Epilepsy is a diverse spectrum that includes over 40 disorders in which patients suffer from seizures of varying severity. Causes of epilepsy include genetic mutations that target ion channels and environmental factors such as brain injury, stroke, or neurotoxic chemicals. These alterations can affect the thalamus, cortex, and the hippocampus. Epilepsy can be diagnosed based on the frequency of seizures and brain scan patterns. This review focuses on the dysfunction of ion channels in presynaptic/postsynaptic neurons and astrocytes, which contribute to hyperexcitability and hypersynchronous firing. In presynaptic neurons, failure of inhibitory circuits and reinforcement of excitatory circuits through mutations in Na+, K+, Ca2+, Cl- ion channels results in epileptogenesis. In postsynaptic neurons, hyperexcitability can result from mutations in sodium channels, other dysfunctional ionotropic receptors and muscarinic receptors of glutamate and GABA. Abnormal astroglial activity is causally linked to the onset of seizures, which can lead to potassium imbalances and overexpression of sodium channels in glia, exacerbating the excitability of presynaptic or postsynaptic neurons and making a further seizure more likely. Anticonvulsant medication is the most common treatment for milder forms of epilepsy, while severe cases require surgery. Future research is heading towards identifying the mechanistic pathways of dysfunctional ion channels and targeting them as a form of treatment.

Introduction

Epilepsy is a spectrum of debilitating neurological disorders characterized by non-elicited seizures that may or may not lead to a loss of consciousness (Chang et al., 2003). These disorders are highly prevalent. At least 17.6 out of 1000 people worldwide are afflicted (Banerjee et al., 2009). Unfortunately, a diagnosis of epilepsy shortens a patient’s lifespan by approximately 20% (Gaitatzis, 2004). The most recent classifications list over 40 types of epileptic seizures, each defined with its own set of symptoms (Engel, 2003). The broadest categorization defines these neurological events as either generalized or partial (Epilepsy Foundation, 2009). Generalized seizures spread a storm of excitatory firing throughout the entire brain, while partial seizures are focused only on a particular region (Chang et al., 2003). In these cases, motor, cognitive, and autonomic symptoms emerge.

Epilepsy can be caused by genetic or environmental factors. Genetic factors include mutations of ion channels, including Na+, Cl-, K+, and Ca2+. Mutations can also take place in upstream molecules of cell proliferation and metabolism (Novarino et al., 2013). Environmental factors such as chemical weapons, toxic byproducts or pesticides can increase glutamate levels, facilitating high-frequency action potentials (Jett et al., 2012). In addition, brain injury and neurodegenerative diseases can lead to epilepsy (Shorvon et al., 2011). Current treatments for epilepsy include phenobarbital (increases GABA activity) and ethosuximide (blocks Ca2+ channels - Schmidt et al. 2014). These treatments do not ameliorate symptoms for 20-30% of epileptic cases. For these cases, a ketogenic diet can have anticonvulsive effects (Stratsform et al. 2004). Finally, surgical lesion of the corpus callosum is an option for the most severe cases (Shimizu et al. 2005).

As shown in Figure 1, dysfunction can lead to epilepsy at three distinct locations in the synapse: the presynaptic neuron, postsynaptic neuron, or astrocytes. This review will focus on ionotropic dysfunctions in each of these sites.

Figure 1. Ionotropic dysfunctions in epilepsy, accounting for depolarization and hyperexcitability. In the presynaptic cell, Cav3.2 [Ca2+]i, Nav1.6 [Na+], CIC-2 [Cl-] and, Kv7.2 [K+] became overactive. In the postsynaptic cell, increased depolarization results from overactivation of channels such as AMPA, NMDA, and Nav1.2. Increased Na+ concentration could lead to a higher frequency of action potentials. In astrocytes, Nav1.6 Na+ channels become overexpressed after a seizure. The Na+/K+/Cl- cotransporter is deactivated (increasing extracellular K+), and Ca2+ transients are abolished.

Hyperexcitable and hypersynchronous neuronal firing gives rise to epileptic seizures. (Beenhakker, 2009). Oscillating circuits between the cortex and thalamus (synchronizing firing during sleep) have been identified as areas especially prone to hyperexcitation and seizures (Beenhakker, 2009). The CA1 and CA3 regions of the hippocampus (as well as the hilus of the dentate gyrus, which atrophies) have also been linked to epileptogenesis (Avoli, 2007). Regardless of the type of seizure or its origin, this neurological event is defined by the emission of synchronized brain waves at a frequency of 10 spikes per second (Gloor, 1998).

Risk factors increase an individual’s likelihood of developing epilepsy. Some examples include brain dysfunction, developmental disorders, neurological disease [Alzheimer’s, cerebral palsy, autism], or brain injury from stroke and exposure to toxic chemicals (Epilepsy Foundation, 2014). Variables including age, ethnic background, and gender add to these risk factors. Idiopathic epilepsy is more common in women and usually strikes after the age of 20 (Christensen et al., 2005 & Nicolson et al., 2004). There are many ways to diagnosis epilepsy. One noninvasive method, known as electroencephalogram or EEG, uses electrodes to record brain activity from neuronal electrical impulses (Smith, 2005). EEG helps doctors differentiate between the type of seizure disorder and the categories into which it fits (focal, generalized, symptomatic, or idiopathic - Smith, 2005). Another technique, neuroimaging, can provide the appropriate diagnosis by identifying structural or metabolic abnormalities (Bano et al., 2011). One hallmark characteristic of epilepsy is the mutation of ion channels which lead to imbalances in the concentration of ions, creating neuronal hyperexcitability (Bernard et al., 2004). For this reason, epilepsy is often categorized as a channelopathy.

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The Role of Presynaptic Neurons

Healthy

Countless neural interactions that occur when an individual moves, feels emotions, maintains homeostasis, and thinks. The information is transmitted as action potentials between the presynaptic and postsynaptic neurons. This process is called neurotransmission (Abbott & Regehr, 2004). For an excitatory signal, the presynaptic neuron initiates an action potential as an electrical event (Murthy & Camilli, 2003). The action potential travels through the synapse as a chemical signal, depolarizes the membrane of the postsynaptic neuron, and is again converted to an electric signal. One type of voltage-gated calcium channel, Cav3.2, allows Ca2+ ions to enter the cytoplasm (Powell et al., 2014; Zamponi et al., 2010; Murthy & Camilli, 2003). In healthy individuals, Ca2+ ions stimulate the fusion of the synaptic vesicles to the presynaptic membrane and the release of the neurotransmitters into the synaptic cleft, eventually binding to the postsynaptic neuron receptors (Murthy & Camilli, 2003). Although Ca2+ ion channels play a critical role, there are many other ion channels involved in the excitability of the presynaptic neuron.

We will be discussing specific voltage-gated ion channels which are responsible for different types of epilepsy. It is important to understand that dysfunction in any one of these channels leads to the same hyperexcitability and hypersynchronous conditions. Overactive sodium channels mediate neuron excitability. One is Nav 1.6, which helps facilitate the action potential of the presynaptic neuron (Makinson et al., 2017; Shao et al., 2017). Potassium channels like Kv7.2 channel regulate neuronal excitability by repolarizing the membrane, restricting repetitive stimulation (Jorge et al., 2011). Chloride channels, like CIC-2, are also important for synaptic inhibition. Their involvement with epilepsy is still not fully understood (Stötting et al., 2014).

Epileptic

In dysfunctional presynaptic neurons, the normal functions of the channels discussed above are reversed. This leads to epileptogenesis (Powell et al., 2014). Research has shown changes in protein expression and function of Cav3.2 channels resulting in overactivation/overexpression (Powell et al., 2014). Researchers have induced seizures through muscarinic receptor agonists or in vitro mutations (Powell et al., 2014). Neurons with dysfunctional Cav3.2 channels exhibited hyperexcitability and hypersynchronous firing. (Powell et al., 2014). This resulted in some pathological hallmarks in acquired temporal lobe epilepsy and general genetic epilepsy like mossy fiber sprouting and gene expression (Powell et al., 2014).

Similar outcomes were seen with sodium channels (their function is described above). Shao et al discusses that Nav1.6 is one of the most common sodium channels expressed in the central nervous system (Shao et al., 2017). The study used lithium pilocarpine induced status epilepticus to mimic seizures. This would result in alterations to Nav1.6’s protein expression and function. It was found that Nav1.6 displayed an increase in Na+ currents, thus leading to hyperexcitability. Bursting action potentials created by overactive sodium channels are a major factor in one most turbulent types of epilepsy, temporal lobe epilepsy and general genetic epilepsy like mossy fiber sprouting and gene expression (Powell et al., 2014).

In 2016, Devaux et al found that the gain of function mutation p.V175L in Kv7.2 potassium channel caused early onset epileptic encephalopathy (EOEE) (Devaux et al. 2016). Counterintuitively, potassium channels experienced a hyperpolarizing shift, leading to overactivation and hyperexcitability. This pathological mechanism needs to be elucidated further (Devaux et al. 2016).

The link of chloride channels, such as the CIC-2 channel, has not been researched as extensively (Stötting et al., 2014). However, patient cases with idiopathic epilepsies have shown missense mutations on the CLCN2 gene for CIC-2 (Stötting et al., 2014). Consequently, the effects of the mutations are coupled with other genes which then have led to hyperexcitability (Stötting et al., 2014). This is due to the increase of inward flow of chloride, allowing a depolarized membrane state (Stötting et al., 2014).

The Role of Postsynaptic Neurons

Healthy

Glutamate is released from presynaptic neurons to the postsynaptic neurons in a nonsynchronous manner. Glutamate can affect the postsynaptic neuron through two kinds of receptors: ionotropic and metabotropic. Three kinds of ionotropic receptors (AMPA, NMDA, and kainate) of glutamate bind to glutamate and open up the channel. This leads to an influx of sodium ions. It specifically opens up AMPA sodium ion channels upon binding. The increased sodium concentration inside repels the magnesium ions in the NMDA receptor and thus opens up the NMDA channel for further depolarization. This depolarization leads to an action potential in the postsynaptic neuron. Then the neurotransmitter is absorbed back by the glutamate transporters. There are many classes of glutamate G-protein coupled receptors (metabotropic). Upon glutamate binding on the receptor site, a signal transduction cascade is activated that opens the sodium channels and allows for depolarization (Armstrong et al., 2000).

Epilepsy can also be due to alterations of the GABA system. GABA, released by the same neurons that release glutamate or by a different neuron, can act upon postsynaptic neurons, similarly to glutamate. However, when GABA binds to either its ionotropic receptor (GABAA) or metabotropic receptor (GABAB), it leads to an inhibitory effect. For ionotropic receptors, chloride channels open up and the influx leads to hyperpolarization of the postsynaptic neurons. Thus, there is no action potential. In the non-epileptic population, the GABA and glutamate firing are balanced and thus there are no epileptic symptoms (Alger et al., 1981).

Epileptic

In dysfunctional neurons, there is increased glutamate release and decreased GABA release, which leads to hyperexcitability of the postsynaptic neurons. Research has shown that mutations in GRIN2A (gene for NMDA receptor), can lead to epilepsy. These mutations can affect the receptor in many ways including removal of the Mg2+ block or alteration of the receptor structure. When glutamate binds to the NMDA receptor, this can make it easier for the postsynaptic neuron to become depolarized and hyperpolarizable (Addis et al., 2017). Rakhade et al. proposed to understand the role of AMPAR in epilepsy. They found that in rat hippocampi the AMPAR were activated compared to normal rats. They also found that the epileptic rats had higher activity of CAMKII, PKA, and PKC (Rakhade et al. 2008). It has also been found that mutations in sodium channel (specifically, Scn2a) contribute to epilepsy. CAMKII has been shown to modify sodium currents. Researchers hypothesized that CAMKII modulates sodium current and leads to hyperexcitability. Inhibition of CAMKII showed that the excitation of postsynaptic neurons decreased (Thompson et al., 2017).

It has also been found that epilepsy can result from decreased GABA activity. It has been shown that death of GABA interneurons decreases the inhibitory effect on the postsynaptic neurons. This happens because less GABA is binding to the GABAA receptor. This can lead to temporal lobe epilepsy (Ghadiri et al. 2016). Surprisingly, it has also been found that upregulation of GABAA receptors can lead to excitation and this can eventually lead to epilepsy (Lewin et al. 2012). The mechanism for this is not entirely known. Some research has uncovered that unlike mature neurons, immature neurons have a high intracellular chloride concentration and a high extracellular potassium ion concentration. The opening of GABAA by GABA can lead to efflux of chloride and result in depolarization of the postsynaptic neuron (Staley et al. 2006). Overall, the mutations in ionotropic receptors, increased activity of ionotropic receptors/metabotropic receptors due to higher glutamate, or lower GABA activity can lead to a higher frequency of action potentials, giving rise to seizures. Diseases involving mutations of GPCRs are rare. The majority of current research targets the ionotropic channels of glutamate or GABA.

The Role of Glial Cells

Healthy

Although not as well-known as their neuronal counterparts, glial cells, specifically astrocytes, have crucial supporting roles. During neurogenesis, they are responsible for secreting neurotrophic factors, guiding axons, and tightly shutting the blood brain barrier (Cabezus, 2014). Astrocytes retain their central role in maturity, providing both structural and metabolic support for neurons (Fink, 2016). Recent studies have elucidated the fact that astrocytes can participate in signaling across synapses, a task previously attributed only to neurons (Min et al., 2012). Recording from pyramidal neurons and associated astrocytes in rat brain slices, researchers discovered that as the concentration of calcium ions in the astrocyte increases, the resulting transient could trigger the release of...
Epileptic

Although astrogliosis is characterized as one of the pathological hallmarks of epilepsy, the role of ionotropic imbalances related to glial cells has remained largely ignored. The question of whether astrogliosis is a cause of seizures or an effect of status epilepticus has hindered researchers. This gap was addressed in 2015, when a mouse model of epilepsy was used to investigate disease onset (Robel et al., 2015). The researchers induced astrogliosis by genetically deleting beta-1 integrin. The mice exhibited no outward pathological signs, but they developed seizures six weeks after the activation of astrocytes. Their brains showed signs of hyperexcitable neuronal firing, poor uptake of glutamate, and low concentrations of GABA. By manipulating astrogliosis and measuring the onset of seizures, this paper showed that the activation of astrocytes is a causal factor in the onset of neuronal hyperexcitability (Robel et al., 2015).

One common theme in the search of astrocytic culprit's hallmarks of epilepsy has been the identification of ion channels whose expression changes after a seizure. These alterations disrupt the synaptic concentrations of their respective ions, increasing the excitability of nearby presynaptic or postsynaptic cells. Robel et al. (2015) found that changes in the expression of two cation-chloride cotransporters were correlated with epileptic seizures. Immediately after a seizure, NKCC1 (a Na+/K+/Cl- co-transporter that drives Cl- efflux from the astrocyte) was overexpressed and KCC2 (a K+/Cl- symport that brings Cl- into the astrocyte) decreased. As a result, the synaptic concentration of Cl- was too high. This imbalance disrupted normal GABAergic inhibitory circuits, facilitating neuronal depolarization and making seizures more likely (Robel et al., 2015).

In 2016, Zhu et al. proposed a similar paradigm, this time involving astrocytic Na+ channels. Nav1.6 is a type of voltage-gated sodium channel found ubiquitously in neurons and astrocytes of the CNS. In a rat model, mRNA and protein analyses confirmed a significant increase in the expression of astrocytic Nav1.6 channels after an epileptic seizure. This contributed to an excess of Na+ ions outside the astrocyte. The overexpression was observed in astrocytes from three regions of the hippocampus that have been linked to epilepsy: CA1, CA3, and the hilus (Zhu et al., 2016). The general assumption is that extraneous Na+ in the synapse depolarizes neurons at this chemical junction, leading to increased firing. As a consequence of a seizure, alterations in the expression of astrocytic ion channels could lead to neuronal hyperexcitability.

Other proposed mechanisms have also involved astrocytic ion imbalances in pathways that lead to depolarization. For example, in 2013, Thrane et al., demonstrated that isoform 1 of the astrocytic Na+/K+ Cl- cotransporter becomes overactivated when mice cannot metabolize ammonia. The cotransporter’s activity results in an increased K+ concentration outside astrocytes, undermining synaptic inhibitory pathways. This pathway shows that seizure propensity mediated by astrocytes could arise from non-neurological dysfunctions (Thrane et al., 2013). Also, a loss-of-function mutation on an astrocytic Na+/Ca2+/K+ co-transporter known as zydeco was found to abolish an oscillating Ca2+ transient (Melom et al., 2013). In Drosophila, this deficiency led to seizures in both adults and larvae. By interacting with neurons, astrocytes could contribute to hyper-excitatory firing in epilepsy. Although the papers highlighted here have focused on some possible mechanisms, some questions remain open. Research has focused on accounting for hyper-excitability, but experiments to explain hyper-synchronous firing from the perspective of astrocytes have not been proposed, and studies investigating genetic anomalies leading to dysfunctional channels have also been insufficient.

Treatments Targeting Ion Channels:

As shown in Figure 2, some current and many novel treatments of epilepsy target ion channels in presynaptic and postsynaptic neurons as well as in astrocytes.

Presynaptic treatments:

Current

Current treatments are available, but the results are short lived. There are many avenues for treatment. However, most treatments entail anti-epileptic drugs (AEDs). Anti-epileptic drugs help control the electrical activity by blocking overactive presynaptic channels, thus normalizing ion concentrations (Epilepsy Foundation 2016). It is important to note that AEDs do not cure, but reduce seizures for whichever type of epilepsy an individual has (Epilepsy Foundation 2016).

Current AEDs for calcium channels are Zonisamide, Valproate, Phenytoin, and Mibebradil. However, Phenytoin targets Cav3.2 directly but has inefficient results. Current AEDs for temporal lobe epilepsy (TLE) are gabapentin, topiramate, lamotrigine, levetiracetam, oxcarbazepine, and zonisamide. In regards to TLE and sodium channel Nav1.6, the complication is whether the channel is targeted. For EOE and Kv7.2 potassium channel, there are difficulties as the AEDs [retigabine, ICA-27243, SF0034, and others] are not effective enough (Devaux et al., 2016). Since there is still disagreement regarding whether chloride channels are linked to epilepsy, current and new possible treatments are still being studied.

Future

One possible novel treatment for Cav3.2 potassium channel is T-type calcium channel antagonists which would suppress seizures from occurring by blocking the channel, reducing the burst of firing (Powell et al., 2014). Gastrodin is a phenolic glucoside which has anticonvulsant effects (Shao et al., 2017). Its positive effects have been reported in different animal models, but the mechanism of its anticonvulsant features remains unknown (Shao et al., 2017). Nav1.6 channels may be one avenue for Gastrodin, as it resulted in reducing the generation of bursting spikes (Shao et al., 2017). Linopirdine is a potential treatment for Kv7.2 because it can inhibit the potassium current (Devaux et al., 2016). With Kv7.2 mutant and its gain of function, inhibiting the potassium current, in theory would reduce epileptic characteristics (Devaux et al., 2016).

Postsynaptic treatments:

Current

Some of the current treatments target the increase of GABA potentiation. This includes drugs such as Vigabatrin and stiripentol. Other drugs, like perampanel (AMP A antagonist) and Striopenol (block Na+ channel), target ionotropic receptors such sodium channels or AMPA receptors. Topiramate, an anticonvulsant, increases GABA potentiation, AMPA and sodium channel inhibition (Schmidt & Schacter, 2014).

Future

One possible treatment is the use of positive allosteric modulators of NMDAR receptor. It binds to a type of mutant NMDAR, which has increased glutamate efficiency and modifies its protein structure. This modification leads to a rescue of NMDAR function and decreases depolarization in postsynaptic neuron (Addis et al. 2017). CAMKII inhibitors prevent CAMKII from activating sodium ion channels. Also, a ketogenic diet contains a fatty acid molecule capable of being an AMPAR antagonist. Blocking the AMPAR prevents sodium ion influx (Rogowski et al., 2016 & Chang et al., 2016). Overall, the prevention of sodium influx can help to reduce action potentials in postsynaptic neurons.

Astrocytic treatments:

Current

Astrocytic ion channels have not been the target of current treatments for epilepsy since these mechanisms of dysfunction have only recently been discovered.

Future

Some papers have suggested that astrocytes could be targeted in a variety of ways to reduce the debilitating effects of epilepsy. The drug bemetamide, a diuretic, was found to inhibit the astrocytic Na+/K+/Cl- cotransporter (Thrane et al., 2013). This could help decrease the K+ concentration outside the astrocyte, reducing the likelihood that inhibitory synaptic pathways will be muted. Another promising source of treatment is the use of drugs that inhibit the enzyme lactate dehydrogenase in astrocytes (Sada et al., 2015). This manipulation hyperpolarizes neurons close to the astrocytes that shuttle lactate, decreasing the likelihood of seizures. Other sources have suggested that by treating astrocytes with...
Figure 2. Treatments and their synaptic targets. For presynaptic neurons, gastrodin could decrease Nav1.6 currents (A), linopiridine could restore K+ currents (B), and Ca2+ T-type channel antagonists could block burst firing from the thalamus (C). In postsynaptic neurons, targeting overactive NMDA receptor with an allosteric modulator (D), CAMKII inhibitors (E), or AMPAR antagonists (F) decreases sodium influx, making postsynaptic neuron less excitable. In astrocytes, bumetanide could inhibit the Na+ /K+ /Cl- cotransporter (G), lactate dehydrogenase inhibition could decrease the synaptic concentration of K+, and optogenetic treatment of the cell could decrease the frequency of action potentials from nearby neurons.

light (optogenetics), it may be possible to restore the normal concentration of K+ outside astrocytes (Ji et al., 2015). These advances show that targeting dysfunctional ion channels throughout the synapse could yield considerable progress in the treatment of epilepsy.

Conclusion
Patients with epileptic disorders suffer from debilitating seizures. The underlying mechanisms that involve dysfunctional ion channels that lead to hyperexcitability and hypersynchronous firing involve ion channel dysfunctions. These channels are dispersed throughout the synapse - on presynaptic and postsynaptic neurons, as well as on astrocytes. The channels are either part of the glutamate or GABA system, and they account for many of the variations in the epileptic spectrum. By targeting these channels with innovative treatments, the factors that lead to increased action potential frequency and seizures could potentially be reversed.

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