Astrocyte-neuron circuits in epilepsy

AUTHOR(S)

Benton S Purnell, Mariana Alves, Detlev Boison

CITATION

Purnell, Benton S; Alves, Mariana; Boison, Detlev (2023): Astrocyte-neuron circuits in epilepsy. Royal College of Surgeons in Ireland. Journal contribution. https://hdl.handle.net/10779/rcsi.22284793.v1

HANDLE

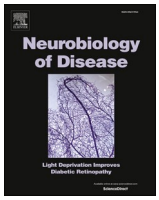
10779/rcsi.22284793.v1

Neurobiology of Disease 179 (2023) 106058

Contents lists available at ScienceDirect

Neurobiology of Disease

journal homepage: www.elsevier.com/locate/ynbdi



Astrocyte-neuron circuits in epilepsy

Benton S. Purnell [a], Mariana Alves [a,b], Detlev Boison [a,c,*]

[a] Department of Neurosurgery, Robert Wood Johnson Medical School, Rutgers University, Piscataway, NJ, United States of America
[b] Department of Physiology and Medical Physics, Royal College of Surgeons in Ireland, Dublin D02 YN77, Ireland
[c] Brain Health Institute, Rutgers University, Piscataway, NJ, United States of America

ARTICLE INFO

Keywords:
Astrocytes
Glia
Metabolism
Epilepsy
Adenosine
Therapy

ABSTRACT

The epilepsies are a diverse spectrum of disease states characterized by spontaneous seizures and associated comorbidities. Neuron-focused perspectives have yielded an array of widely used anti-seizure medications and are able to explain some, but not all, of the imbalance of excitation and inhibition which manifests itself as spontaneous seizures. Furthermore, the rate of pharmacoresistant epilepsy remains high despite the regular approval of novel anti-seizure medications. Gaining a more complete understanding of the processes that turn a healthy brain into an epileptic brain (epileptogenesis) as well as the processes which generate individual seizures (ictogenesis) may necessitate broadening our focus to other cell types. As will be detailed in this review, astrocytes augment neuronal activity at the level of individual neurons in the form of gliotransmission and the tripartite synapse. Under normal conditions, astrocytes are essential to the maintenance of blood-brain barrier integrity and remediation of inflammation and oxidative stress, but in epilepsy these functions are impaired. Epilepsy results in disruptions in the way astrocytes relate to each other by gap junctions which has important implications for ion and water homeostasis. In their activated state, astrocytes contribute to imbalances in neuronal excitability due to their decreased capacity to take up and metabolize glutamate and an increased capacity to metabolize adenosine. Furthermore, due to their increased adenosine metabolism, activated astrocytes may contribute to DNA hypermethylation and other epigenetic changes that underly epileptogenesis. Lastly, we will explore the potential explanatory power of these changes in astrocyte function in detail in the specific context of the comorbid occurrence of epilepsy and Alzheimer's disease and the disruption in sleep-wake regulation associated with both conditions.

1. Introduction

Astrocytes are a type of star-shaped glia whose abundance in the human brain almost reaches that of neurons (von Bartheld et al., 2016). The significance of astrocytes has historically been underappreciated; however, it is now increasingly well understood that astrocytes can substantially augment neuronal activity at the millions of synapses with which they interact (Kimelberg and Nedergaard, 2010; Oberheim et al., 2009). Recent studies have yielded more granular insights into the range of astrocyte diversity based on transcriptomic profiling, immunohistochemistry, and functional read-outs (Chai et al., 2017; Khakh and Sofroniew, 2015; Oberheim et al., 2012). Like neurons, astrocytes are a heterogenous category of cells which vary in critical aspects between neural circuits, developmental time points, and conditions of health and disease (Bayraktar et al., 2014; Ben Haim and Rowitch, 2017). Among their various interrelated physiological functions, astrocytes play a key role in maintaining blood-brain barrier structure and permeability properties by directly interacting with endothelial cells and pericytes (Abbott et al., 2006). They mediate neurovascular coupling (Petzold and Murthy, 2011), and are involved in blood flow regulation and energy metabolism (Boison and Steinhauser, 2018; Koehler et al., 2009), thus providing metabolic support to neurons. They also influence the pH and concentrations of ions in the extracellular space (David et al., 2009; Deitmer et al., 2019). As part of the tripartite synapse, astrocytes regulate extracellular concentrations of neurotransmitters and release signaling molecules of their own in the form of gliotransmission (Halassa et al., 2007). Even the size of the extracellular space itself is largely dictated by the dilation and contraction of astrocytes (Haj-Yasein et al., 2012; Yao et al., 2008).

Epilepsy is a highly prevalent set of disorders characterized by

* Corresponding author at: Department of Neurosurgery, Robert Wood Johnson Medical School, Rutgers University, 683 Hoes Lane West, Piscataway, NJ 08854, United States of America.
E-mail address: detlev.boison@rutgers.edu (D. Boison).

https://doi.org/10.1016/j.nbd.2023.106058
Received 19 December 2022; Received in revised form 20 February 2023; Accepted 27 February 2023
Available online 1 March 2023

spontaneous periods of excessive synchronous neuronal activity in the form of seizures (Sander, 2003; Sander and Shorvon, 1996). Broadly, epilepsy is caused by an imbalance between inhibitory and excitatory signaling in favor of the later (McCormick and Contreras, 2001; Staley, 2015). Despite the broadening array of clinically available anti-seizure medications, rates of drug resistant epilepsy have remained stable for decades (Kwan and Brodie, 2006). Improving our capacity to treat patients with epilepsy may necessitate the identification of different drug targets and novel approaches to pharmacotherapy. The development of most currently available drugs has focused on neurons. Perhaps, the development of astrocyte-based epilepsy therapies will reduce the rate of refractory epilepsy where neuron-focused approaches have failed.

In normal healthy conditions, astrocytes are in a constant state of active participation in the function of neural circuits; however, various pathologies and insults to the central nervous system shift their function to an 'activated state' through the process of reactive astrogliosis (Halassa et al., 2007). Astrogliosis is a complex and dynamic cell response to a variety of pathological conditions which involves a spectrum of genetic, epigenetic, molecular, metabolic, morphological, and functional changes that are context-dependent and regulated by specific signaling events (Halassa et al., 2007; Sofroniew, 2009; Zamanian et al., 2012). Reactive astrogliosis is a hallmark of the epileptic focus both in human epilepsy and animal models (Binder and Steinhauser, 2006; Binder and Steinhauser, 2021; Devinsky et al., 2013). The multiple mechanisms through which astrocytes are linked to epilepsy are summarized in Fig. 1 and will be discussed in more detail in subsequent sections of this article.

The significance of astrocytes and astrogliosis in epilepsy is the subject of intense empirical investigation. Our general knowledge in this area has expanded so rapidly that it is useful to step back and consider its implications in specific contexts. The purpose of this review is (1) to summarize and discuss the multifarious roles played by astrocytes in neural circuits of the epileptic brain in general and (2) to consider the explanatory power of astrocytic dysfunction in the interplay between epilepsy, related neurological disorders with a glial pathology, and their associated comorbidities. We will present the case for the hypothesis that astrocytes contribute to the frequent comorbid occurrence of epilepsy and Alzheimer's disease and the disruption in sleep-wake regulation associated with both conditions.

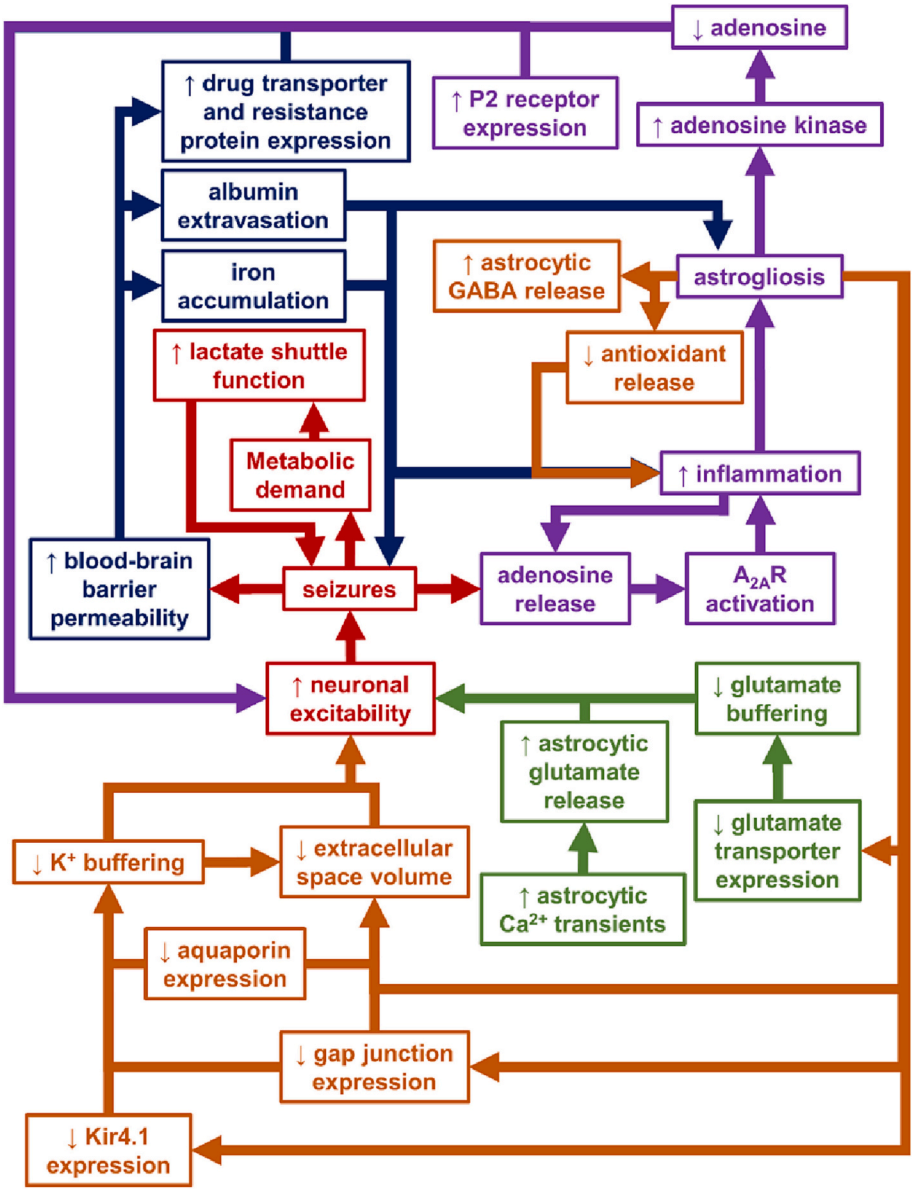


Fig. 1. Schematic diagram depicting the mechanistic interplay in astrocytic function in epilepsy. Mechanistically linked alterations in astrocytic function have been color coded. Red highlights seizures and the increased neuronal excitability that precipitates them. Purple highlights purine related changes. Orange highlights the sequalae of astrogliosis beyond its implications for adenosine signaling. Green highlights alterations in glutamatergic gliotransmission. Blue highlights the consequences of seizure-induced blood-brain barrier dysfunction. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

2. Gliotransmission and the tripartite synapse

Neurons and their synapses do not operate in isolation. Astrocytes, through their elaborate cellular processes, are tightly associated with neuronal synapses, and it is generally agreed that each astrocyte could associate with over 100,000 synapses (Bushong et al., 2002). Revelations regarding the intricate interactions between synapses and astrocytic processes led to the concept of the "tripartite synapse" through which astrocytes have the unique capability to alter the contents of the synaptic space by reuptake of neurotransmitters and release of their own gliotransmitters and thereby to modulate and fine-tune the activity of neurons (Araque et al., 1999). Several gliotransmitters are of importance within the context of epilepsy and may offer opportunities for therapeutic intervention: (1) Astrocytes express all the necessary components for vesicular glutamate release and, indeed, Ca^{2+-}dependent release of glutamate-laden vesicles has been documented in vitro and in vivo (Bezzi et al., 2004; Bohmbach et al., 2018; Nedergaard, 1994; Parpura et al., 1994). Through the activation of neuronal glutamate receptors, astrocytes can directly lower the threshold for ictal activity (Gomez-Gonzalo et al., 2010). Accordingly, intracortical injection of the chemoconvulsant kainic acid increases astrocytic Ca^{2+} levels seconds prior to the onset of seizures (Heuser et al., 2018). Conversely, pharmacological, or genetic approaches to inhibition of astrocytic transmitter release were shown to interfere with the ictogenic and epileptogenic processes (Clasadonte et al., 2013; Riquelme et al., 2020). (2) In addition, astrocytes can control long-term potentiation through the release of the *N*-methyl-D-aspartate receptor (NMDAR) co-agonist D-serine (Henneberger et al., 2010; Papouin et al., 2017). This astrocytic regulation of synaptic plasticity has important implications for learning and memory in conditions of health and for network excitability in epilepsy. (3) ATP is an important astrocyte derived gliotransmitter (Pascual et al., 2005), which can either directly activate pre- and postsynaptic P2 receptors, or – after its sequential dephosphorylation to adenosine – lead to the activation of adenosine receptors, among which the adenosine A_1 receptor (A_1R) mediates the antiseizure effects of adenosine, which is an endogenous seizure terminator (Beamer et al., 2021; Dragunow, 1991; During and Spencer, 1992). In general, ATP and adenosine play opposing roles in epilepsy (Beamer et al., 2021). Extracellular adenosine exerts anticonvulsive and neuroprotective effects by acting on pre- and postsynaptic A_1Rs, whereas the activation of P2 receptors by increased extracellular ATP promotes seizures and the development of epilepsy (Beamer et al., 2021). Accordingly, in epilepsy patients and animal models there is increased P2 receptor expression (Engel et al., 2016; Jimenez-Pacheco et al., 2013; Vianna et al., 2002). Based on its role as an endogenous anticonvulsant, local adenosine augmentation therapies have been developed in experimental models to selectively augment adenosine signaling in the vicinity of the seizure focus (Boison, 2012). Importantly, adenosine has additional, adenosine receptor independent, antiepileptogenic and disease modifying properties as will be discussed in more detail below. Inhibition of astrocytic ATP release through the FDA-approved Panx1 channel blockers probenecid or mefloquine suppressed epileptiform activity in resected cortical tissue from epilepsy patients and decreased seizures in a mouse model of acquired epilepsy, highlighting the proconvulsant roles of ATP (Dossi et al., 2018). (4) Finally, astrocytes can also release GABA, which generates hyperpolarizing currents and tonic inhibition (Yoon and Lee, 2014). At least two studies demonstrate that in rodent models of temporal lobe epilepsy reactive astrocytes overproduce and release GABA, which then activates tonic $GABA_AR$-mediated currents in excitatory neurons and thereby increases seizure thresholds (Muller et al., 2020; Pandit et al., 2020). This increase in astrocytic GABA signaling may be a compensatory mechanism which counterbalances the reduction in GABAergic signaling due to loss of inhibitory interneurons (Muller et al., 2020; Pandit et al., 2020).

The examples enumerated above demonstrate first that aberrant gliotransmission plays an important role in the pathophysiology of epilepsy, and second that gliotransmission is a promising target for therapeutic intervention. In line with this conclusion, commonly used antiseizure medications including phenytoin, valproic acid, and gabapentin suppress astrocytic Ca^{2+} signaling during seizures implying that part of the therapeutic benefits of those agents is mediated by the inhibition of gliotransmission (Tian et al., 2005). The therapeutic targeting of maladaptive gliotransmitter release might be a strategy to modify the etiology of epilepsy and to prevent its genesis and progression (Beamer et al., 2021; Riquelme et al., 2020; Vezzani, 2022; Williams-Karnesky et al., 2013).

2.1. Blood-brain barrier dysfunction

The blood-brain barrier is an actively maintained partition, the selective permeability of which is critical to maintaining the healthy function of the central nervous system (Daneman and Prat, 2015). Generally, the blood-brain barrier passively permits the passage of small lipophilic molecules; however, it is also capable of actively transporting molecules to the brain that would not have passively diffused and metabolizing or pumping out molecules that would have passively diffused into the brain (Abbott, 2013; Loscher and Friedman, 2020). The blood-brain barrier is comprised of an inner layer of tightly joined endothelial cells which envelop capillaries in the brain. These endothelial cells are in turn surrounded by pericytes and the end-feet projections of astrocytes (Daneman and Prat, 2015).

It has long been appreciated that seizures increase the permeability of the blood-brain barrier (Bauer and Leonhardt, 1956; Lorenzo et al., 1975). However, this increased permeability is not a general broadband effect and may be more pronounced for some molecules than others (Kang et al., 2013). Seizures can cause pathophysiological alterations in perivascular astrocytes (Friedman et al., 2009) likely due to the penetrance from the blood into the brain of substances which ordinarily would have been excluded. Perhaps the most thoroughly characterized substance to inappropriately enter the brain during seizures and adversely affect astrocytes is albumin.

Albumin protein is abundantly found in the blood and the deleterious effects of its extravasation into the brain during seizures have been well documented. Albumin extravasation has been observed in epilepsy patients and animal models (Seiffert et al., 2004; van Vliet et al., 2007a). Blood-brain barrier dysfunction or the direct application of albumin to the brain causes astrogliosis, the development of an epileptic focus, and reductions in the clearance of extracellular glutamate and potassium (David et al., 2009; Seiffert et al., 2004). The uptake of albumin into astrocytes is mediated by transforming growth factor β (TGF-β) receptor activation (Ivens et al., 2007). Activation of the TGF-β signaling pathway has been implicated in epileptogenesis and blocking this pathway is therapeutically beneficial in animal models (Cacheaux et al., 2009; Ivens et al., 2007). Furthermore, albumin exposure increases the release of matrix metalloproteinase 9 via activation of the mitogen-activated protein kinase (MAPK) signaling pathway (Ralay Ranaivo et al., 2012). Matrix metalloproteinase 9 has been implicated in epileptogenesis due to its role in seizure-induced cell death and dendritic spine morphology (Kim et al., 2009; Michaluk et al., 2011).

Increased albumin uptake by astrocytes as a result of blood-brain barrier disfunction and the resulting signaling cascade can reduce K^+ buffering capacity and increase neuronal excitability via a decrease in Kir4.1 channels, AQP4 aquaporin channels, glutamate transporters, and an impairment in astrocytic gap junction coupling (Braganza et al., 2012; David et al., 2009; Ivens et al., 2007). The functional significance of many of these changes beyond what is directly pertinent to perivascular astrocytes will be discussed in further detail in other subsections.

In temporal lobe epilepsy patients, AQP4 expression is reduced in the perivascular end feet relative to healthy comparison tissue (Eid et al., 2005). This disruption in AQP4 localization is the result of decreased dystrophin in the perivascular end feet, which normally anchor AQP4

channels (Eid et al., 2005). In a rat seizure model, reduced AQP4 expression colocalizes with blood-brain barrier dysfunction (Bankstahl et al., 2018). As will be discussed in more detail in the subsections on water and K^+ homeostasis, loss of aquaporin expression is significant, as water influx through these channels is thought to be necessary for concomitant K^+ buffering (Holthoff and Witte, 1996; Strohschein et al., 2011). In line with this understanding, a transgenic manipulation, which caused reduced AQP4 in the perivascular end feet of mice, impaired K^+ clearance following neuronal activity and caused more severe seizures during a hyperthermic challenge (Amiry-Moghaddam et al., 2003).

In addition to the potential role of astrocyte dysfunction in the excessive permeability of the blood-brain barrier during seizures, pathological changes in perivascular astrocytes may adversely affect penetrance of anti-seizure drugs into the brain by overexpressing multidrug transporters and multidrug resistance proteins (Aronica et al., 2012b; Lazarowski et al., 2007; Tishler et al., 1995; van Vliet et al., 2005). These changes have been documented in the resected tissue of human epilepsy patients (Tishler et al., 1995). Studies using inducible animal models indicate that the increase in these proteins is the result of the epileptiform activity (Hoffmann et al., 2006; Marchi et al., 2006) as opposed to being a risk factor that predisposes the brain to epilepsy. Studies using animal models have also indicated that these multidrug transporters and multidrug resistance proteins meaningfully alter the concentration of peripherally administered anti-seizure medications in the brain (van Vliet et al., 2007b; van Vliet et al., 2010). Additionally, inhibition of the MAPK signaling pathway in animal models decreases the expression of multidrug transporters and increases the penetrance of anticonvulsant drugs into the brain (Shao et al., 2016).

To summarize, epilepsy and seizures impair blood-brain barrier function allowing harmful materials from the blood into the brain. Perivascular astrocytes are a crucial physical component of the blood-brain barrier, but in epilepsy their function is adversely affected by: (1) overexpression of multidrug transporters and multidrug resistance proteins; (2) decreased glutamate transporter expression and consequent impaired glutamate buffering; (3) a reduction in perivascular AQP4 channels and consequent impairments in K^+ buffering; (4) decreased inward rectifying potassium channel expression causing further impairment in K^+ buffering; and (5) impaired astrocytic gap junction coupling causing yet further impairment of K^+ buffering.

2.2. Inflammation and oxidative stress

Injuries and insults to the brain as varied as traumatic brain injury, stroke, status epilepticus, and viral infection can trigger a surprisingly uniform sequence of events contributing to the development of epilepsy (Klein et al., 2018). Those changes include neuroinflammation, oxidative stress, microglial, and astrocytic activation, culminating in astrogliosis, a pathological hallmark of temporal lobe epilepsy (Pauletti et al., 2019; Terrone et al., 2020; Vezzani, 2022). In this section we focus on mechanisms of astrocyte activation and bi-directional interactions between astrocytes, neuroinflammation, and oxidative stress. In human and experimental epilepsy, it has been shown that astrocytes can produce and release cytokines and chemokines in immunologically relevant concentrations (Aronica et al., 2012a; Devinsky et al., 2013; Vezzani et al., 2019). On the other hand, an inflammatory phenotype in astrocytes can be triggered by a variety of cytokines, chemokines, and danger signals, which can be released by reactive microglia or the microvasculature (Eyo et al., 2017; Liddelow et al., 2017; Ravizza et al., 2008). Acute epilepsy-triggering insults to the brain lead to the release of ATP (Fields, 2011), a major source of adenosine (Yegutkin et al., 2011), of complement factors (Benson et al., 2015; Gruber et al., 2022), prostaglandins (Rojas et al., 2019), and danger signals such as the chromatin associated protein high mobility group box 1 (Maroso et al., 2010; Zurolo et al., 2011), which all contribute to the activation of astrocytes and the induction of an inflammatory phenotype. In particular, increased $A_{2A}R$ activation through excessive injury-induced adenosine production promotes neuro-inflammatory processes and neurodegeneration (Rebola et al., 2011) along with microglial (Madeira et al., 2018) and astroglial (Brambilla et al., 2003) activation. Those processes all contribute to epileptogenesis (Klein et al., 2018). Once activated, altered astrocyte function plays a key role in the reduction of seizure thresholds (Li et al., 2007a; Sano et al., 2021) and the development of epilepsy (Li et al., 2008a; Li et al., 2008b).

Because neuroinflammation and oxidative stress are intricately linked, there is a strong rationale for the astrocyte-mediated redox-based control of hyperexcitability in epilepsy. Astrocytes produce the antioxidant glutathione, and the astrocyte-to-neuron glutathione shuttle replenishes neuronal glutathione pools. This is a major antioxidant defence mechanism of the brain, which rescues mitochondrial function during seizures. In addition, increased neuronal activity promotes the activity of nuclear factor erythroid 2-related factor 2 (Nrf2) in astrocytes and thereby activates anti-inflammatory, antioxidant, and cytoprotective pathways (Habas et al., 2013). Therapeutic activation of the Nrf2 pathway or replenishing the glutathione pool with *N*-acetylcysteine might be promising therapeutic approaches to alter the redox status in epilepsy and thereby increase neuroprotection, decrease seizures, and inhibit epileptogenesis and the development of cognitive comorbidities (Pearson-Smith and Patel, 2017; Terrone et al., 2017; Vezzani et al., 2019). Furthermore, interference with the aforementioned inflammatory cytokine high mobility group box 1 using monoclonal antibodies has shown therapeutic potential in several electrical and chemical animal models of epilepsy and in resected tissue from human patients (Zhao et al., 2017; Zhao et al., 2020).

2.3. The astrocyte to neuron lactate shuttle

Neuronal networks increase their demand for energy during epileptic activity. This energy deficiency is compensated by a stimulation of glycolysis in astrocytes, which triggers a rapid decrease in intracellular glucose concentrations and a simultaneous increase in the metabolic product pyruvate. Pyruvate is then transformed into lactate through the enzyme lactate dehydrogenase. Through this sequence of metabolic reactions lactate, formed in astrocytes, becomes a major energy source to sustain the energy demands of hyperactive neuronal networks. This energy transfer process is enabled by an astrocyte-to-neuron lactate shuttle (Pellerin and Magistretti, 1994). In line with the notion that seizure-induced lactate formation promotes epileptic seizures, lactate dehydrogenase inhibitors, such as the FDA-approved drug stiripentol, provide potent anticonvulsant effects (Sada et al., 2015). Stiripentol, by blocking lactate dehydrogenase, is uniquely suited to dampen glycolysis by reducing the availability of NAD^+ and by promoting the oxidative mitochondrial metabolism of pyruvate as an incoming metabolite to support Krebs cycle activity. This hypothesis has been challenged by findings suggesting that both astrocytic glycolysis as well as the astrocyte-to-neuron lactate shuttle depend on AMP activated protein kinase (AMPK) in astrocytes (Muraleedharan et al., 2020), which acts as a major energy sensor. In line with those findings, AMPK knockout mice were characterized by a depletion of lactate and reduced seizure propensity; however, the cell-type selective disruption of AMPK in astrocytes promoted neurodegeneration (Muraleedharan et al., 2020). While these findings appear to be contradictory, they demonstrate the complexity of astrocyte metabolism in the regulation of lactate and brain energy homeostasis.

2.4. Gap junction coupling

In healthy conditions, astrocytes are connected by specialized pore forming transmembrane proteins that link their intracellular space called gap junctions (Fischer and Kettenmann, 1985; Lee et al., 1994). The cytosolic connections afforded by gap junctions provide an opportunity for the rapid passage of electrical currents, ions, and small molecules (Giaume et al., 1997; Wallraff et al., 2006). Gap junctions have

been implicated in astrocytic water homeostasis and, by proxy, extracellular space volume dynamics (Pannasch et al., 2011). Astrocytic connections via gap junctions increase their capacity to provide metabolic resources to neighboring neurons, which is ostensibly adaptive under normal conditions, but may prolong seizures (Giaume et al., 1997; Rouach et al., 2008).

Transgenic downregulation of astrocyte coupling via gap junctions in mice marginally impairs K^+ buffering, increases vulnerability to seizure activity, decreases glutamate clearance, and increases astrocytic swelling (Pannasch et al., 2011; Wallraff et al., 2006). Observations of the expression of gap junctions in resected human epileptic tissue have generated mixed results with some studies reporting no change (Elisevich et al., 1997b), other studies reporting increases (Aronica et al., 2001; Fonseca et al., 2002; Naus et al., 1991), and yet others reporting a mix of up and downregulation depending on the specific protein in question (Collignon et al., 2006). Similarly inconsistent observations have been made in animal models with decreased (Elisevich et al., 1997a; Xu et al., 2009), increased (Condorelli et al., 2002; Takahashi et al., 2010), or unchanged gap junction expression (Khurgel and Ivy, 1996) all having been observed depending on experimental parameters and the specific protein being examined.

The mixed results pertaining to gap junction expression in epilepsy patients and experimental models is likely a function of the disparate effects that these channels might have on tissue excitability. On the one hand, gap junction expression is thought to contribute to normal astrocytic K^+ buffering and volume compensation (Pannasch et al., 2011; Wallraff et al., 2006). Unchecked increases in extracellular K^+ and decreases in the volume of the extracellular space consequent to gap junction downregulation would be expected to increase excitability. On the other hand, gap junctions may also increase astrocytic Ca^{2+} signaling and their capacity to provide metabolic support for local neurons (Fujii et al., 2017; Rouach et al., 2008). Decreased propagation of Ca^{2+} waves and reduced neuronal energetic support consequent to gap junction downregulation would be expected to decrease excitability. Additional experimentation is needed to further elucidate the adaptive and pathological roles of gap junctions in different epilepsy subtypes.

2.5. Dysregulation of water homeostasis

Aquaporins are membrane bound channels that allow the transport of water molecules across cell membranes in the direction of osmotic gradients (Verkman, 2009). AQP4 is the most abundantly expressed of the aquaporin channels in glial cells of the central nervous system (Mader and Brimberg, 2019; Papadopoulos and Verkman, 2013). Expression of AQP4 is most concentrated in glial cells adjacent to blood vessels along with the subarachnoid and ventricular spaces (Hubbard et al., 2015). Hydration status influences seizure susceptibility with rapid increases in water content being proconvulsant and dehydration being anticonvulsant (Andrew, 1991; Andrew et al., 1989).

There is a net increase in AQP4 expression in the sclerotic hippocampi of human epilepsy patients (Eid et al., 2005; Lee et al., 2004). Decreases in AQP4 expression have been reported in other epilepsy subtypes (Kandratavicius et al., 2015; Lapato et al., 2020). Closer examination reveals that these disparate changes in net AQP4 expression belie a consistent focal decrease in AQP4 in perivascular end feet where they are normally enriched (Alvestad et al., 2013; Eid et al., 2005; Lapato et al., 2020). Model dependent increases and decreases in AQP4 expression have been observed in animal models of seizures and epilepsy (Hubbard et al., 2016; Lee et al., 2012; Szu et al., 2020a); however, as with human tissue, decreased AQP4 localization on astrocytic end feet is a consistent feature (Lee et al., 2012; Szu et al., 2020a). As was discussed in greater detail above, this decrease in astrocytic perivascular AQP4 may have consequences for local blood-brain barrier permeability (Bankstahl et al., 2018). The primary functional significance of altered aquaporin expression in the context of epilepsy and seizures is related to the effect of these channels on K^+ buffering which will be discussed in detail in its own subsection (Holthoff and Witte, 1996; Strohschein et al., 2011).

The passage of water through AQP4 channels and the consequent contraction or expansion of astrocytes can alter the volume of the extracellular space (Haj-Yasein et al., 2012; Yao et al., 2008). Seizures themselves reduce the volume of the extracellular space (Lux et al., 1986; Tonnesen et al., 2018). Changes in the volume of the extracellular space alter neuronal excitability with decreases in the extracellular space being proconvulsant (Chebabo et al., 1995; Kilb et al., 2006; Schwartzkroin et al., 1998). AQP4 itself is not necessary for activity dependent decreases in the volume of the extracellular space (Colbourn et al., 2021; Haj-Yasein et al., 2012; Toft-Bertelsen et al., 2021); however, activity dependent contraction of the extracellular space may be compounded by tonic alterations to the extracellular space due to altered AQP4 expression in astrocytes to aggravate and prolong seizure activity.

Characterization of mice which lack AQP4 has generated interesting and somewhat disparate results. Mice lacking AQP4 channels display an increased seizure threshold in electrical stimulation (Binder et al., 2006) and chemoconvulsant (Binder et al., 2004) seizures models. These mice also exhibit an increase in evoked (Binder et al., 2006) and spontaneous (Szu et al., 2020b) seizure duration. Furthermore, aquaporin deficient mice subjected to PTZ exposure following traumatic brain injury display a decreased latency to seizures and an increase in seizure severity (Lu et al., 2021).

Transient cerebral edema is observed following status epilepticus in humans (Liu et al., 2021; Sammaritano et al., 1985) and in animal models (Kim et al., 2010; Nelson and Olson, 1987). Edema following status epilepticus is exacerbated in mice with transgenic downregulation of AQP4 (Lee et al., 2012), a finding consistent with other observations in these mice which are indicative of a general impairment in the clearance of tissue water (Bloch et al., 2005; Papadopoulos et al., 2004).

To summarize, astrocytes abundantly express AQP4 channels which are essential to their role in maintaining water homeostasis. In both human patients and epilepsy models, alterations in AQP4 of varying directionality are commonly observed; however, decreased localization of AQP4 on astrocytic end feet is quite consistent. Altered AQP4 expression has functional implications in the resolution of cerebral edema following prolonged seizure activity, the volume of the extracellular space, blood-brain barrier permeability, and K^+ buffering.

2.6. Dysregulation of ion homeostasis

2.6.1. Potassium

The Na^+/K^+-ATPase pumps Na^+ ions out of neurons in exchange for K^+ ions thereby progressively reversing the changes in ionic balance induced by neuronal activity; however, this process in neurons alone is not adequate for timely clearance of extracellular K^+, particularly during seizures (Heinemann et al., 1977; Hodgkin and Huxley, 1952). Astrocytes regulate extracellular K^+ levels by (1) direct K^+ uptake from the extracellular space and (2) K^+ spatial buffering within astrocytes linked by gap junctions (Amédée et al., 1998; Kofuji and Newman, 2004). Astrocytes, like neurons, also express Na^+/K^+-ATPases; however, due to a difference in subunit composition (Cameron et al., 1994; McGrail et al., 1991), the function of Na^+/K^+-ATPases in astrocytes is enhanced by increased extracellular K^+ concentrations (Henn et al., 1972; Hertz and Chen, 2016). As a result, astrocytes have a superior capacity to rapidly take up and sequester extracellular K^+ in comparison to adjacent neurons (Henn et al., 1972; Walz and Hertz, 1983). Additionally, $Na^+/K^+/Cl^-$ cotransporters pump $Na^+/K^+/Cl^-$ into the intracellular space of cells, including astrocytes (MacVicar et al., 2002; Walz, 1992). Lastly, inwardly rectifying K^+ channels, specifically Kir4.1, allow the inward and outward passage of K^+ ions with a preference towards inward flow (Doupnik et al., 1995; Kofuji and Newman, 2004). K^+ ions taken up from the extracellular space can be transferred between astrocytes connected by gap junctions in a process called spatial buffering (Holthoff and

Witte, 2000; Kofuji and Newman, 2004). K^+ that was taken up by astrocytes is slowly released back into the extracellular space through Kir4.1 channels (Bay and Butt, 2012).

Impairments in K^+ homeostasis have been observed in chemoconvulsant (Gabriel et al., 1998) and traumatic brain injury (D'Ambrosio et al., 1999) models of epilepsy as well as in tuberous sclerosis complex (Xu et al., 2009). Transgenic downregulation of gap junction expression impedes spatial K^+ buffering and increases susceptibility to seizures in mice (Wallraff et al., 2006). Reduced Kir4.1 expression decreases the capacity of astrocytes to take up K^+ and glutamate from the extracellular space in vitro and in vivo (Djukic et al., 2007; Kucheryavykh et al., 2007). Astrocyte specific deletion of Kir4.1 causes spontaneous seizures (Djukic et al., 2007). Blood-brain barrier disruption or direct application of albumin to the brain reduces Kir4.1 expression and impedes K^+ buffering prior to the development of seizures (Ivens et al., 2007). Electrophysiological investigations of excised tissue samples from human patients have revealed a downregulation in inwardly rectifying K^+ currents in epileptic foci (Bordey and Sontheimer, 1998; Hinterkeuser et al., 2000; Kivi et al., 2000). Most, but not all, available evidence suggests that downregulation of Kir4.1 channels contributes to epilepsy. However, there are reports of increased Kir4.1 expression (Nagao et al., 2013) and a lack of any change in Kir dependent currents (Takahashi et al., 2010) in rodent models of epilepsy brought about by prior chemoconvulsant-induced status epilepticus.

Given the diminished capacity for K^+ buffering observed in epilepsy and the ictogenic effects of experimental interference with K^+ buffering, interventions to improve K^+ buffering may be therapeutic. In line with this premise, optogenetic activation of astrocytes suppresses seizures in a Na^+/K^+-ATPase dependent manner following cortical kainic acid injection and in a model of focal cortical dysplasia in rats (Zhao et al., 2022). Similarly, bolstering K^+ buffering suppresses seizures in the well characterized 'Shaker' Drosophila line (Li et al., 2021; Lones and DiAntonio, 2023).

2.6.2. Calcium

Voltage-gated calcium channels (VGCCs) mediate entry of Ca^{2+} into excitable cells (Hofmann et al., 1999) and thereby control the release of neurotransmitters. Not surprisingly, mutations in neuronal VGCCs have been associated with epilepsy (Brill et al., 2004). In astrocytes, calcium transport is mediated by the sodium/calcium exchanger (NCX) (Parpura et al., 2016; Rose et al., 2020). The directionality of the passage of ions through the NCX is dependant on ion concentrations and the membrane potential of the astrocyte (Khananshvili, 2014). Generally, NCX function is thought to facilitate seizures given the observation that pharmacological inhibition of NCX function (Saito et al., 2009) and downregulation of NCX expression both have anticonvulsant effects (Akinfiresoye et al., 2023; Newton et al., 2021; Saito et al., 2009). Experiments utilizing pharmacological inhibition specific to the reverse function of the NCX (in which Ca^{2+} enters the cell) indicate that it is this aspect of NCX function which confers its proconvulsant properties (Akinfiresoye et al., 2023; Martinez and N'Gouemo, 2010).

Alterations in astrocytic Ca^{2+} signaling have been implicated both in the initiation of seizures as well as the progression of epileptogenesis. Ca^{2+} imaging experiments indicate that increases in astrocytic Ca^{2+} precede seizure activity in the cortex and in CA1 of the hippocampus in mice and in the developing nervous systems of zebrafish larvae (Diaz Verdugo et al., 2019; Heuser et al., 2018; Tian et al., 2005). It has been demonstrated using photolytic Ca^{2+} uncaging within astrocytes in rodent hippocampal slices that intracellular astrocytic Ca^{2+} signaling elicits slow inward depolarizing currents in adjacent neurons (Fellin et al., 2004). Subsequent experimentation using a similar astrocytic Ca^{2+} uncaging approach suggests that these neuronal slow depolarizing shifts are mediated by Ca^{2+} dependant glutamate release (Tian et al., 2005).

In addition to periictal astrocytic calcium signaling contributing to excitatory gliotransmission, more chronic changes in astrocyte signaling may contribute to epileptogenesis. Chemoconvulsant-induced status epilepticus increases the astrocytic Ca^{2+} transients elicited by the activation of local neurons (Szokol et al., 2015). Similarly, the astrocytes of mice subjected to chemoconvulsant-induced status epilepticus have increased ambient Ca^{2+} signaling for days after the initial insult (Ding et al., 2007). These increases in astrocytic Ca^{2+} are correlated with neuronal cell death and this cell death can be counteracted by pharmacological interference with astrocytic glutamate signaling (Ding et al., 2007). Generally, hypertrophic activation of astrocytes increases their Ca^{2+} signaling and, correspondingly, astrocyte atrophy is associated with weaker Ca^{2+} signaling (Plata et al., 2018; Shigetomi et al., 2019).

2.6.3. Iron

Increased iron deposition in the brain is observed in experimental models and in epilepsy patients (Gorter et al., 2005; Zimmer et al., 2021). The increased accumulation of iron in the brain associated with seizures and epilepsy is hypothesized to be the result of iron rich proteins in the blood crossing into the brain following seizure-induced blood-brain barrier dysfunction (van Vliet et al., 2020; Zimmer et al., 2021). Iron deposition in the brain may contribute to epileptogenesis as the introduction of exogenous iron into the brain triggers spontaneous recurrent seizures (Sharma et al., 2007; Willmore et al., 1978). Neurons, microglia, and astrocytes are all capable of taking up iron from the extracellular space (Rouault, 2013; Zimmer et al., 2021). Human astrocyte cell cultures that are exposed to iron increase expression of antioxidant and iron handling genes; however, chronic exposure results in pro-inflammatory changes (Zimmer et al., 2021). Iron accumulation in cells increases reactive oxygen species within the cell and can result in cell death in the form of ferroptosis (Li et al., 2020). Tissue resected from patients with temporal lobe epilepsy or taken post-mortem from patients who died of status epilepticus display increased astrocytic iron sequestration and a shift in iron uptake from microglia to astrocytes (Zimmer et al., 2021).

2.7. Dysregulation of glutamate and glutamine metabolism

Glutamate, a ubiquitous biological molecule, has important roles in the central nervous system in the synthesis of proteins, as a source of energy, and as a neurotransmitter (Mahmoud et al., 2019). In its most discussed role, glutamate is the most common excitatory neurotransmitter (Meldrum, 2000). However, accumulation in the extracellular space of glutamate released from excitatory neurons can be highly neurotoxic (Lau and Tymianski, 2010). Although multiple cell types are involved in extracellular glutamate removal, astrocytes are the most efficient in this process, removing almost 90% of all glutamate released (Mahmoud et al., 2019). Astrocytes maintain glutamate homeostasis by the release or the uptake of glutamate from the synaptic cleft thereby fine-tuning neuronal function and preventing glutamate excitotoxicity (Mahmoud et al., 2019). Therefore, it is important to comprehend how glutamate is regulated during physiological and pathological conditions to establish new strategies to maintain its homeostasis and to prevent the development of diseases that are associated with an increase of excitatory neurotransmission, such as epilepsy.

During physiological conditions, astrocytes take up glutamate in the synaptic cleft through excitatory amino acid transporters. Glutamine synthetase then converts this neuronal glutamate into glutamine (Anlauf and Derouiche, 2013). Glutamine is released from astrocytes into the extracellular space and is used as a precursor for the synthesis of neurotransmitters, such as glutamate or GABA that will be released again during exocytosis events. The remaining glutamate is then metabolized into α-ketoglutarate, which is used as a substrate for ATP production (McKenna, 2013). On the other hand, when glutamate is taken up by astrocytes, it increases intracellular calcium contributing to astrocytic glutamate release. This glutamate will act on different glutamatergic receptors, contributing to balanced neuron-neuron and neuron-glia

interactions (Schousboe et al., 2014).

During pathological conditions, astrocytes enter an activated state through molecular and morphological alterations that affect their basal functions, including glutamate homeostasis. Loss of astroglial glutamate uptake or excessive glutamate release increases the concentration of glutamate in the synapse resulting in excitotoxicity and neuronal hyperexcitability. Administration of glutamate or glutamate analogues into the hippocampus in animal models triggers seizures, but simultaneous injection of glutamate antagonists blocks these seizures (McNamara, 1994; Olney et al., 1972; Pitkanen and Lukasiuk, 2009). Additionally, in patients with mesial temporal lobe epilepsy, extracellular glutamate concentrations are increased during a seizure and remain elevated for several hours after the seizure has ended (During and Spencer, 1993). The accumulation of glutamate can be accentuated by a reduction in the volume of the extracellular space. The swelling of reactive astrocytes can alter the extracellular space volume through the disturbances in the expression of aquaporin-4 water channels or volume-regulated anion channels. Reduction in the volume of extracellular space has been associated with increased hyperexcitability in several forms of epilepsy (During and Spencer, 1993; Tonnesen et al., 2018). Excessive glutamate in the synaptic cleft can also act on NMDA receptors, increasing slow inward currents and neuronal hyperexcitability, or on metabotropic glutamate receptors (mGluRs), which indirectly leads to the activation of NMDARs and neuronal hyperexcitability. Studies performed in both experimental models of epilepsy and in resected tissue from patients with temporal lobe epilepsy indicate that mGluR1 expression is increased in the hippocampus (Blumcke et al., 2000). mGluR1 activation has been also shown to potentiate NMDAR currents through a mechanism involving increased intracellular calcium signaling (Heidinger et al., 2002). Correspondingly, inhibition of mGluR1 decreases PTZ-kindled seizures (Watanabe et al., 2011). Similar observations have been made regarding mGluR5 and mGluR3 (Aronica et al., 2000; Ding et al., 2007). mGluR5, which is expressed in astrocytes, is increased in expression in animal models of epilepsy (Ding et al., 2007; Kelly et al., 2018; Umpierre et al., 2019). Unfortunately, activation of mGluR5 potentiates NMDAR activity by increasing calcium levels, which consequently drives neuronal hyperexcitability (Ding et al., 2007). In contrast, mGluR2/3 is down regulated in a pilocarpine mouse model (Garrido-Sanabria et al., 2008; Pacheco Otalora et al., 2006; Tang et al., 2004), suggesting a neuroprotective role of these receptors when activated in epilepsy models (Kelly et al., 2018; Watanabe et al., 2011).

In summary, astrocytes are crucial to synaptic transmission due to their ability to modulate neuronal firing and maintain a balance between glutamate uptake, metabolism, and release. When their function is disrupted, astrocytes can become active contributors to excessive glutamatergic signaling and may potentiate seizures, excitotoxic cell damage, and epilepsy development.

2.8. Dysregulation of adenosine metabolism

Astrocytes play a key role in regulating concentrations of the brain's own anticonvulsant and seizure terminator, adenosine (Dragunow, 1991; During and Spencer, 1992; Lado and Moshe, 2008). Adenosine is an evolutionary ancient regulator of energy homeostasis (Boison and Yegutkin, 2019; Yegutkin and Boison, 2022). Through expression of the adenosine metabolizing enzyme adenosine kinase, astrocytes form a metabolic sink for the efficient clearance of adenosine (Studer et al., 2006). In line with this role, the genetic disruption of adenosine kinase in embryonic stem cells has been used to engineer implantable glia for the therapeutic delivery of adenosine (Fedele et al., 2004). The inhibitory effect of adenosine in the brain depends predominantly on the activation of G_i protein coupled adenosine A_1 receptors, which can be blocked by the non-selective adenosine receptor antagonists caffeine and theophylline, which therefore can aggravate seizures (Boison, 2011). Maladaptive overexpression of adenosine kinase leads to enhanced metabolic clearance of adenosine through astrocytes and is considered a pathological hallmark of temporal lobe epilepsy (Aronica et al., 2013). Specifically, this overexpression of adenosine kinase is mediated by reactive astrogliosis, and the resulting adenosine deficiency can both trigger seizures (Li et al., 2008b) and contribute to the epileptogenic process (Williams-Karnesky et al., 2013). Therapeutic augmentation of adenosine is effective in the suppression of seizures and also in interference with the epileptogenic process itself (see below for details) (Williams-Karnesky et al., 2013). Therefore, augmented adenosine metabolism in astrocytes affects the pathophysiology of epilepsy broadly and provides unexplored opportunities for metabolism-based therapies (Boison and Rho, 2020; Rho and Boison, 2022).

2.9. Dysregulation of DNA methylation

The role of epigenetic factors in epilepsy and specifically the role of maladaptive DNA methylation changes has received increased attention recently (Kobow and Blumcke, 2018; Murugan et al., 2021; Younus and Reddy, 2017). According to the methylation hypothesis of epileptogenesis, first proposed in 2011, maladaptive changes in DNA methylation drive the processes that contribute to the development and progression of epilepsy (Kobow and Blumcke, 2011; Kobow et al., 2013a). In support of this hypothesis, an increasing number of publications document that epigenetic processes, specifically increased DNA methyltransferase activity and maladaptive DNA methylation, are closely linked to epileptogenesis (Kobow et al., 2009; Kobow et al., 2013b; Martins-Ferreira et al., 2022; Miller-Delaney et al., 2015; Miller-Delaney et al., 2012; Mohandas et al., 2019; Williams-Karnesky et al., 2013). Clinical support for the methylation hypothesis of epileptogenesis has been derived from the analysis of surgically resected tissue from patients with temporal lobe epilepsy and hippocampal sclerosis. Tissue from these patients, revealed progressive changes in DNA methylation that were tightly associated with the upregulation of genes supporting neuroinflammation (Martins-Ferreira et al., 2022).

Biochemically, all S-adenosylmethionine (SAM) dependent transmethylation reactions, including DNA methylation, lead to the production of adenosine through S-adenosylhomocysteine hydrolase (SAHH) and depend on efficient metabolic adenosine clearance through adenosine kinase (Boison et al., 2002; Williams-Karnesky et al., 2013). Thus, the genetic disruption or pharmacological inhibition of adenosine kinase leads to an increase in adenosine, which shifts the thermodynamic equilibrium of the SAHH reaction towards the formation of S-adenosylhomocysteine (SAH), which is a potent inhibitor of DNA methyltransferases. Conversely, pathological overexpression of adenosine kinase as part of the epileptogenic process drives increased DNA methylation and thereby promotes epileptogenesis (Williams-Karnesky et al., 2013). Strikingly, a selective splice variant of adenosine kinase, ADK-L, has been identified in the cell nucleus (Cui et al., 2009). ADK-L is needed for the cell cycle to permit the methylation of newly formed DNA during the S-phase of the cell cycle. Consequently, in the adult brain, ADK-L is only expressed in cells with proliferative capacity, such as neurons of the olfactory bulb and dentate gyrus, as well as in astrocytes (Gebril et al., 2021; Studer et al., 2006). Because maladaptive increases in adenosine kinase are part of the epileptogenic cascade (Gouder et al., 2004; Li et al., 2008b), increases in ADK-L drive maladaptive DNA methylation as a contributing factor for epilepsy development (Williams-Karnesky et al., 2013). Consequently, adenosine kinase inhibitors, which act on ADK-L have potent antiepileptogenic properties and are the subject of intense drug discovery efforts (Sandau et al., 2019; Toti et al., 2016).

2.10. Metabolic therapies for epilepsy and its prevention

As outlined above, astrocytes play a crucial role in maintaining the intricate balance of brain metabolism. Consequently, maladaptive changes in biochemical pathways and metabolism are intricately linked

to both ictogenesis and epileptogenesis. Therefore, astrocytes are logical targets for metabolic therapies. The most widely employed metabolic therapy is the ketogenic diet, which has been in clinical use for a century (Wheless, 2008). This diet is high in fats and low in carbohydrates and forces the brain to use fat-derived ketone bodies instead of glucose as the major energy source. Metabolic therapies have not only been shown to be highly effective in suppressing seizures in a wide spectrum of pharmacoresistant epilepsies (deCampo and Kossoff, 2019), but also to exert lasting disease modifying, and possibly antiepileptogenic properties in various clinical and experimental applications (Boison and Rho, 2020). The diet works through multiple mechanisms based on a combination of glucose restriction and elevation in ketone bodies (Simeone et al., 2018). Consequently, in rodent models the direct disruption of glycolysis with the glycolytic inhibitor 2-deoxy-D-glucose (2DG) has been explored as a therapeutic alternative to the stringent restrictions of the ketogenic diet (Ockuly et al., 2012). In those studies, 2DG provided both acute and chronic antiepileptic effects likely through a presynaptic mechanism (Pan et al., 2019). Likewise, in a traumatic brain injury model in mice, 2DG reduced the development of epileptiform activity in vitro and in vivo supporting the disease modifying potential of metabolic therapies (Koenig et al., 2019). These studies suggest that glucose restriction rather than ketone production is at the core of the therapeutic benefits of metabolic therapies. In line with this notion, medium-chain fatty acids, such as decanoic acid, can work through a ketone-independent mechanism and directly inhibit AMPA receptors and promote mitochondrial biogenesis (Augustin et al., 2018). Thus, medium-chain fatty acids can play a direct role in blocking seizure onset and raising seizure thresholds independent of ketones.

A further ketone-independent mechanism of metabolic therapies is an increase in adenosine in conjunction with downregulation of adenosine kinase. This is a likely explanation for the disease modifying properties of metabolic therapies (Lusardi et al., 2015; Masino et al., 2011). As outlined above, increased adenosine kinase expression drives the epileptogenic process through increased DNA methylation. Therefore, therapeutic adenosine augmentation is a rational approach for epilepsy prevention. We have shown that adenosine, delivered via cell-based brain implants, suppressed epileptogenesis in both kindling and post status epilepticus models of acquired epilepsy (Li et al., 2009; Li et al., 2008b; Li et al., 2007b). In line with those findings, the transient local delivery of adenosine to the hippocampus of rats through silk-

Table 1
Summary of mechanisms by which astrocytes are associated with epilepsy and their potential therapeutic implications.

Mechanism	Primary consequences	Potential therapeutic implications
Increased blood brain barrier permeability	• ↓blood-brain barrier integrity during seizures • ↓ AQP4 expression • ↓ clearance of glutamate and K^+ • ↓ penetrance of anticonvulsant drugs • ↑ astrogliosis • ↑ cell death • ↑ penetrance of iron and albumin from blood	• counteract increased blood - brain barrier permeability. • reduce multidrug transporter expression. • maintain AQP4 expression, particularly in perivascular end feet. • intervention in TGF-β or MAPK signaling pathways may be therapeutically beneficial.
Alteration in gap junction expression	• Variable ↓ or ↑ in gap junction expression • ↓ gap junction expression ⇒ ↓ K^+ buffering & ↓ adaptive volume regulation • ↑ gap junction expression ⇒ ↑ astrocytic Ca^{2+} signaling & ↑ metabolic support for local neurons.	• unclear whether benefits of increased gap junction expression in areas of K^+ buffering and volume regulation outweigh potential harm due to increased Ca^{2+} signaling and metabolic support for seizure activity. • more preclinical experimentation is needed to understand the conditions in which alterations in gap junction expression are beneficial.
Dysregulation of water homeostasis	• Variable ↓ or ↑ in net AQP4 expression • Consistent ↓ in AQP4 expression in perivascular end feet • ↓ blood-brain barrier function • ↓ volume regulation • ↓ K^+ buffering • ↓ reversal of cerebral edema following seizures	• facilitation of normal water homeostasis. • improvement in K^+ buffering. • prevention of the loss of localization of AQP4 channels in perivascular end feet may be therapeutically beneficial.
Impaired K^+ buffering	• ↓ K^+ buffering can result from: (1) ↓ gap junction expression (2) ↓ Kir4.1 channel expression (3) ↓ capacity for volume regulation (4) ↓ glutamate reuptake • ↓ K^+ buffering ⇒ ↑ vulnerability to seizures	Interventions which: • interfere with pathways causally upstream of pathological alterations in K^+ buffering (e.g., albumin extravasation, gap junction or aquaporin dysregulation, activation of the TGF-β or MAPK signaling pathways) or, • act directly to bolster K^+ buffering (e.g., enhancing the function or expression of the Kir4.1 channel or the Na^+/K^+-ATPase), may be therapeutic.
Dysregulation of glutamate and glutamine metabolism	• ↓ extracellular volume ⇒ ↑ extracellular glutamate concentrations • ↑ mGluR expression • ↑ release and ↓ reuptake of astrocytic glutamate associated with: (1) ↑ astrocytic Ca^{2+} signaling (2) ↑ vulnerability to seizures (3) ↑ excitotoxicity (4) ↓ glutamate reuptake	Interventions which: • increase astrocytic glutamate uptake, • decrease astrocytic glutamate release, • prevent overexpression of mGluR receptors during epileptogenesis, • increase the production of GABA from glutamate via glutamic acid decarboxylase, or • preserve the volume of the extracellular space, may be therapeutic.
Dysregulation of adenosine metabolism	• ↑ extracellular adenosine <u>*acutely*</u> during and after seizures • A_{2A} receptor activation ⇒ astrogliosis ⇒ ↑ adenosine kinase expression ⇒ ↑ metabolic adenosine clearance ⇒ <u>*chronic*</u> ↓ extracellular adenosine • ↑ vulnerability to seizures	Interventions which • reduce astrogliosis, • prevent astrogliosis and adenosine kinase overexpression, • decrease clearance of adenosine, such as adenosine kinase inhibitors, • increase release of adenosine, or • increase adenosine receptor expression, may be therapeutic.
Dysregulation of DNA methylation	• increased DNA methyltransferase activity • increased metabolic clearance of S-adenosylhomocysteine (SAH), which is a potent inhibitor of DNA methyltransferases. • upregulation of genes involved in neuroinflammation	• adenosine kinase inhibitors have potent antiepileptogenic properties though epigenetic mechanisms and are promising candidates for epilepsy prevention therapies.

based implants prevented epilepsy progression (Williams-Karnesky et al., 2013). Furthermore, a transient systemic dose of a small-molecule adenosine kinase inhibitor suppressed the epileptogenic process in mice after intrahippocampal kainic acid administration (Sandau et al., 2019). Importantly, a transient boost of adenosine during the latent period of epileptogenesis is sufficient to provide lasting suppression of epilepsy development and its progression (Sandau et al., 2019; Williams-Karnesky et al., 2013).

In conclusion, metabolic strategies that target astrocyte-dependent adenosine metabolism have the unique potential to disrupt the epileptogenic process through an epigenetic mechanism (Williams-Karnesky et al., 2013). Transient metabolic treatment approaches exert powerful disease modifying effects and may be able overcome challenges associated with conventional anti-seizure medications, such as pharmacoresistance, side effects, and a lack of lasting efficacy.

3. Conclusions and case studies in potential explanatory power

In the body of this review, we have outlined advances in our understanding of astrocyte function in epilepsy (Table 1). We will now turn our attention on how this knowledge can be usefully integrated. Detailing the explanatory power of what is known regarding astrocyte function in epilepsy in an exhaustive manner would necessitate a book-length format. Instead, here we will focus on one specific example: the role of astrocytes in the relationship between Alzheimer's disease, epilepsy, and the sleep-wake dysregulation observed in both conditions (Fig. 2). We hope that this example will be illustrative of the practical applicability of the information described in the body of this review. We will describe (1) the well characterized comorbid occurrence of epilepsy and Alzheimer's disease; (2) the similar sleep disturbances observed in persons with epilepsy and Alzheimer's disease; (3) the similar dysregulation in astrocytic and adenosinergic function seen in persons with epilepsy and Alzheimer's. We will then present the premise for the hypotheses that disrupted adenosine clearance due to astrogliosis contributes to (4) the etiology of sleep disruptions in Alzheimer's disease and epilepsy and (5) to the increased risk of seizures in patients with Alzheimer's disease.

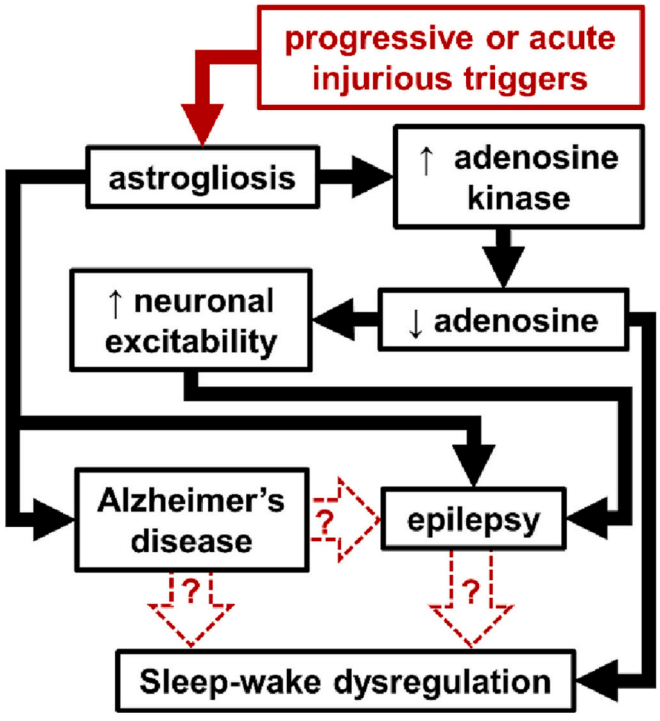


Fig. 2. The role of astrocytes in Alzheimer's disease, epilepsy, and associated sleep-wake dysregulation.
Schematic diagram of the hypothesized mechanism by which astrocytes contribute to epilepsy, Alzheimer's disease, and the associated disruption in sleep-wake regulation via disruption in adenosine signaling. Progressive or acute brain injury has been outlined with a red box to indicate that it is the causal point of origin. Red dotted arrows with question marks indicate known associations of unclear mechanistic cause.

3.1. Comorbid occurrence of epilepsy and Alzheimer's disease

Persons with Alzheimer's disease are at an increased risk of developing epilepsy (Pandis and Scarmeas, 2012) and may proceed to develop convulsive seizures (Vossel et al., 2016); however, prospective EEG studies in patients with Alzheimer's, but without an epilepsy diagnosis, have observed that approximately 40% of individuals with Alzheimer's have undiagnosed subclinical epileptiform activity (Vossel et al., 2016). These observations have led to the conjecture that epilepsy, particularly that characterized by nonconvulsive seizures, might be much more common in persons with Alzheimer's than had been previously understood and that the behavioral manifestations of Alzheimer's disease might be masking these seizures (Pandis and Scarmeas, 2012; Vossel et al., 2016).

3.2. Sleep disturbances in epilepsy and Alzheimer's disease patients

Self-reported sleep problems are common in patients with epilepsy, the most frequent complain being sleep maintenance insomnia characterized by frequent awakenings during the night (Hoeppner et al., 1984; Quigg et al., 2016). These subjective observations are corroborated by polysomnography data which indicates that epilepsy patients have an increase in wakefulness after sleep onset (Sudbrack-Oliveira et al., 2019). Despite these issues with maintaining nocturnal sleep, persons with epilepsy commonly experience excessive daytime sleepiness (Malow et al., 1997; Sudbrack-Oliveira et al., 2019).

Sleep problems are common in persons with Alzheimer's disease and are similar to those observed in individuals with epilepsy. Sleep maintenance insomnia, characterized by frequent awakenings during the night, is frequently associated with Alzheimer's disease (McCurry et al., 1999). These subjective observations are corroborated by polysomnography data which indicates that persons with Alzheimer's have an increase in wakefulness after sleep onset (Bonanni et al., 2005). Also like persons with epilepsy, individuals with Alzheimer's experience excessive daytime sleepiness (Bonanni et al., 2005).

3.3. Astrocytic and adenosinergic dysfunction in epilepsy and Alzheimer's

What insights can be gained into the etiology of sleep disturbances in epilepsy and Alzheimer's disease by looking through the star-shaped lens of augmented astrocytic function? Astrogliosis is a commonly observed feature of Alzheimer's disease and epilepsy in both human patients (Devinsky et al., 2013; Osborn et al., 2016) and animal models (Khurgel and Ivy, 1996; Olabarria et al., 2010). Astrocytes express adenosine kinase, the primary enzyme for adenosine metabolism, and are responsible for the regulation of extracellular adenosine concentrations (Etherington et al., 2009; Lloyd and Fredholm, 1995). The hypertrophy of astrocytes seen in astrogliosis causes overexpression of adenosine kinase and decreases extracellular adenosine concentrations (Aronica et al., 2013; Fedele et al., 2005). In Alzheimer's disease there are disparate focal alterations in astrocyte morphology and adenosine concentrations (Alonso-Andres et al., 2018; Verkhratsky et al., 2019). Adenosine levels are decreased in the frontal cortex, but increased in the temporal and parietal cortices in patients with Alzheimer's disease (Alonso-Andres et al., 2018; Rodriguez et al., 2009; Verkhratsky et al., 2019). On a much smaller scale, astrocytes in the vicinity of Aβ plaques undergo robust hypertrophy whereas astrocytes that are more distal to Aβ plaques become atrophied (Rodriguez et al., 2009; Verkhratsky et al.,

2019). It is conceivable that the balance between hypertrophied and atrophied astrocytes is responsible for the regional differences in adenosine concentrations seen in patients with Alzheimer's disease.

Hypothesis 1. *Disrupted adenosine clearance due to astrogliosis contributes to the etiology of sleep disruption in Alzheimer's disease and epilepsy.*

Adenosine signaling is essential to normal sleep-wake regulation and is widely considered to be a principal contributor to homeostatic sleep pressure in the two-process model of sleep-wake regulation (Borbely et al., 2016; Landolt, 2008). Extracellular adenosine levels increase with prolonged wakefulness (Porkka-Heiskanen et al., 1997) and increased extracellular adenosine promotes sleep (Portas et al., 1997). The sleep promoting effect of adenosine is most pronounced in the basal forebrain where it inhibits wake-promoting upwardly projecting cholinergic neurons (Arrigoni et al., 2006; Bjorness and Greene, 2009). Notably, the basal forebrain is just ventral to the frontal cortex, where adenosine levels are decreased in Alzheimer's patients (Alonso-Andres et al., 2018). Reduction in astrocytic, but not neuronal, adenosine kinase increases homeostatic sleep drive (Bjorness et al., 2016). Fittingly, transgenic upregulation of adenosine kinase, which increases the metabolic clearance of adenosine, causes a decrease in sleep (Palchykova et al., 2010). Considering the effect of adenosine on sleep and the largely astrocytic regulation of adenosine signaling it has been hypothesized that homeostatic sleep pressure should be conceptualized as an emergent property of a neuronal-glial circuit (Bjorness et al., 2016). We hypothesize that disrupted adenosine clearance due to astrogliosis contributes to the etiology of sleep disruptions in Alzheimer's disease and chronic epilepsy.

Hypothesis 2. *Disruption in adenosinergic signaling due to astrogliosis contribute to the increased risk of seizures in persons with Alzheimer's disease.*

The increased risk of epilepsy in patients with Alzheimer's disease may be the result of astroglial pathology (Boison, 2010). Adenosine is an endogenous anticonvulsant which is critical to the prevention and cessation of seizures (Beamer et al., 2021; Dragunow, 1991; During and Spencer, 1992). Adenosine kinase overexpression, without any other insult or injury, causes adenosine deficits and spontaneous seizures (Li et al., 2008b). We hypothesize that disruptions in adenosinergic signaling due to astrogliosis contribute to the increased risk of seizures in patients with Alzheimer's disease. Furthermore, we posit that the focal disparities in adenosine kinase expression between hypertrophied astrocytes that are close to amyloid plaques and atrophied astrocytes that are further away cause variations in adenosine concentrations which prevent epileptiform activity from generalizing and becoming convulsive.

In conclusion, astrocytes play an indispensable role in regulating neuronal activity in health and disease. As our understanding of astrocytic function improves, the potential implications of astrocyte dysfunction become clearer. In this review we summarize the derangement of astrocyte function observed in epilepsy and present astrocyte focused hypotheses on the interplay between epilepsy, Alzheimer's disease, and sleep-wake dysregulation. We proffer these hypotheses as examples of useful insights based on our rapidly advancing understanding of astrocytes in epilepsy.

Declaration of Competing Interest

DB is co-founder and CDO of PrevEp Inc.

Data availability

No data was used for the research described in the article.

Acknowledgements

This work was supported by funding from the National Institutes of Health (NIH) through grants NS103740, NS065957, NS127846, to DB and grant NS117792 to BSP; from the US Department of the Army through contract W81XWH2210638 to DB; a Catalyst award from CURE Epilepsy to DB; and the EpiPurines grant 101032321 from the European Commission to MA. We would also like to apologize to those authors whose relevant work was not cited here for space reasons.

References

Abbott, N.J., 2013. Blood-brain barrier structure and function and the challenges for CNS drug delivery. J. Inherit. Metab. Dis. 36, 437–449.
Abbott, N.J., et al., 2006. Astrocyte-endothelial interactions at the blood-brain barrier. Nat. Rev. Neurosci. 7, 41–53.
Akinfiresoye, L.R., et al., 2023. Targeted inhibition of upregulated sodium-calcium exchanger in rat inferior colliculus suppresses alcohol withdrawal seizures. Mol. Neurobiol. 60, 292–302.
Alonso-Andres, P., et al., 2018. Purine-related metabolites and their converting enzymes are altered in frontal, parietal and temporal cortex at early stages of Alzheimer's disease pathology. Brain Pathol. 28, 933–946.
Alvestad, S., et al., 2013. Mislocalization of AQP4 precedes chronic seizures in the kainate model of temporal lobe epilepsy. Epilepsy Res. 105, 30–41.
Amédée, T., et al., 1998. Potassium homeostasis and glial energy metabolism. Glia. 21, 46–55.
Amiry-Moghaddam, M., et al., 2003. Delayed K+ clearance associated with aquaporin-4 mislocalization: phenotypic defects in brains of alpha-syntrophin-null mice. Proc. Natl. Acad. Sci. U. S. A. 100, 13615–13620.
Andrew, R.D., 1991. Seizure and acute osmotic change: clinical and neurophysiological aspects. J. Neurol. Sci. 101, 7–18.
Andrew, R.D., et al., 1989. Seizure susceptibility and the osmotic state. Brain Res. 498, 175–180.
Anlauf, E., Derouiche, A., 2013. Glutamine synthetase as an astrocytic marker: its cell type and vesicle localization. Front. Endocrinol. (Lausanne). 4, 144.
Araque, A., et al., 1999. Tripartite synapses: glia, the unacknowledged partner. Trends Neurosci. 22, 208–215.
Aronica, E., et al., 2000. Upregulation of metabotropic glutamate receptor subtype mGluR3 and mGluR5 in reactive astrocytes in a rat model of mesial temporal lobe epilepsy. Eur. J. Neurosci. 12, 2333–2344.
Aronica, E., et al., 2001. Expression of connexin 43 and connexin 32 gap-junction proteins in epilepsy-associated brain tumors and in the perilesional epileptic cortex. Acta Neuropathol. 101, 449–459.
Aronica, E., et al., 2012a. Astrocyte immune responses and epilepsy. Glia. 60, 1258–1268.
Aronica, E., et al., 2012b. Cerebral expression of drug transporters in epilepsy. Adv. Drug Deliv. Rev. 64, 919–929.
Aronica, E., et al., 2013. Glial adenosine kinase - a neuropathological marker of the epileptic brain. Neurochem. Int. 63, 688–695.
Arrigoni, E., et al., 2006. Adenosine inhibits basal forebrain cholinergic and noncholinergic neurons in vitro. Neuroscience. 140, 403–413.
Augustin, K., et al., 2018. Mechanisms of action for the medium-chain triglyceride ketogenic diet in neurological and metabolic disorders. Lancet Neurol. 17, 84–93.
Bankstahl, M., et al., 2018. Blood-brain barrier leakage during early Epileptogenesis is associated with rapid Remodeling of the neurovascular unit. eNeuro. 5.
von Bartheld, C.S., et al., 2016. The search for true numbers of neurons and glial cells in the human brain: a review of 150 years of cell counting. J. Comp. Neurol. 524, 3865–3895.
Bauer, K.F., Leonhardt, H., 1956. A contribution to the pathological physiology of the blood-brain-barrier; megaphen stabilises the blood-brain-barrier. J. Comp. Neurol. 106, 363–370.
Bay, V., Butt, A.M., 2012. Relationship between glial potassium regulation and axon excitability: a role for glial Kir4.1 channels. Glia. 60, 651–660.
Bayraktar, O.A., et al., 2014. Astrocyte development and heterogeneity. Cold Spring Harb. Perspect. Biol. 7, a020362.
Beamer, E., et al., 2021. ATP and adenosine-two players in the control of seizures and epilepsy development. Prog. Neurobiol. 204, 102105.
Ben Haim, L., Rowitch, D.H., 2017. Functional diversity of astrocytes in neural circuit regulation. Nat. Rev. Neurosci. 18, 31–41.
Benson, M.J., et al., 2015. A novel anticonvulsant mechanism via inhibition of complement receptor C5ar1 in murine epilepsy models. Neurobiol. Dis. 76, 87–97.
Bezzi, P., et al., 2004. Astrocytes contain a vesicular compartment that is competent for regulated exocytosis of glutamate. Nat. Neurosci. 7, 613–620.
Binder, D.K., Steinhauser, C., 2006. Functional changes in astroglial cells in epilepsy. Glia. 54, 358–368.
Binder, D.K., Steinhauser, C., 2021. Astrocytes and epilepsy. Neurochem. Res. 46, 2687–2695.
Binder, D.K., et al., 2004. Increased seizure threshold in mice lacking aquaporin-4 water channels. Neuroreport. 15, 259–262.
Binder, D.K., et al., 2006. Increased seizure duration and slowed potassium kinetics in mice lacking aquaporin-4 water channels. Glia. 53, 631–636.

Bjorness, T.E., Greene, R.W., 2009. Adenosine and sleep. Curr. Neuropharmacol. 7, 238–245.
Bjorness, T.E., et al., 2016. An adenosine-mediated glial-neuronal circuit for homeostatic sleep. J. Neurosci. 36, 3709–3721.
Bloch, O., et al., 2005. Aquaporin-4 gene deletion in mice increases focal edema associated with staphylococcal brain abscess. J. Neurochem. 95, 254–262.
Blumcke, I., et al., 2000. Temporal lobe epilepsy associated up-regulation of metabotropic glutamate receptors: correlated changes in mGluR1 mRNA and protein expression in experimental animals and human patients. J. Neuropathol. Exp. Neurol. 59, 1–10.
Bohmbach, K., et al., 2018. The structural and functional evidence for vesicular release from astrocytes in situ. Brain Res. Bull. 136, 65–75.
Boison, D., 2010. Adenosine dysfunction and adenosine kinase in epileptogenesis. Open Neurosci. J. 4, 93–101.
Boison, D., 2011. Methylxanthines, seizures and excitotoxicity. Handb. Exp. Pharmacol. 200, 251–266.
Boison, D., 2012. Adenosine augmentation therapy for epilepsy. In: Noebels, J.L., et al. (Eds.), Jasper's Basic Mechanisms of the Epilepsies. Oxford University Press, Oxford, pp. 1150–1160.
Boison, D., Rho, J.M., 2020. Epigenetics and epilepsy prevention: the therapeutic potential of adenosine and metabolic therapies. Neuropharmacology. 167, 107741.
Boison, D., Steinhauser, C., 2018. Epilepsy and astrocyte energy metabolism. Glia. 66, 1235–1243.
Boison, D., Yegutkin, G.G., 2019. Adenosine metabolism: emerging concepts for cancer therapy. Cancer Cell 36, 582–596.
Boison, D., et al., 2002. Neonatal hepatic steatosis by disruption of the adenosine kinase gene. Proc. Natl. Acad. Sci. U. S. A. 99, 6985–6990.
Bonanni, E., et al., 2005. Daytime sleepiness in mild and moderate Alzheimer's disease and its relationship with cognitive impairment. J. Sleep Res. 14, 311–317.
Borbely, A.A., et al., 2016. The two-process model of sleep regulation: a reappraisal. J. Sleep Res. 25, 131–143.
Bordey, A., Sontheimer, H., 1998. Properties of human glial cells associated with epileptic seizure foci. Epilepsy Res. 32, 286–303.
Braganza, O., et al., 2012. Albumin is taken up by hippocampal NG2 cells and astrocytes and decreases gap junction coupling. Epilepsia. 53, 1898–1906.
Brambilla, R., et al., 2003. Blockade of A2A adenosine receptors prevents basic fibroblast growth factor-induced reactive astrogliosis in rat striatal primary astrocytes. Glia. 43, 190–194.
Brill, J., et al., 2004. *Entla:* a novel epileptic and ataxic *Cacna2d2* mutant of the mouse. J. Biol. Chem. 279, 7322–7330.
Bushong, E.A., et al., 2002. Protoplasmic astrocytes in CA1 stratum radiatum occupy separate anatomical domains. J. Neurosci. 22, 183–192.
Cacheaux, L.P., et al., 2009. Transcriptome profiling reveals TGF-beta signaling involvement in epileptogenesis. J. Neurosci. 29, 8927–8935.
Cameron, R., et al., 1994. Neurons and astroglia express distinct subsets of Na,K-ATPase alpha and beta subunits. Brain Res. Mol. Brain Res. 21, 333–343.
Chai, H., et al., 2017. Neural circuit-specialized astrocytes: transcriptomic, proteomic, morphological, and functional evidence. Neuron. 95 (531–549), e9.
Chebabo, S.R., Hester, M.A., Aitken, P.G., Somjen, G.G., 1995. Hypotonic exposure enhances synaptic transmission and triggers spreading depression in rathippocampal tissue slices. Brain Res 695 (2), 203–216.
Clasadonte, J., et al., 2013. Astrocyte control of synaptic NMDA receptors contributes to the progressive development of temporal lobe epilepsy. Proc. Natl. Acad. Sci. U. S. A. 110, 17540–17545.
Colbourn, R., et al., 2021. Rapid volume pulsation of the extracellular space coincides with epileptiform activity in mice and depends on the NBCe1 transporter. J. Physiol. 599, 3195–3220.
Collignon, F., et al., 2006. Altered expression of connexin subtypes in mesial temporal lobe epilepsy in humans. J. Neurosurg. 105, 77–87.
Condorelli, D.F., et al., 2002. Connexin-30 mRNA is up-regulated in astrocytes and expressed in apoptotic neuronal cells of rat brain following kainate-induced seizures. Mol. Cell. Neurosci. 21, 94–113.
Cui, X.A., et al., 2009. Subcellular localization of adenosine kinase in mammalian cells: the long isoform of AdK is localized in the nucleus. Biochem. Biophys. Res. Commun. 388, 46–50.
D'Ambrosio, R., et al., 1999. Impaired K(+) homeostasis and altered electrophysiological properties of post-traumatic hippocampal glia. J. Neurosci. 19, 8152–8162.
Daneman, R., Prat, A., 2015. The blood-brain barrier. Cold Spring Harb. Perspect. Biol. 7, a020412.
David, Y., et al., 2009. Astrocytic dysfunction in epileptogenesis: consequence of altered potassium and glutamate homeostasis? J. Neurosci. 29, 10588–10599.
deCampo, D.M., Kossoff, E.H., 2019. Ketogenic dietary therapies for epilepsy and beyond. Curr. Opin. Clin. Nutr. Metab. Care. 22, 264–268.
Deitmer, J.W., et al., 2019. Energy dynamics in the brain: contributions of astrocytes to metabolism and pH homeostasis. Front. Neurosci. 13, 1301.
Devinsky, O., et al., 2013. Glia and epilepsy: excitability and inflammation. Trends Neurosci. 36, 174–184.
Diaz Verdugo, C., et al., 2019. Glia-neuron interactions underlie state transitions to generalized seizures. Nat. Commun. 10, 3830.
Ding, S., et al., 2007. Enhanced astrocytic Ca2+ signals contribute to neuronal excitotoxicity after status epilepticus. J. Neurosci. 27, 10674–10684.
Djukic, B., et al., 2007. Conditional knock-out of Kir4.1 leads to glial membrane depolarization, inhibition of potassium and glutamate uptake, and enhanced short-term synaptic potentiation. J. Neurosci. 27, 11354–11365.
Dossi, E., et al., 2018. Pannexin-1 channels contribute to seizure generation in human epileptic brain tissue and in a mouse model of epilepsy. Sci. Transl. Med. 10.
Doupnik, C.A., et al., 1995. The inward rectifier potassium channel family. Curr. Opin. Neurobiol. 5, 268–277.
Dragunow, M., 1991. Adenosine and seizure termination. Ann. Neurol. 29, 575.
During, M.J., Spencer, D.D., 1992. Adenosine: a potential mediator of seizure arrest and postictal refractoriness. Ann. Neurol. 32, 618–624.
During, M.J., Spencer, D.D., 1993. Extracellular hippocampal glutamate and spontaneous seizure in the conscious human brain. Lancet. 341, 1607–1610.
Eid, T., et al., 2005. Loss of perivascular aquaporin 4 may underlie deficient water and K + homeostasis in the human epileptogenic hippocampus. Proc. Natl. Acad. Sci. U. S. A. 102, 1193–1198.
Elisevich, K., et al., 1997a. Connexin 43 mRNA expression in two experimental models of epilepsy. Mol. Chem. Neuropathol. 32, 75–88.
Elisevich, K., et al., 1997b. Hippocampal connexin 43 expression in human complex partial seizure disorder. Exp. Neurol. 145, 154–164.
Engel, T., et al., 2016. ATPergic signalling during seizures and epilepsy. Neuropharmacology. 104, 140–153.
Etherington, L.A., et al., 2009. Astrocytic adenosine kinase regulates basal synaptic adenosine levels and seizure activity but not activity-dependent adenosine release in the hippocampus. Neuropharmacology. 56, 429–437.
Eyo, U.B., et al., 2017. Microglia-neuron communication in epilepsy. Glia. 65, 5–18.
Fedele, D.E., et al., 2004. Engineering embryonic stem cell derived glia for adenosine delivery. Neurosci. Lett. 370, 160–165.
Fedele, D.E., et al., 2005. Astrogliosis in epilepsy leads to overexpression of adenosine kinase resulting in seizure aggravation. Brain. 128, 2383–2395.
Fellin, T., et al., 2004. Neuronal synchrony mediated by astrocytic glutamate through activation of extrasynaptic NMDA receptors. Neuron. 43, 729–743.
Fields, R.D., 2011. Nonsynaptic and nonvesicular ATP release from neurons and relevance to neuron-glia signaling. Semin. Cell Dev. Biol. 22, 214–219.
Fischer, G., Kettenmann, H., 1985. Cultured astrocytes form a syncytium after maturation. Exp. Cell Res. 159, 273–279.
Fonseca, C.G., et al., 2002. Upregulation in astrocytic connexin 43 gap junction levels may exacerbate generalized seizures in mesial temporal lobe epilepsy. Brain Res. 929, 105–116.
Friedman, A., et al., 2009. Blood-brain barrier breakdown-inducing astrocytic transformation: novel targets for the prevention of epilepsy. Epilepsy Res. 85, 142–149.
Fujii, Y., et al., 2017. Astrocyte calcium waves propagate proximally by gap junction and distally by extracellular diffusion of ATP released from volume-regulated anion channels. Sci. Rep. 7, 13115.
Gabriel, S., et al., 1998. Effects of barium on stimulus induced changes in extracellular potassium concentration in area CA1 of hippocampal slices from normal and pilocarpine-treated epileptic rats. Neurosci. Lett. 242, 9–12.
Garrido-Sanabria, E.R., et al., 2008. Impaired expression and function of group II metabotropic glutamate receptors in pilocarpine-treated chronically epileptic rats. Brain Res. 1240, 165–176.
Gebril, H., et al., 2021. Developmental role of adenosine kinase in the cerebellum. eNeuro. 8.
Giaume, C., et al., 1997. Metabolic trafficking through astrocytic gap junctions. Glia. 21, 114–123.
Gomez-Gonzalo, M., et al., 2010. An excitatory loop with astrocytes contributes to drive neurons to seizure threshold. PLoS Biol. 8, e1000352.
Gorter, J.A., et al., 2005. Increased expression of ferritin, an iron-storage protein, in specific regions of the parahippocampal cortex of epileptic rats. Epilepsia. 46, 1371–1379.
Gouder, N., et al., 2004. Overexpression of adenosine kinase in epileptic hippocampus contributes to epileptogenesis. J. Neurosci. 24, 692–701.
Gruber, V.E., et al., 2022. Increased expression of complement components in tuberous sclerosis complex and focal cortical dysplasia type 2B brain lesions. Epilepsia. 63, 364–374.
Habas, A., et al., 2013. Neuronal activity regulates astrocytic Nrf2 signaling. Proc. Natl. Acad. Sci. U. S. A. 110, 18291–18296.
Haj-Yasein, N.N., et al., 2012. Aquaporin-4 regulates extracellular space volume dynamics during high-frequency synaptic stimulation: a gene deletion study in mouse hippocampus. Glia. 60, 867–874.
Halassa, M.M., et al., 2007. The tripartite synapse: roles for gliotransmission in health and disease. Trends Mol. Med. 13, 54–63.
Heidinger, V., et al., 2002. Metabotropic glutamate receptor 1-induced upregulation of NMDA receptor current: mediation through the Pyk2/Src-family kinase pathway in cortical neurons. J. Neurosci. 22, 5452–5461.
Heinemann, U., et al., 1977. Extracellular free calcium and potassium during paroxsmal activity in the cerebral cortex of the cat. Exp. Brain Res. 27, 237–243.
Henn, F.A., et al., 1972. Glial cell function: active control of extracellular K + concentration. Brain Res. 43, 437–443.
Henneberger, C., et al., 2010. Long-term potentiation depends on release of D-serine from astrocytes. Nature. 463, 232–236.
Hertz, L., Chen, Y., 2016. Importance of astrocytes for potassium ion (K(+)) homeostasis in brain and glial effects of K(+) and its transporters on learning. Neurosci. Biobehav. Rev. 71, 484–505.
Heuser, K., et al., 2018. Ca2+ signals in astrocytes facilitate spread of Epileptiform activity. Cereb. Cortex 28, 4036–4048.
Hinterkeuser, S., et al., 2000. Astrocytes in the hippocampus of patients with temporal lobe epilepsy display changes in potassium conductances. Eur. J. Neurosci. 12, 2087–2096.
Hodgkin, A.L., Huxley, A.F., 1952. Currents carried by sodium and potassium ions through the membrane of the giant axon of Loligo. J. Physiol. 116, 449–472.

Hoeppner, J.B., et al., 1984. Self-reported sleep disorder symptoms in epilepsy. Epilepsia. 25, 434–437.
Hoffmann, K., et al., 2006. Expression of the multidrug transporter MRP2 in the blood-brain barrier after pilocarpine-induced seizures in rats. Epilepsy Res. 69, 1–14.
Hofmann, F., et al., 1999. Voltage-dependent calcium channels: from structure to function. Rev. Physiol. Biochem. Pharmacol. 139, 33–87.
Holthoff, K., Witte, O.W., 1996. Intrinsic optical signals in rat neocortical slices measured with near-infrared dark-field microscopy reveal changes in extracellular space. J. Neurosci. 16, 2740–2749.
Holthoff, K., Witte, O.W., 2000. Directed spatial potassium redistribution in rat neocortex. Glia. 29, 288–292.
Hubbard, J.A., et al., 2015. Expression of the astrocyte water channel Aquaporin-4 in the mouse brain. ASN Neuro. 7.
Hubbard, J.A., et al., 2016. Regulation of astrocyte glutamate transporter-1 (GLT1) and aquaporin-4 (AQP4) expression in a model of epilepsy. Exp. Neurol. 283, 85–96.
Ivens, S., et al., 2007. TGF-beta receptor-mediated albumin uptake into astrocytes is involved in neocortical epileptogenesis. Brain. 130, 535–547.
Jimenez-Pacheco, A., et al., 2013. Increased neocortical expression of the P2X7 receptor after status epilepticus and anticonvulsant effect of P2X7 receptor antagonist A-438079. Epilepsia. 54, 1551–1561.
Kandratavicius, L., et al., 2015. Mesial temporal lobe epilepsy with psychiatric comorbidities: a place for differential neuroinflammatory interplay. J. Neuroinflammation 12, 38.
Kang, E.J., et al., 2013. Blood-brain barrier opening to large molecules does not imply blood-brain barrier opening to small ions. Neurobiol. Dis. 52, 204–218.
Kelly, E., et al., 2018. mGluR5 modulation of behavioral and epileptic phenotypes in a mouse model of tuberous sclerosis complex. Neuropsychopharmacology. 43, 1457–1465.
Khakh, B.S., Sofroniew, M.V., 2015. Diversity of astrocyte functions and phenotypes in neural circuits. Nat. Neurosci. 18, 942–952.
Khananshvili, D., 2014. Sodium-calcium exchangers (NCX): molecular hallmarks underlying the tissue-specific and systemic functions. Pflugers Arch. 466, 43–60.
Khurgel, M., Ivy, G.O., 1996. Astrocytes in kindling: relevance to epileptogenesis. Epilepsy Res. 26, 163–175.
Kilb, W., Dierkes, P.W., Sykova, E., Vargova, L., Luhmann, H.J., 2006. Hypoosmolar conditions reduce extracellular volume fraction and enhance epileptiform activity in the CA3 region of the immature rat hippocampus. J. Neurosci. Res. 84 (1), 119–129.
Kim, G.W., et al., 2009. The role of MMP-9 in integrin-mediated hippocampal cell death after pilocarpine-induced status epilepticus. Neurobiol. Dis. 36, 169–180.
Kim, J.E., et al., 2010. Astroglial loss and edema formation in the rat piriform cortex and hippocampus following pilocarpine-induced status epilepticus. J. Comp. Neurol. 518, 4612–4628.
Kimelberg, H.K., Nedergaard, M., 2010. Functions of astrocytes and their potential as therapeutic targets. Neurotherapeutics. 7, 338–353.
Kivi, A., et al., 2000. Effects of barium on stimulus-induced rises of [K+]o in human epileptic non-sclerotic and sclerotic hippocampal area CA1. Eur. J. Neurosci. 12, 2039–2048.
Klein, P., et al., 2018. Commonalities in epileptogenic processes from different acute brain insults: do they translate? Epilepsia. 59, 37–66.
Kobow, K., Blumcke, I., 2011. The methylation hypothesis: do epigenetic chromatin modifications play a role in epileptogenesis? Epilepsia. 52 (Suppl. 4), 15–19.
Kobow, K., Blumcke, I., 2018. Epigenetics in epilepsy. Neurosci. Lett. 667, 40–46.
Kobow, K., et al., 2009. Increased reelin promoter methylation is associated with granule cell dispersion in human temporal lobe epilepsy. J. Neuropathol. Exp. Neurol. 68, 356–364.
Kobow, K., et al., 2013a. The methylation hypothesis of pharmacoresistance in epilepsy. Epilepsia. 54 (Suppl. 2), 41–47.
Kobow, K., et al., 2013b. Deep sequencing reveals increased DNA methylation in chronic rat epilepsy. Acta Neuropathol. 126, 741–756.
Koehler, R.C., et al., 2009. Astrocytes and the regulation of cerebral blood flow. Trends Neurosci. 32, 160–169.
Koenig, J.B., et al., 2019. Glycolytic inhibitor 2-deoxyglucose prevents cortical hyperexcitability after traumatic brain injury. JCI. Insight. 5.
Kofuji, P., Newman, E.A., 2004. Potassium buffering in the central nervous system. Neuroscience. 129, 1045–1056.
Kucheryavykh, Y.V., et al., 2007. Downregulation of Kir4.1 inward rectifying potassium channel subunits by RNAi impairs potassium transfer and glutamate uptake by cultured cortical astrocytes. Glia. 55, 274–281.
Kwan, P., Brodie, M.J., 2006. Refractory epilepsy: mechanisms and solutions. Expert. Rev. Neurother. 6, 397–406.
Lado, F.A., Moshe, S.L., 2008. How do seizures stop? Epilepsia. 49, 1651–1664.
Landolt, H.P., 2008. Sleep homeostasis: a role for adenosine in humans? Biochem. Pharmacol. 75, 2070–2079.
Lapato, A.S., et al., 2020. Astrocyte glutamate uptake and water homeostasis are dysregulated in the Hippocampus of multiple sclerosis patients with seizures. ASN Neuro. 12, 1759091420979604.
Lau, A., Tymianski, M., 2010. Glutamate receptors, neurotoxicity and neurodegeneration. Pflugers Arch. 460, 525–542.
Lazarowski, A., et al., 2007. ABC transporters during epilepsy and mechanisms underlying multidrug resistance in refractory epilepsy. Epilepsia. 48 (Suppl. 5), 140–149.
Lee, D.J., et al., 2012. Decreased expression of the glial water channel aquaporin-4 in the intrahippocampal kainic acid model of epileptogenesis. Exp. Neurol. 235, 246–255.
Lee, S.H., et al., 1994. Astrocytes exhibit regional specificity in gap-junction coupling. Glia. 11, 315–325.
Lee, T.S., et al., 2004. Aquaporin-4 is increased in the sclerotic hippocampus in human temporal lobe epilepsy. Acta Neuropathol. 108, 493–502.
Li, H., et al., 2021. Bidirectional regulation of glial potassium buffering - glioprotection versus neuroprotection. Elife. 10.
Li, J., et al., 2020. Ferroptosis: past, present and future. Cell Death Dis. 11, 88.
Li, T., et al., 2007a. Adenosine dysfunction in astrogliosis: cause for seizure generation? Neuron Glia Biol. 3, 353–366.
Li, T., et al., 2007b. Suppression of kindling epileptogenesis by adenosine releasing stem cell-derived brain implants. Brain. 130, 1276–1288.
Li, T., et al., 2008a. Uncoupling of astrogliosis from epileptogenesis in adenosine kinase (ADK) transgenic mice. Neuron Glia Biol. 4, 91–99.
Li, T., et al., 2008b. Adenosine kinase is a target for the prediction and prevention of epileptogenesis in mice. J. Clin. Invest. 118, 571–582.
Li, T., et al., 2009. Human mesenchymal stem cell grafts engineered to release adenosine reduce chronic seizures in a mouse model of CA3-selective epileptogenesis. Epilepsy Res. 84, 238–241.
Liddelow, S.A., et al., 2017. Neurotoxic reactive astrocytes are induced by activated microglia. Nature. 541, 481–487.
Liu, K., et al., 2021. Attenuation of cerebral edema facilitates recovery of glymphatic system function after status epilepticus. JCI. Insight. 6.
Lloyd, H.G., Fredholm, B.B., 1995. Involvement of adenosine deaminase and adenosine kinase in regulating extracellular adenosine concentration in rat hippocampal slices. Neurochem. Int. 26, 387–395.
Lones, L., DiAntonio, A., 2023. SIK3 and Wnk converge on fray to regulate glial K+ buffering and seizure susceptibility. PLoS Genet. 19, e1010581.
Lorenzo, A.V., et al., 1975. Increased penetration of horseradish peroxidase across the blood-brain barrier induced by Metrazol seizures. Brain Res. 88, 136–140.
Loscher, W., Friedman, A., 2020. Structural, molecular, and functional alterations of the blood-brain barrier during Epileptogenesis and epilepsy: a cause, consequence, or both? Int. J. Mol. Sci. 21.
Lu, D.C., et al., 2021. Aquaporin-4 reduces post-traumatic seizure susceptibility by promoting astrocytic glial scar formation in mice. J. Neurotrauma 38, 1193–1201.
Lusardi, T.A., et al., 2015. Ketogenic diet prevents epileptogenesis and disease progression in adult mice and rats. Neuropharmacology. 99, 500–509.
Lux, H.D., et al., 1986. Ionic changes and alterations in the size of the extracellular space during epileptic activity. Adv. Neurol. 44, 619–639.
MacVicar, B.A., et al., 2002. Intrinsic optical signals in the rat optic nerve: role for K(+) uptake via NKCC1 and swelling of astrocytes. Glia. 37, 114–123.
Madeira, M.H., et al., 2018. Blockade of microglial adenosine A2A receptor impacts inflammatory mechanisms, reduces ARPE-19 cell dysfunction and prevents photoreceptor loss in vitro. Sci. Rep. 8, 2272.
Mader, S., Brimberg, L., 2019. Aquaporin-4 Water Channel in the brain and its implication for health and disease. Cells. 8.
Mahmoud, S., et al., 2019. Astrocytes maintain glutamate homeostasis in the CNS by controlling the balance between glutamate uptake and release. Cells. 8.
Malow, B.A., et al., 1997. Predictors of sleepiness in epilepsy patients. Sleep. 20, 1105–1110.
Marchi, N., et al., 2006. Determinants of drug brain uptake in a rat model of seizure-associated malformations of cortical development. Neurobiol. Dis. 24, 429–442.
Maroso, M., et al., 2010. Toll-like receptor 4 and high-mobility group box-1 are involved in ictogenesis and can be targeted to reduce seizures. Nat. Med. 16, 413–419.
Martinez, Y., N'Gouemo, P., 2010. Blockade of the sodium calcium exchanger exhibits anticonvulsant activity in a pilocarpine model of acute seizures in rats. Brain Res. 1366, 211–216.
Martins-Ferreira, R., et al., 2022. Epilepsy progression is associated with cumulative DNA methylation changes in inflammatory genes. Prog. Neurobiol. 209, 102207.
Masino, S.A., et al., 2011. A ketogenic diet suppresses seizures in mice through adenosine A1 receptors. J. Clin. Invest. 121, 2679–2683.
McCormick, D.A., Contreras, D., 2001. On the cellular and network bases of epileptic seizures. Annu. Rev. Physiol. 63, 815–846.
McCurry, S.M., et al., 1999. Characteristics of sleep disturbance in community-dwelling Alzheimer's disease patients. J. Geriatr. Psychiatry Neurol. 12, 53–59.
McGrail, K.M., et al., 1991. Immunofluorescent localization of three Na,K-ATPase isozymes in the rat central nervous system: both neurons and glia can express more than one Na, K-ATPase. J. Neurosci. 11, 381–391.
McKenna, M.C., 2013. Glutamate pays its own way in astrocytes. Front. Endocrinol. (Lausanne). 4, 191.
McNamara, J.O., 1994. Cellular and molecular basis of epilepsy. J. Neurosci. 14, 3413–3425.
Meldrum, B.S., 2000. Glutamate as a neurotransmitter in the brain: review of physiology and pathology. J. Nutr. 130, 1007S–15S.
Michaluk, P., et al., 2011. Influence of matrix metalloproteinase MMP-9 on dendritic spine morphology. J. Cell Sci. 124, 3369–3380.
Miller-Delaney, S.F., et al., 2012. Differential DNA methylation patterns define status epilepticus and epileptic tolerance. J. Neurosci. 32, 1577–1588.
Miller-Delaney, S.F., et al., 2015. Differential DNA methylation profiles of coding and non-coding genes define hippocampal sclerosis in human temporal lobe epilepsy. Brain. 138, 616–631.
Mohandas, N., et al., 2019. Evidence for type-specific DNA methylation patterns in epilepsy: a discordant monozygotic twin approach. Epigenomics. 11, 951–968.
Muller, J., et al., 2020. Astrocytic GABA accumulation in experimental temporal lobe epilepsy. Front. Neurol. 11, 614923.
Muraleedharan, R., et al., 2020. AMPK-regulated astrocytic lactate shuttle plays a non-cell-autonomous role in neuronal survival. Cell Rep. 32, 108092.
Murugan, M., et al., 2021. Adenosine kinase: an epigenetic modulator in development and disease. Neurochem. Int. 147, 105054.

Nagao, Y., et al., 2013. Expressional analysis of the astrocytic Kir4.1 channel in a pilocarpine-induced temporal lobe epilepsy model. Front. Cell. Neurosci. 7, 104.
Naus, C.C., et al., 1991. Gap junction gene expression in human seizure disorder. Exp. Neurol. 111, 198–203.
Nedergaard, M., 1994. Direct signaling from astrocytes to neurons in cultures of mammalian brain cells. Science. 263, 1768–1771.
Nelson, S.R., Olson, J.P., 1987. Role of early edema in the development of regional seizure-related brain damage. Neurochem. Res. 12, 561–564.
Newton, J., et al., 2021. Inhibition of the sodium calcium exchanger suppresses alcohol withdrawal-induced seizure susceptibility. Brain Sci. 11.
Oberheim, N.A., et al., 2009. Uniquely hominid features of adult human astrocytes. J. Neurosci. 29, 3276–3287.
Oberheim, N.A., et al., 2012. Heterogeneity of astrocytic form and function. Methods Mol. Biol. 814, 23–45.
Ockuly, J.C., et al., 2012. Behavioral, cognitive, and safety profile of 2-deoxy-2-glucose (2DG) in adult rats. Epilepsy Res. 101, 246–252.
Olabarria, M., et al., 2010. Concomitant astroglial atrophy and astrogliosis in a triple transgenic animal model of Alzheimer's disease. Glia. 58, 831–838.
Olney, J.W., et al., 1972. Glutamate-induced brain damage in infant primates. J. Neuropathol. Exp. Neurol. 31, 464–488.
Osborn, L.M., et al., 2016. Astrogliosis: an integral player in the pathogenesis of Alzheimer's disease. Prog. Neurobiol. 144, 121–141.
Pacheco Otalora, L.F., et al., 2006. Abnormal mGluR2/3 expression in the perforant path termination zones and mossy fibers of chronically epileptic rats. Brain Res. 1098, 170–185.
Palchykova, S., et al., 2010. Manipulation of adenosine kinase affects sleep regulation in mice. J. Neurosci. 30, 13157–13165.
Pan, Y.Z., et al., 2019. 2-Deoxy-d-glucose reduces epileptiform activity by presynaptic mechanisms. J. Neurophysiol. 121, 1092–1101.
Pandis, D., Scarmeas, N., 2012. Seizures in Alzheimer disease: clinical and epidemiological data. Epilepsy Curr. 12, 184–187.
Pandit, S., et al., 2020. Bestrophin1-mediated tonic GABA release from reactive astrocytes prevents the development of seizure-prone network in kainate-injected hippocampi. Glia. 68, 1065–1080.
Pannasch, U., et al., 2011. Astroglial networks scale synaptic activity and plasticity. Proc. Natl. Acad. Sci. U. S. A. 108, 8467–8472.
Papadopoulos, M.C., Verkman, A.S., 2013. Aquaporin water channels in the nervous system. Nat. Rev. Neurosci. 14, 265–277.
Papadopoulos, M.C., et al., 2004. Aquaporin-4 facilitates reabsorption of excess fluid in vasogenic brain edema. FASEB J. 18, 1291–1293.
Papouin, T., et al., 2017. Astrocytic control of synaptic function. Philos. Trans. R. Soc. Lond. Ser. B Biol. Sci. 372.
Parpura, V., et al., 1994. Glutamate-mediated astrocyte-neuron signalling. Nature. 369, 744–747.
Parpura, V., et al., 2016. Plasmalemmal and mitochondrial Na(+) -ca(2+) exchange in neuroglia. Glia. 64, 1646–1654.
Pascual, O., et al., 2005. Astrocytic purinergic signaling coordinates synaptic networks. Science. 310, 113–116.
Pauletti, A., et al., 2019. Targeting oxidative stress improves disease outcomes in a rat model of acquired epilepsy. Brain. 142, e39.
Pearson-Smith, J.N., Patel, M., 2017. Metabolic dysfunction and oxidative stress in epilepsy. Int. J. Mol. Sci. 18.
Pellerin, L., Magistretti, P.J., 1994. Glutamate uptake into astrocytes stimulates aerobic glycolysis: a mechanism coupling neuronal activity to glucose utilization. Proc. Natl. Acad. Sci. U. S. A. 91, 10625–10629.
Petzold, G.C., Murthy, V.N., 2011. Role of astrocytes in neurovascular coupling. Neuron. 71, 782–797.
Pitkanen, A., Lukasiuk, K., 2009. Molecular and cellular basis of epileptogenesis in symptomatic epilepsy. Epilepsy Behav. 14 (Suppl. 1), 16–25.
Plata, A., et al., 2018. Astrocytic atrophy following status epilepticus parallels reduced ca (2+) activity and impaired synaptic plasticity in the rat Hippocampus. Front. Mol. Neurosci. 11, 215.
Porkka-Heiskanen, T., et al., 1997. Adenosine: a mediator of the sleep-inducing effects of prolonged wakefulness. Science. 276, 1265–1268.
Portas, C.M., et al., 1997. Role of adenosine in behavioral state modulation: a microdialysis study in the freely moving cat. Neuroscience. 79, 225–235.
Quigg, M., et al., 2016. Insomnia in epilepsy is associated with continuing seizures and worse quality of life. Epilepsy Res. 122, 91–96.
Ralay Ranaivo, H., et al., 2012. Albumin induces upregulation of matrix metalloproteinase-9 in astrocytes via MAPK and reactive oxygen species-dependent pathways. J. Neuroinflammation 9, 68.
Ravizza, T., et al., 2008. Innate and adaptive immunity during epileptogenesis and spontaneous seizures: evidence from experimental models and human temporal lobe epilepsy. Neurobiol. Dis. 29, 142–160.
Rebola, N., et al., 2011. Adenosine A2A receptors control neuroinflammation and consequent hippocampal neuronal dysfunction. J. Neurochem. 117, 100–111.
Rho, J.M., Boison, D., 2022. The metabolic basis of epilepsy. Nat. Rev. Neurol. 18, 333–347.
Riquelme, J., et al., 2020. Gliotransmission: a novel target for the development of Antiseizure drugs. Neuroscientist. 26, 293–309.
Rodriguez, J.J., et al., 2009. Astroglia in dementia and Alzheimer's disease. Cell Death Differ. 16, 378–385.
Rojas, A., et al., 2019. The COX-2/prostanoid signaling cascades in seizure disorders. Expert Opin. Ther. Targets 23, 1–13.
Rose, C.R., et al., 2020. On the special role of NCX in astrocytes: translating Na(+)-transients into intracellular ca(2+) signals. Cell Calcium 86, 102154.
Rouach, N., et al., 2008. Astroglial metabolic networks sustain hippocampal synaptic transmission. Science. 322, 1551–1555.
Rouault, T.A., 2013. Iron metabolism in the CNS: implications for neurodegenerative diseases. Nat. Rev. Neurosci. 14, 551–564.
Sada, N., et al., 2015. Epilepsy treatment. Targeting LDH enzymes with a stiripentol analog to treat epilepsy. Science. 347, 1362–1367.
Saito, R., et al., 2009. Involvement of Na+/Ca2+ exchanger in pentylenetetrazol-induced convulsion by use of Na+/Ca2+ exchanger knockout mice. Biol. Pharm. Bull. 32, 1928–1930.
Sammaritano, M., et al., 1985. Prolonged focal cerebral edema associated with partial status epilepticus. Epilepsia. 26, 334–339.
Sandau, U.S., et al., 2019. Transient use of a systemic adenosine kinase inhibitor attenuates epilepsy development in mice. Epilepsia. 60, 615–625.
Sander, J.W., 2003. The epidemiology of epilepsy revisited. Curr. Opin. Neurol. 16, 165–170.
Sander, J.W., Shorvon, S.D., 1996. Epidemiology of the epilepsies. J. Neurol. Neurosurg. Psychiatry 61, 433–443.
Sano, F., et al., 2021. Reactive astrocyte-driven epileptogenesis is induced by microglia initially activated following status epilepticus. JCI. Insight. 6.
Schousboe, A., et al., 2014. Glutamate metabolism in the brain focusing on astrocytes. Adv. Neurobiol. 11, 13–30.
Schwartzkroin, P.A., Baraban, S.C., Hochman, D.W., 1998. Osmolarity, ionic flux,and changes in brain excitability. Epilepsy Res. 32 (1–2), 275–285.
Seiffert, E., et al., 2004. Lasting blood-brain barrier disruption induces epileptic focus in the rat somatosensory cortex. J. Neurosci. 24, 7829–7836.
Shao, Y., et al., 2016. Inhibition of p38 mitogen-activated protein kinase signaling reduces multidrug transporter activity and anti-epileptic drug resistance in refractory epileptic rats. J. Neurochem. 136, 1096–1105.
Sharma, V., et al., 2007. Iron-induced experimental cortical seizures: electroencephalographic mapping of seizure spread in the subcortical brain areas. Seizure. 16, 680–690.
Shigetomi, E., et al., 2019. Aberrant calcium signals in reactive astrocytes: a key process in neurological disorders. Int. J. Mol. Sci. 20.
Simeone, T.A., et al., 2018. Do ketone bodies mediate the anti-seizure effects of the ketogenic diet? Neuropharmacology. 133, 233–241.
Sofroniew, M.V., 2009. Molecular dissection of reactive astrogliosis and glial scar formation. Trends Neurosci. 32 (12), 638–647. PMC2787735.
Staley, K., 2015. Molecular mechanisms of epilepsy. Nat. Neurosci. 18, 367–372.
Strohschein, S., et al., 2011. Impact of aquaporin-4 channels on K+ buffering and gap junction coupling in the hippocampus. Glia. 59, 973–980.
Studer, F.E., et al., 2006. Shift of adenosine kinase expression from neurons to astrocytes during postnatal development suggests dual functionality of the enzyme. Neuroscience. 142, 125–137.
Sudbrack-Oliveira, P., et al., 2019. Sleep architecture in adults with epilepsy: a systematic review. Sleep Med. 53, 22–27.
Szokol, K., et al., 2015. Augmentation of Ca(2+) signaling in astrocytic endfeet in the latent phase of temporal lobe epilepsy. Front. Cell. Neurosci. 9, 49.
Szu, J.I., et al., 2020a. Aquaporin-4 dysregulation in a controlled cortical impact injury model of posttraumatic epilepsy. Neuroscience. 428, 140–153.
Szu, J.I., et al., 2020b. Modulation of posttraumatic epileptogenesis in aquaporin-4 knockout mice. Epilepsia. 61, 1503–1514.
Takahashi, D.K., et al., 2010. Increased coupling and altered glutamate transport currents in astrocytes following kainic-acid-induced status epilepticus. Neurobiol. Dis. 40, 573–585.
Tang, F.R., et al., 2004. Expression of different isoforms of protein kinase C in the rat hippocampus after pilocarpine-induced status epilepticus with special reference to CA1 area and the dentate gyrus. Hippocampus. 14, 87–98.
Terrone, G., Salamone, A., Vezzani, A., 2017. Inflammation and Epilepsy: Preclinical Findings and Potential Clinical Translation. Curr. Pharm. Des. 23 (37), 5569–5576.
Terrone, G., et al., 2020. Inflammation and reactive oxygen species as disease modifiers in epilepsy. Neuropharmacology. 167, 107742.
Tian, G.F., et al., 2005. An astrocytic basis of epilepsy. Nat. Med. 11, 973–981.
Tishler, D.M., et al., 1995. MDR1 gene expression in brain of patients with medically intractable epilepsy. Epilepsia. 36, 1–6.
Toft-Bertelsen, T.L., et al., 2021. Clearance of activity-evoked K(+) transients and associated glia cell swelling occur independently of AQP4: a study with an isoform-selective AQP4 inhibitor. Glia. 69, 28–41.
Tonnesen, J., et al., 2018. Super-resolution imaging of the extracellular space in living brain tissue. Cell. 172 (1108–1121), e15.
Toti, K.S., et al., 2016. South (S)- and north (N)-Methanocarba-7-Deazaadenosine analogues as inhibitors of human adenosine kinase. J. Med. Chem. 59, 6860–6877.
Umpierre, A.D., et al., 2019. Conditional Knock-out of mGluR5 from astrocytes during epilepsy development impairs high-frequency glutamate uptake. J. Neurosci. 39, 727–742.
Verkhratsky, A., et al., 2019. Astroglial atrophy in Alzheimer's disease. Pflugers Arch. 471, 1247–1261.
Verkman, A.S., 2009. Physiological importance of aquaporin water channels. Ann. Med. 34, 192–200.
Vezzani, A., et al., 2019. Neuroinflammatory pathways as treatment targets and biomarkers in epilepsy. Nat. Rev. Neurol. 15, 459–472.
Vezzani, A., et al., 2022. Astrocytes in the initiation and progression of epilepsy. Nat. Rev. Neurol. 18, 707–722.
Vianna, E.P., et al., 2002. Evidence that ATP participates in the pathophysiology of pilocarpine-induced temporal lobe epilepsy: fluorimetric, immunohistochemical, and Western blot studies. Epilepsia. 43 (Suppl. 5), 227–229.

van Vliet, E.A., et al., 2005. Expression of multidrug transporters MRP1, MRP2, and BCRP shortly after status epilepticus, during the latent period, and in chronic epileptic rats. Epilepsia. 46, 1569–1580.
van Vliet, E.A., et al., 2007a. Blood-brain barrier leakage may lead to progression of temporal lobe epilepsy. Brain. 130, 521–534.
van Vliet, E.A., et al., 2007b. Region-specific overexpression of P-glycoprotein at the blood-brain barrier affects brain uptake of phenytoin in epileptic rats. J. Pharmacol. Exp. Ther. 322, 141–147.
van Vliet, E.A., et al., 2010. COX-2 inhibition controls P-glycoprotein expression and promotes brain delivery of phenytoin in chronic epileptic rats. Neuropharmacology. 58, 404–412.
van Vliet, E.A., et al., 2020. Long-lasting blood-brain barrier dysfunction and neuroinflammation after traumatic brain injury. Neurobiol. Dis. 145, 105080.
Vossel, K.A., et al., 2016. Incidence and impact of subclinical epileptiform activity in Alzheimer's disease. Ann. Neurol. 80, 858–870.
Wallraff, A., et al., 2006. The impact of astrocytic gap junctional coupling on potassium buffering in the hippocampus. J. Neurosci. 26, 5438–5447.
Walz, W., 1992. Role of Na/K/cl cotransport in astrocytes. Can. J. Physiol. Pharmacol. 70 (Suppl), S260–S262.
Walz, W., Hertz, L., 1983. Functional interactions between neurons and astrocytes. II. Potassium homeostasis at the cellular level. Prog. Neurobiol. 20, 133–183.
Watanabe, Y., et al., 2011. Participation of metabotropic glutamate receptors in pentetrazol-induced kindled seizure. Epilepsia. 52, 140–150.
Wheless, J.W., 2008. History of the ketogenic diet. Epilepsia. 49 (Suppl. 8), 3–5.
Williams-Karnesky, R.L., et al., 2013. Epigenetic changes induced by adenosine augmentation therapy prevent epileptogenesis. J. Clin. Invest. 123, 3552–3563.
Willmore, L.J., et al., 1978. Recurrent seizures induced by cortical iron injection: a model of posttraumatic epilepsy. Ann. Neurol. 4, 329–336.
Xu, L., et al., 2009. Impaired astrocytic gap junction coupling and potassium buffering in a mouse model of tuberous sclerosis complex. Neurobiol. Dis. 34, 291–299.
Yao, X., et al., 2008. Aquaporin-4-deficient mice have increased extracellular space without tortuosity change. J. Neurosci. 28, 5460–5464.
Yegutkin, G.G., Boison, D., 2022. ATP and adenosine metabolism in Cancer: exploitation for therapeutic gain. Pharmacol. Rev. 74, 797–822.
Yegutkin, G.G., et al., 2011. Chronic hypoxia impairs extracellular nucleotide metabolism and barrier function in pulmonary artery vasa vasorum endothelial cells. Angiogenesis. 14, 503–513.
Yoon, B.E., Lee, C.J., 2014. GABA as a rising gliotransmitter. Front. Neural. Circ. 8, 141.
Younus, I., Reddy, D.S., 2017. Epigenetic interventions for epileptogenesis: a new frontier for curing epilepsy. Pharmacol. Ther. 177, 108–122.
Zamanian, J.L., Xu, L., Foo, L.C., Nouri, N., Zhou, L., Giffard, R.G., Barres, B.A., 2012. Genomic analysis of reactive astrogliosis. J. Neurosci. 32 (18), 6391–6410. PMC3480225.
Zhao, J., et al., 2017. Therapeutic potential of an anti-high mobility group box-1 monoclonal antibody in epilepsy. Brain Behav. Immun. 64, 308–319.
Zhao, J., et al., 2022. Activated astrocytes attenuate neocortical seizures in rodent models through driving Na(+)-K(+)-ATPase. Nat. Commun. 13, 7136.
Zhao, J.L., et al., 2020. HMGB1 is a therapeutic target and biomarker in diazepam-refractory status epilepticus with wide time window. Neurotherapeutics. 17, 710–721.
Zimmer, T.S., et al., 2021. Seizure-mediated iron accumulation and dysregulated iron metabolism after status epilepticus and in temporal lobe epilepsy. Acta Neuropathol. 142, 729–759.
Zurolo, E., et al., 2011. Activation of toll-like receptor, RAGE and HMGB1 signalling in malformations of cortical development. Brain. 134, 1015–1032.