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EDITORIAL

Cav3 T-type channels as drug targets for treating epilepsy



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• non-REM sleep

Epilepsy is a neurological disorder characterized by spontaneous brain seizures. Approximately 50 million people worldwide have epilepsy and approximately 30% of patients have seizures that are not under control with current anti-epileptic drugs [1]. Epilepsy can be focally localized to a particular brain region (such as those caused by lesions or tumors) or more generalized to both hemispheres [1]. Generalized epilepsy can be subdivided as epilepsy that is derived from an identifiable symptom, (such as brain damage caused by an infection or trauma) or is idiopathic, where the cause of origin is hidden, such as those with a genetic underpinning [1]. Idiopathic generalized epilepsies are typically associated with absence seizures, originally dubbed 'petit mal', or 'little illness', characterized by brief 'blanking out' or 'absence' and quick return to consciousness and typically occur without convulsions and without postseizure fatigue [2]. Absence seizures commonly begin in children ages 4–8 years, and is often difficult to diagnose, because a child often just appears to be daydreaming, rolling up their eyes a bit and staring, unresponsively and unaware for brief 10-s periods, which can repeat often, up to 100-times per day in children with recurrent absence seizures [2].

The EEG recordings during an absence seizure is a characteristic 1.5–4 Hz (cycles per second) spike and wave discharge (SWD) observable in the neural circuit between groups of interconnected neurons in the thalamus and cortex [3] normally associated with non-rapid eye movement (REM) category of sleep state, including delta waves, sleep spindles and k-complexes [4]. It is believed that the pathological SWD of absence epilepsy is to alter the synchrony of firing patterns normally associated with the sleep-related circuit between this intrathalamic set of neurons and pyramidal neurons on the cerebral cortex layers V–VI [5].

'Transient' or 'T-type' class of calcium channels are 'rhythm generator' ion channels, supporting the heartbeat and an even more prominent role in setting brain circuit rhythms within the thalamus [6]. T-type channels are at their highest density in the human body within the thalamus intersecting the circuits underlying normal sleep patterns and the abnormal, pathological rhythms during absence epilepsy. T-type channels have ion selective pores

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that are selective for the inflow of positive calcium ions (and some sodium ions [7]) generating excitability across nerve cell membranes. T-type channels are 'first responders' from the resting state with a low voltage threshold for ion gating [6]. They facilitate a rebound burst of excitation due to input from inhibitory synapses, and can also respond quickly to opening after small inputs from excitatory neurons [6]. T-type channels are a primary engine for driving the rhythmic, periodic firing pattern between sets of interconnected neurons, sustaining nerve excitability with any input that pulls neurons out of the resting state [3]. T-type channels can even generate a trickle of excitatory 'window' current at rest itself [8], contributing to the transition from the 'downstate' to 'upstate' that underlies the (<1 Hz) oscillations of slow wave sleep. Sensory input (hearing, smell, touch, taste, vision) first passes through the thalamus, and thus the thalamus serves as a gatekeeper to conscious awareness. It is the oscillatory patterns of firing within the thalamus, primed by T-type channel activity which dampens our sensations (hearing, smell, touch) during non-REM sleep [3].

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Thalamocortical rhythms become pro-epileptic with increases in T-type channel activity. Transgenic mice overexpressing CACNA1G T-type channels, are more prone to absence seizures [9]. T-type calcium channel activity also rises significantly in animal models of absence epilepsy including those generated by kindling (epilepsy induced by repeated electrical stimulation), in the Generalized Epilepsy Rat of Strasbourg (GAERS) and the Epilepsy Wistar Albino Glaxo/Rij (WAG/Rij) rat, as well as seizure-susceptible mice with descriptive phenotypes: tottering, stargazer and lethargic where the underlying mutation is in calcium channel subunits that are not T-type channels [5]. Increased human susceptibility to epilepsy has been linked to single nucleotide mutations in T-type calcium channel genes. Twelve specific single-nucleotide polymorphisms have been identified in the T-type channel gene, CACNA1H from a study of 118 cases of childhood absence epilepsy from Chinese patients [10]. These mutations enhance expression levels of T-type channels and often their functional activity too (gating and kinetics), which is modeled to reduce the threshold for burst firing and increasing the spontaneous firing rate in thalamocortical neurons [11]. Animals and human studies provide a convincing link between overactive T-type channels and a susceptibility to absence seizures.

Selective impairment of T-type channel activity will prevent absence seizures. Knockout animals of the T-type channel gene, CACNA1G lack the burst firing mode of excitation in thalamocortical neurons that is consistent with absence seizures, and these animals are resistant to the SWD seizures induced by some, but not all drugs [12]. No medicines clinically available today are specific for blocking T-type calcium channels alone, but many of the current therapeutics for treating epilepsy do reduce T-type channel activity as one of its many drugs targets. Standard childhood absence epilepsy drugs that block T-type channels include Zarontin® (ethosuximide; Parke-Davis, Pfizer, NY, USA) [13] and Depakote® (valproate; Abbott Laboratories, IL, USA) [14], and newer generation, anti-epileptic drug, Zonegran® (zonisamide; Élan, Dublin, Ireland) [15] introduced this century and old formulation (1938), Dilantin® (Phenytoin; Parke-Davis) [16]. Posicor® (mibefradil; Roche Laboratories, NJ, USA) originally marketed as a selective antihypertensive agent, will block T-type channels and prevent seizure activity in animal models [14], but is no longer in clinical use because of its side effects on drug metabolism.

Promising novel and more potent T-type channel antagonists have been recently identified that have clinical potential for treatment of absence epilepsy. Merck (PA, USA [formerly Neuromed Technologies, Vancouver, Canada]) and Epirus Biopharmaceuticals (MA, USA [formerly Zalicus Pharmaceuticals, MA, USA]) isolated drug compounds including TTA-P1, TTA-P2 and Z941, Z944 [17], respectively, from screening a library of variations with either a piperidine or piperazine scaffold. Merck has a number of potent drug compounds besides TTA-P1 and TTA-P2 [18], including amide-derivatives, TTA-A2 [19] and quinazolinone derivative, TTA-Q4 [20]. All these new generation of T-type channel antagonists block in the nM concentration range, and block with a much greater potency for T-type channels than ethosuximide or valproate, the most commonly used absence epilepsy drugs. These newer T-type channel antagonists block thalamic burst firing, and suppress absence seizures in animal models. Z944, is the only drug so far in clinical trials, with positive results after Phase Ib and is currently being put into a modified-release formulation for Phase II clinical development in 2014.

Dampening excitability of over-active T-type channels with more specific antagonists is clearly on the horizon as a potential treatment for absence epilepsy. However, it is still early to tell how useful new drug formations will be compared with currently available anti-epileptic drugs. First, even highly specific drugs may have variable effectiveness in different patients. Absence epilepsy is a complex polygenic disorder, where changes in one gene merely tips the balance slightly to a more excitable phenotype, and no single gene is solely responsible for the susceptibility in absence epilepsy [1]. Different patients may require more tailored drug treatments because of differing genetic underpinnings for their absence epilepsy.

We also have to consider that there are three different human T-type channel genes expressed in most tissues including the thalamus, and each gene is similarly blocked by known T-type channel drugs [5]. It is not clear at this stage which one or all of the T-type channel genes is more important as a drug target for treatment in absence epilepsy. All three T-type channel genes appear to have some overlap to functionally compensate for each others' absence in animals where a specific T-type channel gene is knocked out during development [3]. Effects on sleep or epilepsy in these T-type channel knockout animals can be less severe to muted, compared with drug application of T-type channel antagonists or transient gene knockdown, where developmental compensation between T-type channels is not possible [3]. My laboratory has discovered a potential determinant for generating more selective blocking antagonists for each of the three T-type channels. We recently found that T-type channels have an extracellular, cysteine scaffold above the pore of T-type channels that can alter the accessibility of drugs and is highly variable among T-type channels [7]. This extracellular scaffold could be a useful determinant to develop more selective T-type channel blockers.

It may be the case though that a generalized, pan-selective T-type channel antagonist may be less effective than a drug that is selective for one of the T-Type channel genes. Functional differences among T-type channels are nuanced, but highly tailored for differing roles. Each T-type channels has unique expression profiles in the nervous system, the cardiovascular system and endocrine systems [6]. Further complicating the diversity is that each of the three T-type channels is subject to unique gene splicing [6]. The differing genes and their spliced variants create T-type channels with a unique subset of biophysical properties and spatial distributions, contributing in novel ways to excitability within different cell types [6]. There is no question that T-type channels contribute to susceptibility to absence epilepsy, and that specifically blocking T-type channels may be more effective than ethosuximide or valproate in treatment of absence epilepsy. The question is which of the T-type channel genes is most important as a drug target, and whether specific T-type channel antagonists will be an equally effective treatment in all patients, given that the absence epilepsy phenotype varies with patients of different genetic backgrounds.

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