaptic release from a variety of neuronal cell types, both excitatory and inhibitory, in the cortex, hippocampus, thalamus and cerebellum.[15,42–49] The loss of $Ca_V2.1$ channels is compensated by upregulation of other voltage-gated calcium channels at most central synapses,[15,44,47–49] although with different efficiency,[50] resulting in synaptic dysfunction of particular cell types leading to pathological manifestations. In cerebellar networks, $Ca_V2.1$ channels regulate the whole-cell calcium current density and the intrinsic excitability of Purkinje cells and granule cells,[44,51–53] and exert major control over glutamate release at the parallel fiber onto Purkinje-cell synapses.[50,54–56] Furthermore, targeted deletions of *Cacna1a* in cerebellar granule cells[57] or in Purkinje cells[58] result in altered cerebellar output by respectively decreasing the excitatory drive on Purkinje cells or their ability to release neurotransmitters, causing ataxia and dyskinesia in mice.

Within the thalamus, constitutive dysfunction of $Ca_V2.1$ channels in the *Cacna1a*$^{tg/tg}$ mutant mice result in a gain of function of Cav3.1 T-type calcium channels in the reticular nucleus and in a persistent tonic thalamic $GABA_A$ current in thalamocortical projection neurons, which together enhance thalamocortical excitation and contribute to the spike-wave absence seizures phenotype.[59–63] In the neocortex and hippocampus, $Ca_V2.1$ channels have been demonstrated to mediate GABA release and synaptic efficiency from cortical GABAergic parvalbumin-positive fast-spiking interneurons (FS-INs)[15,42,43,47] as well as from cortical pyramidal cells.[15] We recently demonstrated that a selective deletion of *Cacna1a* from cortical and hippocampal GABAergic interneurons, while sparing the thalamus and cerebellum, selectively impairs GABA release from FS-INs, despite an upregulation of N-type channels, and that this is sufficient to cause generalized epilepsy in conditional mutant mice.[15] Of note, the selective deletion of *Cacna1a* in cortical pyramidal cells did not cause seizures but its combination with forebrain GABAergic interneuron *Cacna1a* deletion reduced seizure severity. These studies revealed the importance of $Ca_V2.1$ channels in regulating synaptic release from cortical FS-INs and the potential involvement of these cell types in *Cacna1a*-associated epilepsy.

The mechanisms underlying cognitive dysfunction in *Cacna1a* mutants are uncertain. Progressive cognitive deficits have been reported in the heterozygous leaner mutant mice *Cacna1a*$^{tg(la)/+}$ [64,65] and in the heterozygous Nogoya mutant mice.[66] The cerebellum projects directly and indirectly to many cortical and limbic structures involved in learning and cognition,[67–69] and deregulation of these projections might impair cortical and limbic processes, including motor memory consolidation.[70] In addition, cortical inhibitory defects have been postulated to result in a variety of neurobehavioral phenotypes in humans and rodents, including cognitive dysfunction, social deficits and autism,[71–76] and could contribute to cognitive impairment following *Cacna1a* mutations. Indeed, cortical and limbic GABAergic interneurons regulate the synchrony of neuronal firing in populations of neurons and participate in the generation of high-frequency gamma oscillations involved in cognitive processes and attention.[77–86]

In summary, the current report illustrates the spectrum of neurobehavioral symptoms associated with *CACNA1A* loss-of-function mutations in humans. Such behavioral phenotypes might be overlooked in patients when the epilepsy or ataxia are predominant, but a careful consideration of potential cognitive and behavioral consequences in these patients might allow for an earlier instauration of cognitive-behavioral interventions and improve long-term outcome. Furthermore, we propose that targeted sequencing of the *CACNA1A* gene should be considered in children presenting with downbeat nystagmus together with epileptic encephalopathy, cognitive impairment or ASD.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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