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# Role of voltage-gated calcium channels in epilepsy

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## Abstract

It is well established that idiopathic generalized epilepsies (IGEs) show a polygenic origin and may arise from dysfunction of various types of voltage- and ligand-gated ion channels. There is an increasing body of literature implicating both high and low voltage-activated (HVA and LVA) calcium channels and their ancillary subunits in IGEs. $Ca_v2.1$ (P/Q-type) calcium channels control synaptic transmission at presynaptic nerve terminals, and mutations in the gene encoding the $Ca_v2.1$ α1 subunit (*CACNA1A*) have been linked to absence seizures in both humans and rodents. Similarly, mutations and loss of function mutations in ancillary HVA calcium channel subunits known to coassemble with $Ca_v2.1$ result in IGE phenotypes in mice. It is important to note that in all these mouse models with mutations in HVA subunits there is a compensatory increase in thalamic LVA currents, which likely leads to the seizure phenotype. In fact, gain of function mutations have been identified in $Ca_v3.2$ (an LVA or T-type calcium channel encoded by the *CACNA1H* gene) in patients with congenital forms of IGEs, consistent with increased excitability of neurons as a result of enhanced T-type channel function. Here we provide a broad overview of the roles of voltage-gated calcium channels, their mutations, and how they might contribute to the river that terminates in epilepsy.

### Keywords

calcium channel; P/Q-type channels; T-type channels; epilepsy; seizures

## Introduction

Epilepsy is a disorder that results from abnormal hyperexcitable and hypersynchronous activity of neurons [28]. Epileptic seizures can have a wide variety of origins, including brain damage and genetic causes [85]. Broadly, seizures can be classified into focal and generalized seizures. Focal seizures are typically localized to one brain hemisphere and may arise from insults such as localized brain lesions, or tumors. In contrast, generalized epilepsies are characterized by seizure activity over both brain hemispheres. They can be further classified into symptomatic and idiopathic epilepsy. Symptomatic generalized seizures can be caused by insults such as brain infection or oxygen deprivation. Idiopathic seizures often do not have a clear etiology, but may often include a genetic component [10, 59]. One of the key defining attributes of idiopathic generalized epilepsies (IGEs) is the

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occurrence of absence seizures [22]. Absence seizures are typically short in duration and manifest themselves as sudden behavioral arrest and impaired consciousness followed by sudden termination and return to normal behavior. In electroencephalogram (EEG) recordings, absence seizures are characterized by spike and wave discharges (SWDs) that arise from synchronous firing of thalamocortical networks [11].

The onset of IGE typically occurs in childhood, although adult onset may also occur [52]. Indeed, among epilepsies involving absence seizures, childhood absence epilepsy CAE) and juvenile absence epilepsy (JAE) are the most common forms of IGE. CAE has been extensively investigated at the cellular and network levels in both animals and humans, revealing a strong causal link to mutations in various types of voltage-gated and ligand-gated ion channels, including voltage-gated potassium, sodium, and calcium channels, as well as $GABA_A$ receptors [33, 50, 59, 82]. Here, we shall focus on the role of voltage-gated calcium channels in the development of IGEs.

## Subtypes, subunit composition, and selected physiological roles of voltage gated calcium channels

Voltage-gated calcium channels can be classified, based on their biophysical characteristics, into high voltage activated (HVA) and low voltage activated (LVA) channels, with HVA channels requiring larger membrane depolarizations for activation. HVA channels can be further subdivided into L-, N-, P-, Q- and R-types, by virtue of their distinct functional and pharmacological profiles (Fig. 1A). Members of the HVA channel family are heteromultimers of a pore forming α1 subunit that coassembles with ancillary β, $\alpha_2\delta$ (and in some cases a γ subunit) into a functional channel complex (Fig. 1B) [6, 19]. In contrast, LVA channels (also referred to as T-type) are thought to be α1 subunit monomers (Fig. 1C). The α1 subunit defines the channel subtype, whereas the ancillary subunits modulate α1 subunit function and surface expression. The α1 subunit is comprised of four homologous membrane repeats (each containing six transmembrane helices termed S1 through S6, plus a pore lining P-loop motif) that are connected via cytoplasmic linker regions and flanked by cytoplasmic N- and C-termini. Ten different α1 subunit subtypes belonging to three large families are expressed in the mammalian genome (Fig. 1A). The $Ca_v1$ family encodes L-type calcium channels and includes four different members ($Ca_v1.1$ through $Ca_v1.4$). The $Ca_v2$ family includes $Ca_v2.1$, $Ca_v2.2$ and $Ca_v2.3$, which encode P/Q-type, N-type and R-type channels, respectively. The $Ca_v3$ encompasses the family of T-type channels with three members ($Ca_v3.1$ through $Ca_v3.3$). Of particular note, P- and Q-type channels appear to arise from alternate splicing of $Ca_v2.1$ and possibly the coassembly with distinct β subunits [12, 67]. Vertebrates express four different types of calcium channel β subunits ($\beta_1$ through $\beta_4$), four different types of $\alpha_2\delta$ subunits ($\alpha_2\delta1$ through $\alpha_2\delta4$), and as many as 8 different γ subunits [6, 9] (Fig. 1B). The β subunits are cytoplasmic proteins that interact with the linker joining repeats I & II of the calcium channel α1 subunit and strongly modulate channel properties such as inactivation rates [6]. Moreover, these subunits play a major role in targeting the channels to the plasma membrane. The $\alpha_2\delta$ subunit is encoded by a single gene, and it is post-translationally cleaved into a transmembrane δ subunit that remains di-sulfide linked to an extracellular $\alpha_2$ subunit [25, 44]. The γ subunit includes four transmembrane helices and was first identified as part of the skeletal muscle L-type channel complex [3]. Since then, a number of neuronal γ subunits have been cloned; however, it is not entirely clear if they are bona fide calcium channel subunits. Functional effects on both HVA and LVA calcium channel activity have been reported in expression systems [68], but a much more pronounced role has been ascribed to γ subunits in the context of regulating AMPA receptor trafficking and function [78].

The different types of calcium channel α1 subunits are differentially distributed within neurons. In the context of this review, it is important to note that P/Q-type, N-type, and, to some extent, R-type channels are expressed highly at presynaptic nerve terminals where their activities evoke neurotransmitter release [80]. T-type calcium channels tend to be expressed at cell bodies and dendrites [55], where they contribute to the regulation of neuronal excitability [63]. Taken together, P/Q-type and T-type calcium channels show distinct functional properties, subunit composition, and subcellular distributions, and they serve distinct physiological roles, and yet, they both are major contributors to the development of absence seizures and IGE.

## P/Q-type channels and absence seizures

As noted above, $Ca_v2.1$ (P/Q-type) calcium channels are important mediators of synaptic transmission in central and peripheral neurons. As a result, changes in the functional properties of $Ca_v2.1$ channels may be expected to alter the behavior of neuronal networks. A possible role of P/Q-type channels in seizure disorders is readily apparent when considering the phenotypes of $Ca_v2.1$ channel KO mice. These mice show severe ataxia and die about four weeks after birth [42]. Moreover, they display absence seizures, indicating that P/Q-type channels may protect from the occurrence of such seizure activity during normal physiology. Additional evidence implicating $Ca_v2.1$ channels in absence seizures comes from several murine mouse models with P/Q-type channel mutations (Fig. 2A). In *tottering* (*tg*) mice, a proline residue in the repeat II S5–S6 region of $Ca_v2.1$ is substituted by a leucine residue [26, 32], leading to a reduction of P/Q-type channel activity as assayed in both neurons and in expression systems [88]. Consistent with their role in neurotransmitter release, the reduced P/Q-type channel function in these mice is paralleled by altered neurotransmitter release in cortical neurons [4], and reduced excitatory synaptic potentials evoked in thalamic neurons [17]. Similar to the $Ca_v2.1$ KO mice, *tg* mice show cerebellar ataxia and absence seizures with 5–7 Hz SWDs [32, 61]. The $Ca_v2.1$ calcium channel gene of *leaner* ($tg^{la}$) mice exhibits a frameshift mutation in the C-terminal region, which results in a premature stop [26, 32]. This reportedly results in altered P/Q-type channel currents, as well as reduced neurotransmitter release at neocortical synapses [4]. The $tg^{la}$ mice display cortical SWDs that are consistent with absence seizures and, are, again, severely ataxic [32, 51]. Finally, in the $Ca_v2.1$ channels of *Rocker* (*rkr*) mice, a lysine residue in the repeat III S5–S6 region is replaced with a threonine [98]; however, it remains unclear what effects this mutation has on the biophysics of the channel. Nonetheless, these mice show spontaneous bilateral 6–7 Hz SWDs accompanied by behavioral arrest, typical of absence seizures [98].

A few other mutations associated with seizure activity have been mapped to the $Ca_v2.1$ subunit in mice, including *rolling Nagoya* ($tg^{rol}$) and *wobbly* (*wb*; Fig. 2A). Most of these mouse genotypes and their associated phenotypes, except *wobbly* [91], show a recessive inheritance pattern and, at least in the *tg* and $tg^{la}$ mice, seem to parallel the phenotype of the $Ca_v2.1$ channel KO mouse. This may be due to the fact that both mutant mice show reduced $Ca_v2.1$ channel activity, which would be equivalent to a partial $Ca_v2.1$ knockdown. Importantly, all these mouse mutants also exhibit ataxia, possibly due to reduced P/Q-type channel activity and unbalanced excitatory/inhibitory neurotransmitter release in the cerebellum, since the P/Q-current accounts for ~90% of the total calcium current in Purkinje neurons [58].

In humans, mutations in P/Q-type calcium channels are more frequently associated with conditions such as episodic ataxia type 2 (EA-2) and familial hemiplegic migraine type 1 (FHM1) [reviewed in [64] and [65]], both of which are dominantly inherited (Fig. 2B). Ataxia is then a common phenotype in both mice and men with $Ca_v2.1$ mutations. Importantly, there are some instances in which patients with $Ca_v2.1$ mutations present with

absence seizures (illustrated in Fig. 2B). For example, a truncation mutation in the C-terminal region of $Ca_v2.1$ that results in a non-functional channel has been associated with the occurrence of childhood episodes of absence epilepsy and primary generalized seizures in a patient with episodic ataxia type 2 [41]. Another mutation in the repeat I-S2 region is found in a different family of patients, which reduces P/Q-type channel activity and gives rise to an ataxic and epileptic phenotype [40]. An 11 year old girl with a missense mutation (I712V) in the $Ca_v2.1$ channel was recently described to have a range of symptoms, including seizures, headache, and ataxia [35], but the effect of the mutation on channel gating has not yet been determined. In patients with FHM associated with P/Q-type channel mutations, the occurrence of seizures is exceedingly rare and limited to only a few case reports [27]. This may be due to the notion that FHM appears to arise from a gain of function of $Ca_v2.1$, rather than a loss of function [18, 79]. Altogether, in both humans and rodents, mutations that give rise to diminished P/Q-type channel function have the propensity to cause absence seizures. As noted below, in these rodent models there is also a compensatory increase in T-currents of thalamic relay neurons [94], which likely underlies their seizure phenotype.

## Ancillary calcium channel subunits and seizures

As noted above, HVA calcium channels are multi-subunit complexes where the ancillary subunits regulate channel function and/or membrane expression of the pore-forming α1 subunit. As such, one may expect loss of function mutations in ancillary subunits of $Ca_v2.1$ channels would reduce its channel activity and perhaps produce an epileptic phenotype in vivo. In mice, mutations associated with seizure activity have indeed been found in all major classes of ancillary calcium channel subunits (Fig. 2A). The *lethargic* (*lh*) mouse arises from a frameshift mutation in the calcium channel $\beta_4$ subunit, thus resulting in an effective knockout of this subunit. These mice exhibit absence seizures, ataxia, and reduced excitatory neurotransmitter release [16]. It is interesting to note that similar mutations at splice junctions have been found in the $\beta_4$ gene in two families displaying IGE [30].

There are also several mouse phenotypes associated with mutations in $\alpha_2\delta$ subunits, including two mutations which cause a loss of $\alpha_2\delta 2$ subunit protein and which give rise to two strains of *ducky* mice (*du* and $du^{2J}$). These mice show 5–7 Hz SWD during seizure episodes, as well as behavioral arrest and ataxia [5]. In transient expression systems, the coexpression of the mutated recombinant subunits reduces the amount of $Ca_v2.1$ current [14], again, consistent with the idea that reduced $Ca_v2.1$ channel function may lead to the appearance of absence seizures. In the *entla* (*ent*) mouse, a duplication of an exon 3 interferes with the disulfide linkage of the $\alpha_2$ and δ2 subunits, thus rendering the subunits non-functional [13]. These mice also have reduced P/Q-type currents in the hippocampus, as well as 2–4 Hz SWDs in the cortex and hippocampus.

The *stargazer* (*stg*) and *waggler* (*wgl*) mice arise from dysfunctional calcium channel $\gamma_2$ subunits (also referred to as stargazin). Their phenotypes are characterized by absence seizures and episodes of head tossing [45, 46]. Seizures are further exacerbated in *waggler* mice by knockout of the $\gamma_4$ subunit [47]. In *stg* mice, there is a small reduction in P/Q-type channel activity due to shifts in the midpoint of the steady state inactivation curve of the channel, as assayed in *Xenopus* oocytes [reviewed in [9]]. In view of the known effects of stargazin on AMPA receptors, it is difficult to unequivocally attribute the physiological effects of truncation mutations in stargazin to an alteration of calcium channel function.

## Role of T-type channels in neuronal excitability

Entry of $Ca^{2+}$ ions through T-channels leads to depolarization of the membrane, allowing T-currents to generate low threshold spikes (LTS) that trigger bursts of Na-dependent action

potentials [49]. This role is especially prominent in thalamic neurons, which express T-currents at very high densities (reviewed in [63]). Thalamic neurons form a reciprocally connected circuit that oscillates during natural processes like sleep, but can also oscillate at inappropriate times, as during a generalized seizure [53]. This circuit is composed of thalamic reticular neurons (nRT; GABAergic), thalamocortical neurons that reside in the relay nuclei (TC, glutamatergic), and cerebral cortical neurons (glutamatergic). The unique voltage dependence of T-channels allows them to generate LTS after *either* an inhibitory or excitatory post-synaptic potential (IPSP, EPSP). Rebound firing after an IPSP occurs because T-channels are inactivated at the resting membrane potential of many neurons, then recover from inactivation during an IPSP. As the IPSP decays and the resting membrane potential is restored, T-channels can open and create a LTS. Another very interesting property of T-channels is their ability to generate "window currents." Window currents arise when a channel is available to open at a given potential and not totally inactivated. They are operationally defined by the overlap of the steady-state activation and inactivation curves. Notably, T-window currents occur at the resting membrane potential of most neurons, allowing them to contribute to membrane bistability; e.g. thalamic neurons have two "resting" membrane potentials, one around −77 mV, and a second around −60 mV [23]. $Ca_V3.2$ channels have been shown to generate window currents and increase resting basal $Ca^{2+}$ concentrations [20].

T-channels open and close at negative membrane potentials, where there is a large driving force for $Ca^{2+}$ entry. Their opening can lead to robust increases in intracellular $Ca^{2+}$, especially in small compartments such as dendrites [60, 97]. $Ca^{2+}$ entry via T-channels has also been shown to cause Ca-induced Ca-release (CICR) from internal stores, which can trigger a form of long-term depression in hippocampal neurons [62, 89]. In some neurons, $Ca^{2+}$ influx via T-channels dampens excitability by opening Ca-activated $K^+$ channels, which contributes to spike repolarization and after-hyperpolarizations [24, 48, 83, 90]. In summary, T-channels play important physiological roles by controlling membrane potential and by controlling intracellular $Ca^{2+}$ concentrations.

## Upregulation of T-type channels in epilepsy

There is significant evidence that T-channels play a role in epilepsy and pain. In particular, T-channels are thought to play an important role in idiopathic generalized epilepsies (IGE), such as absence epilepsy, due to their high expression in the thalamus. In addition to being supported by pharmacology studies, this hypothesis is further supported by studies in the GAERS model of absence epilepsy. Reticular thalamic neurons isolated from these rats have 55% larger T-currents than the control strain and show a 16% increase in $Ca_V3.2$ mRNA [75, 81]. Similar results were found in the WAG/Rij model of absence epilepsy [15]. It is interesting to note that increased T-type calcium channel activity was first implicated in studies of mice with absence seizure phenotypes, where the underlying mutation is in high voltage-activated channels [94]. This result provides a clear example of how misleading the correlation between phenotype and gene knockout can be. Examples of this phenomenon include: *tottering* mice with $Ca_V2.1$ mutations; *stargazer* mice with mutations in γ2; and *lethargic* mice with mutations in β4. Mutations in non-calcium channel genes also lead to absence epilepsy in the mouse model *Coloboma*, and increases in T-currents increase prior to the onset of epilepsy [95]. The hypothesis that increased T-currents play a direct role in epileptogenesis is provided by inducing epilepsy by forced overexpression of $Ca_V3.1$ currents [29], and by "curing" *tottering*, *stargazer*, and *lethargic* mice by crossing with $Ca_V3.1$ knockout mice [71]. It remains to be determined whether the absence seizures arise from the ability of increased T-currents to alter membrane excitability or intracellular calcium concentrations, although it should be noted that early modeling studies suggested a 2-fold increase in T-currents would be sufficient to induce oscillations of thalamocortical

circuitry [54]. Similarly, a recent modeling study concluded that small changes (2–3 mV, which would be virtually undetectable) in the voltage dependence of activation of $Na^+$ or $Ca^{2+}$ channels could underlie seizures [77]. Indeed, over-activity of T-channels appears critical, as demonstrated by the observation that transgenic mice overexpressing $Ca_v3.1$ channels display spike-and wave-discharges, the EEG signature of absence epilepsy [29]. This provided strong evidence that primary elevation of T-type channel activity is sufficient to induce a pure absence epilepsy phenotype. Conversely, a small reduction in T-currents by the anti-absence seizure medication, ethosuximide, may be sufficient to reduce oscillation of thalamocortical circuits in vitro [39]. Indeed, a recent study has documented that a *selective* T-channel blocker exhibits strong efficacy in suppression of absence seizures in the genetic rat model WAG/Rij [93].

T-currents are also upregulated in CA1 hippocampal neurons in the pilocarpine model of temporal lobe epilepsy (TLE) [7, 74]. The original observation was that pilocarpine-induced status epilepticus promoted spontaneous firing of CA1 neurons [74]. A subsequent study using focal application of $Ni^{2+}$ and amiloride implicated upregulation of T-currents in apical dendrites [92]. A recent study established the role of $Ca_v3.2$ channels in this form of epilepsy using $Ca_v3.2$ (−/−) mice [7]. Interestingly, the strength of synaptic inputs into CA1 neurons is altered in this model of TLE with a dramatic increase in the entorhinal cortex (EC) input via the temporoammonic pathway [1]. These inputs terminate in the most distal dendrites in the stratum lacunosum-moleculare [56]. Although other factors may be involved, a plausible explanation is that upregulation of $Ca_v3.2$ channels boosts EPSPs triggered in distal dendrites. Hippocampal T-current density is also increased after electrical kindling [31]. These animal models mimic human temporal lobe and focal epilepsies, implicating possible upregulation of T-currents in many forms of epilepsy.

## *CACNA1H* is an epilepsy susceptibility gene

The word *mutation* has two connotations. To a molecular biologist, a mutation is any change in the nucleotide sequence that differs from a standard reference sequence (e.g. the GenBank). To a geneticist, a mutation is a change in nucleotide sequence that has been shown to *cause* disease. This strict interpretation requires that the term *mutation* only applies to monogenic autosomal dominant disorders. Monogenic disorders that cause epilepsy are relatively rare [57]. In contrast, idiopathic generalized epilepsy (IGE) is a polygenic disorder [34, 38, 69, 76, 84]. The first evidence that IGE was a polygenic disorder came from Dr. Lennox's study of epilepsy in twins [84]. He found that even in monozygotic twins there was only a 75% concordance for absence epilepsy, with a 0% concordance in dizygotic twins. Other characteristics of polygenic disorders found in absence epilepsy include a variable age of onset, variable age of recovery, and periodicity [34]. Furthermore, generalized spike-and-wave discharges have been observed in family members who do not have seizures [69]. Finally, the conclusion from many linkage studies is that the complex pattern of IGE inheritance is not due to a single gene, but that "each gene contributes a small or modest effect to the epilepsy phenotype, and by itself is insufficient to cause epilepsy. These are 'susceptibility genes'" [76]. Therefore, common single nucleotide polymorphisms (SNPs) can contribute to epilepsy susceptibility. More importantly, a large number of SNPs have been found only in epilepsy patients (Fig. 3). We will refer to these as "*variants.*" The first of these studies was performed on Childhood Absence Epilepsy (CAE) patients in China, leading to the identification of 12 specific variants in *CACNA1H* associated with CAE [21]. Interestingly, patients from 3 distinct families harbored both the G773D variant and "common" SNP R788C, which was found in 20% of CAE patients and in 10% of the general population [87]. A second study extended the finding of *CACNA1H* variants to many related epilepsies, such as juvenile absence, juvenile myoclonic epilepsy (JME), febrile seizures, and temporal lobe epilepsy (TLE) [36]. Sequence variations in the $Ca_v3.2$

gene, *CACNA1H*, have also been linked to Autism Spectrum Disorders (ASD) [72]. In addition, variants were detected in the $Ca_v3.1$ gene, *CACNA1G*, in IGE patients, although no phenotypic differences were noted in channel behavior [70]. Studies on the Generalized Absence Epilepsy Rat of Strasbourg (GAERS) confirmed this conclusion, as a variant in the III–IV loop (Fig. 1) accounts for some, but not all, of the seizure phenotype [66]. Notably, this variant, R1584P, alters the ratio of III–IV loop splice variants, favoring the variant with faster recovery from inactivation. Similarly, IGE of the human *CACNA1H* gene, supporting the hypothesis that variants simply alter the ratio of splice variants with unique properties, but do not themselves induce novel phenotypes [96]. In crosses of GAERS with control rats, the R1584P mutation segregated with seizures (percentage time and total number of seizures). Interestingly, a subset of rats did not inherit the mutation, yet were still prone to seizures, and vice versa. Taken together, these studies on both humans and animal models clearly establish that IGE is a polygenic disorder, and that sequence variations in *CACNA1H* can contribute to seizure susceptibility.

Despite considerable progress in defining the role of calcium channels in epilepsy, there remain many unanswered questions. How do these $Ca_v3$ variants increase seizure susceptibility? In the most simplistic sense, epilepsy is a disorder in which the balance between excitatory and inhibitory neurotransmission is tipped towards excitability. Therefore, one can predict that T-channel variants might cause changes in gating that would increase seizure susceptibility, such as: 1) shifting the voltage-dependence of activation; 2) shifting the voltage dependence of inactivation; 3) accelerating channel opening; slowing channel inactivation; 4) slowing deactivation (open to close transitions); 5) accelerating recovery; 6) increasing the probability of channel opening ($P_o$); and 7) increasing single channel conductance. Notably, variants associated with IGE do affect many of these properties [43, 87]. Surprisingly, some of the $Ca_v3.2$ variants found in IGE patients had no detectable effect on channel biophysics. During a structure-function study it was discovered that the loop connecting repeat I to II (I–II loop; Fig. 1) played a role in channel expression [2]. This study went on to show that all CAE variants located in this loop increased surface expression, thereby providing a unifying mechanism for their role in seizure susceptibility [86]. Mechanisms by which this might occur include: enhanced trafficking out of ER/Golgi to plasma membrane (PM); slower retrieval from PM; altered fate of internalized early endosomes from degradation to PM recycling; and altered distribution of T-channels in neurons, e.g. enhanced trafficking into dendrites or axon hillock. Another unresolved question is: Do the $Ca_v3$ variants alter trafficking to dendrites? Based on their known roles in dendritic physiology [73], this would likely have significant effects on burst firing and synaptic integration. Finally, we would like to emphasize the difficulty in demonstrating that a channel variant is the root cause of epilepsy, because IGE is a polygenic disorder and by definition, requires the co-inheritance of variants in other genes (nature), as well as contributions from the environment (nurture) [8, 77].

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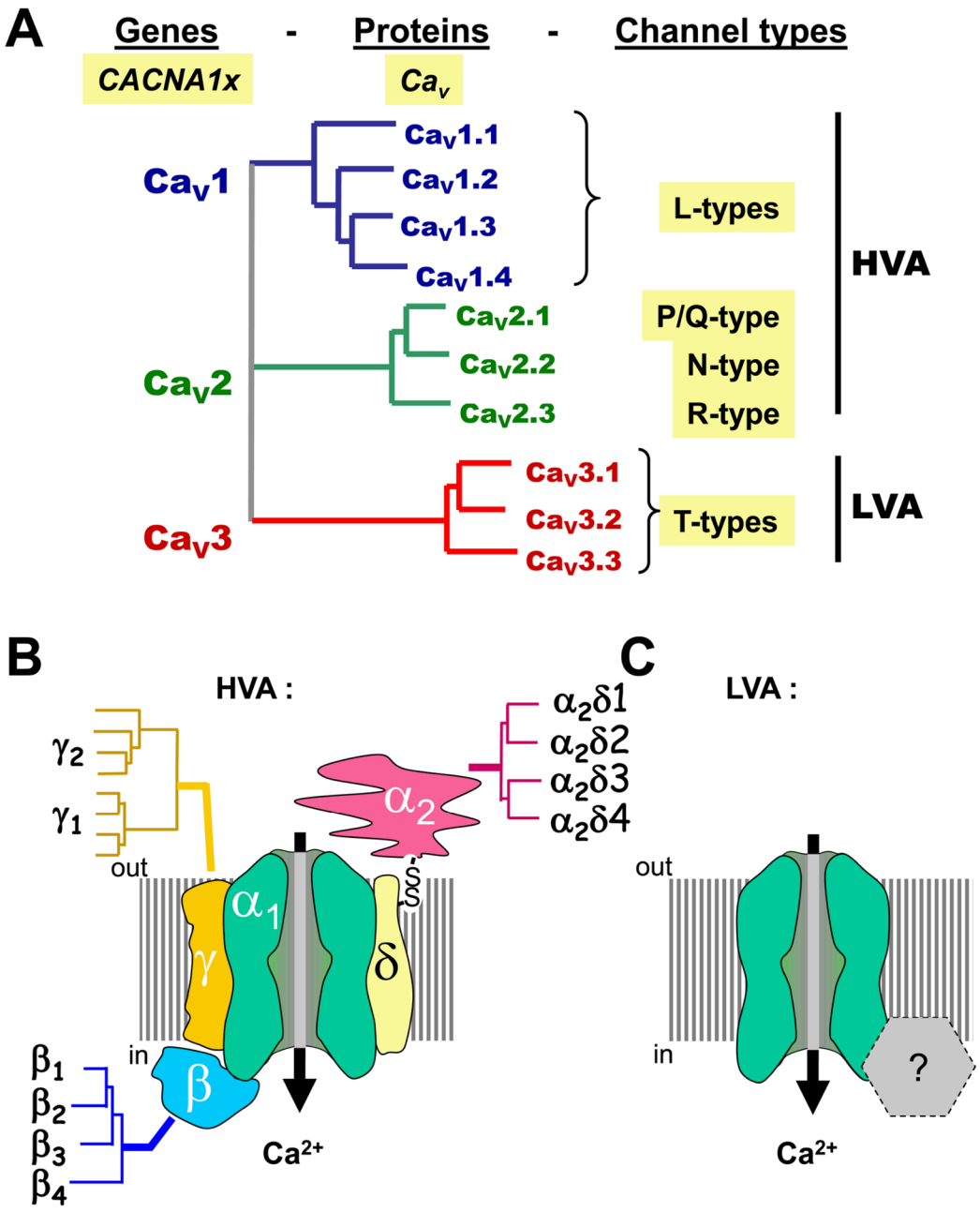


**Figure 1. Voltage-gated calcium channels (VGCCs): naming and structure**

**A.** Dendrogram illustrating the three subfamilies of VGCCs: the $Ca_v1$ (L-types), the $Ca_v2$ (neuronal types), and the $Ca_v3$ (T-types). $Ca_v1$ and $Ca_v2$ channels are high-voltage activated (HVA), in contrast to $Ca_v3$ channels (T-types), which are low-voltage activated (LVA). **B.** The molecular structure of HVA channels comprises ancillary subunits $\alpha_2\delta$, β, and γ, each encoded by several subunits. In contrast, the subunit composition of LVA/T-type channels is not yet resolved.

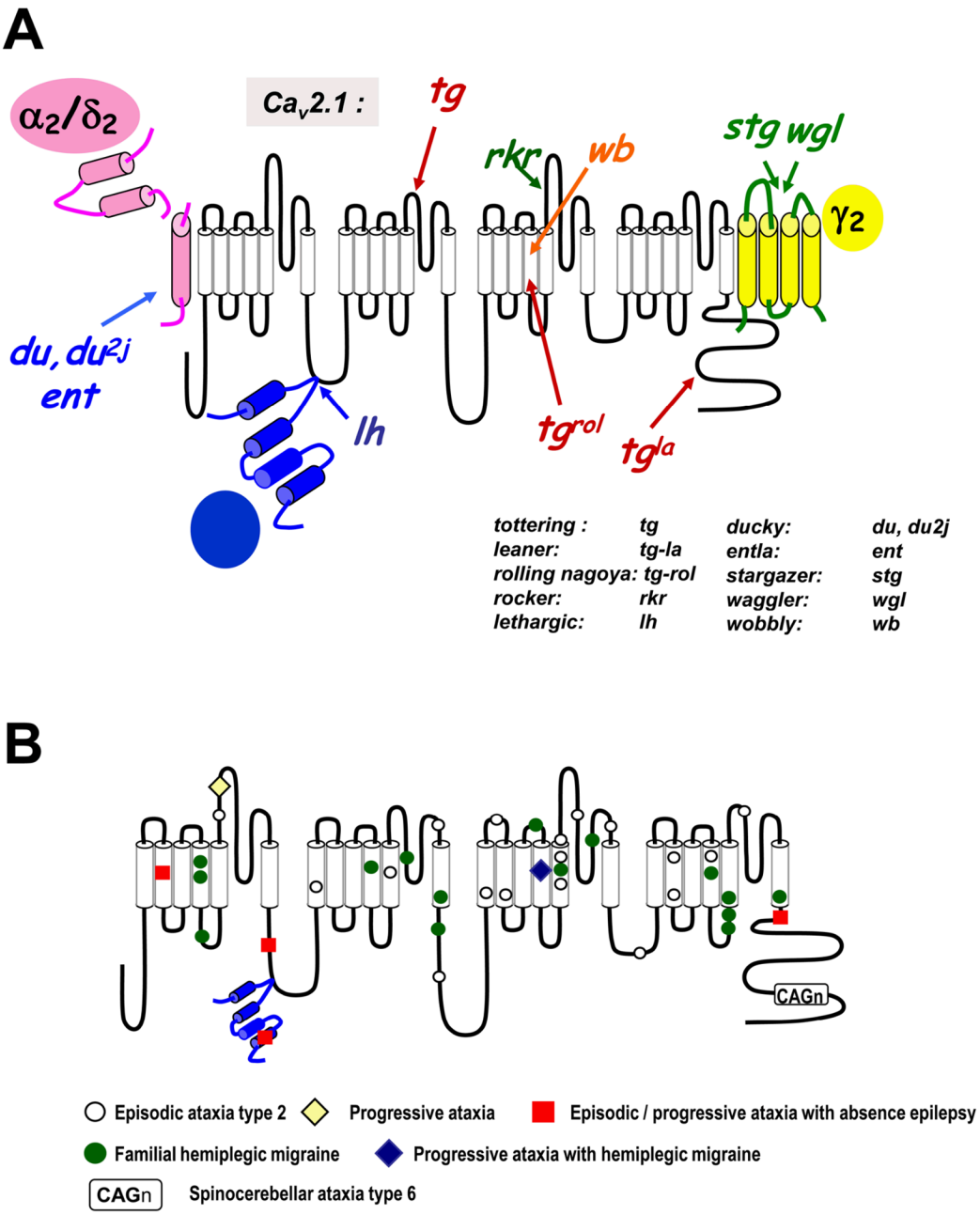


**Figure 2. Mutations in P/Q-type calcium channels in mouse (A) and humans (B)**

**A.** Schematic representation of the multiple mutations that affect the various subunits of the P/Q-type calcium channels in mouse and the corresponding mouse phenotypes. **b** Location of mutations in human P/Q-type α1 subunits that related to episodic ataxia type 2 (EA2), familial hemiplegic migraine type 1 (FHM1), spinocerebellar ataxia type 6 (SCA6), as well as more complex clinical patterns such as progressive ataxia with absence epilepsy (red squares) are indicated.

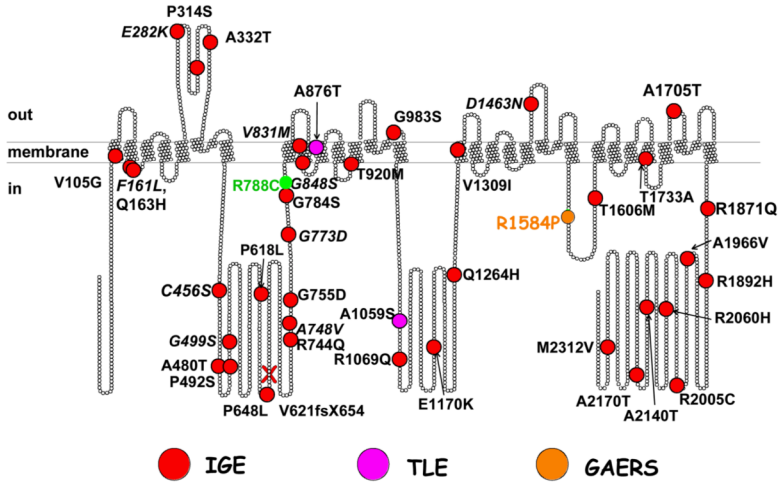


**Figure 3. Variants of $Ca_V3.2$**

Location of IGE variants are mapped on a scaled model of $Ca_V3.2$ (each ball represents an amino acid). Key: red balls, IGE variants reported by Chen et al., [21] and Heron et al., [37]; orange ball, the GAERS variant [66]; pink balls, TLE variants [37]; and green, the common SNP, R788C. A frame-shift mutation, V621fsX654, was found in patients with febrile seizures, which leads to premature truncation of the protein at a.a. residue 654 [37].