Warped rhythms: Epileptic activity during critical periods disrupts the development of neural networks for human communication

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Abstract
It is well established that temporal lobe epilepsy—the most common and well-studied form of epilepsy—can impair communication by disrupting social-emotional and language functions. In pediatric epilepsy, where seizures co-occur with the development of critical brain networks, age of onset matters: The earlier in life seizures begin, the worse the disruption in network establishment, resulting in academic hardship and social isolation. Yet, little is known about the processes by which epileptic activity disrupts developing human brain networks. Here we take a synthetic perspective—reviewing a range of research spanning studies on molecular and oscillatory processes to those on the development of large-scale functional networks—in support of a novel model of how such networks can be disrupted by epilepsy. We seek to bridge the gap between research on molecular processes, on the development of human brain circuitry, and on clinical outcomes to propose a model of how epileptic activity disrupts brain development.
Introduction.

Before 10 years of age, one out of every 150 children is diagnosed with epilepsy, and approximately 20% of these cases of epilepsy are resistant to drug treatment. In pediatric epilepsy, where seizures co-occur with the development of critical brain networks, age of onset matters: The earlier in life seizures begin, the worse the disruption in network establishment, resulting in academic hardship and social isolation. Indeed, childhood epilepsy is associated with a number of resultant problems, which often have a larger impact on patients’ lives than the seizures themselves. Relative to healthy controls, children with epilepsy show marked impairment in language development, with poorer outcomes associated with earlier seizure onset. They also suffer overall poorer mental health associated with lower social competence and more interpersonal problems. Additionally, adults with childhood-onset epilepsy have difficulty establishing and maintaining marital relationships and experience greater social isolation than healthy controls, suggesting difficulties persist throughout the lifespan. In particular, temporal lobe epilepsy (TLE)—the most common and well-studied form of epilepsy—can impair communication by disrupting social-emotional and language functions (for review see ). TLE in particular is associated with high incidence of depression and anxiety, which have been associated with gray matter volume loss and altered medial temporal lobe connectivity with prefrontal regions of the brain. With regard to language, in children, processing of sound sequences and durations is less acute, and this low-level challenge likely underlies some receptive and expressive language impairments. Yet how the timing of seizure onset and focus of seizure activity influences the development of brain networks underlying cognitive processes that serve communication remains poorly understood. This knowledge gap, which presents an obstacle to treatment, is the result of multiple challenges to conducting systematic neuroscience research in this population. Such challenges include wide variation in patient age range and heterogeneity in pediatric epilepsy as well as isolation between communities of basic researchers, clinical researchers, and physicians.

Sensitive periods in brain network development.

Just as a radio needs to be tuned to the correct frequency to receive information, nodes of brain networks need to be tuned to receive information from distant regions to function efficiently. In this paper we will propose that such tuning occurs gradually from birth through adolescence within and between networks, as well as during temporally circumscribed sensitive periods in local brain hubs. To lay the foundation for this proposal, we will first review evidence for disruption of large-scale functional brain networks in temporal lobe epilepsy within the context of what is known about development of large scale brain networks.

Healthy brain network development.

Canonical intrinsic networks in adults. A number of large-scale brain networks — comprised of distributed regions whose activities fluctuate together at low frequencies as indexed by the Blood Oxygen Level Dependent (BOLD) response measured with functional magnetic resonance imaging (fMRI) — have been consistently observed in healthy adults. These canonical intrinsic networks also contribute to brain activity observed during specific types of experimental task. For example, activity in the dorsal attention network (DAN), which
subdivides deliberate attentional processes, is not only correlated across the network’s nodes when the mind is left to its own devices\textsuperscript{17} but also activated when explicit attention is paid to goal-relevant aspects of the external world\textsuperscript{20}. In contrast, activity in the default mode network (DMN)\textsuperscript{21,22} observed when participants are not performing an externally-oriented task, is typically anti-correlated with that of externally-oriented “task-positive” networks, including the DAN\textsuperscript{23}, and is active during more internally generated mental processes involving self-relevance, episodic memory and imagination\textsuperscript{24,25}. Another reliably observed intrinsic network, the salience network, is active when motivationally or emotionally relevant events, or “homeostatic challenges,” occur\textsuperscript{18,26}. Recent research using large-scale datasets has supported the hypothesis that intrinsic networks shape activity during experimental tasks and contribute to overall functional network structure.\textsuperscript{19}

Much of the recent research on brain network development and network structure in epilepsy has taken a graph theoretic approach to characterizing networks. Here primary measures of interest are path length and clustering coefficient. Path length indicates the number of functional connections between nodes of a network, with shorter path lengths indexing greater integration and network efficiency. A clustering coefficient measures the likelihood that closely neighboring nodes are connected to each other, and a larger coefficient is a measure of greater segregation. A healthy adult brain shows a configuration that has been classically described as “small-world” (but see\textsuperscript{27}). A small-world configuration is an efficient configuration, characterized by relatively high clustering and short path lengths, which includes a few long-range “short-cuts” between nodes. In contrast a regular network is characterized by long path lengths and high clustering coefficients, and a random network is low on both measures.

Network development in neonates/infancy. Studies investigating network structure and connectivity in infants, as well as extended network development over childhood and adolescence, tell a relatively consistent story. Recent reviews indicate that, at a global scale, macroscale projections are in place\textsuperscript{28} and adult-like properties of network organization are observable at birth.\textsuperscript{28,29} Preterm infants have been found to differ from full-term infants in the quality rather than the layout of connections\textsuperscript{28}; however, in infancy and childhood the brain is more modular and less integrated than in young and middle-aged adults.\textsuperscript{28,30} Long-range “shortcut” connections, which characterize network efficiency in the mature brain, begin to emerge only at around one year and strengthen substantially over the second year of age.\textsuperscript{29} Functional imaging measures indicate that thalamocortical connectivity, as well as inter- and intra-hemispheric connectivity, all increase substantially in the first year.\textsuperscript{28} White matter development of long-range tracts, facilitating conductivity, occurs very rapidly in the first three years, with frontal regions characterized by more gradual trajectories.\textsuperscript{31}

Intrinsic networks. With regard to canonical intrinsic networks, there is evidence that a “proto-intrinsic” network structure is observable in fetuses as well as in pre-term infants.\textsuperscript{32,33} Yet, at birth, specific intrinsic networks show different degrees of configurational maturity, and develop toward more adult-like form at distinct rates that are congruent with what is known about the development of associated cognitive capacities. For example, primary sensory and motor networks show relatively mature configurations at birth, whereas the DMN is restricted to core regions that are relatively fragmented, and undergoes rapid development in the first year\textsuperscript{29,34}. Development of salience and dorsal attention networks is more extended, although
“proto-salience” networks have been identified in neonates.\(^{35}\) Over the first two years, connectivity between the thalamus and default and salience network nodes increases substantially. Moreover, thalamo-salience network connectivity at one year predicts both visuospatial working memory performance and a measures of overall cognitive performance (e.g., visual reception, fine motor performance and expressive and receptive language) at two years\(^{35}\). Unsurprisingly, the frontoparietal control network associated with executive functions is still relatively immature at the end of the first year.\(^{29}\) Different timelines of maturation suggest potentially distinct sensitive periods for the development of hubs or connectivity between them within these networks, but such sensitive periods have yet to be identified.

**Development of intrinsic networks through adolescence.** Overall, studies of structural development indicate an extended process of increasing integration within and primarily decreasing segregation between networks over childhood and adolescence\(^{28}\). Measures of functional connectivity indicate that canonical intrinsic networks continue to differentiate from each other well beyond infancy\(^{26}\). For example, the dissociation between activity in the DMN and DAN, which is not observed in infants, becomes increasingly pronounced over childhood and adolescence\(^{29,37}\), and in 9-10 year olds the degree of anti-correlation is associated with a measure of general cognitive ability\(^{38}\). There also is convergent evidence that in general, between late childhood and adulthood, connections between nodes within networks strengthen and those between networks weaken\(^{39,40}\), conveying both greater network segregation and greater long-range efficiency\(^{41,42}\). Providing a more nuanced view, a large-scale study of 780 participants aged 8-22 years found that, while there was overall greater connectivity within, and less between, canonical networks with age, this was true to different degrees for different intrinsic networks.\(^{43}\) The DMN showed greater within- and between- system connectivity with age, suggesting increasing capacity for flexible co-activation with other networks, and this pattern was associated with better cognitive performance. In contrast, high-order networks showed relatively less within-network connectivity than sensorimotor systems, the better to flexibly fractionalize into specialized subsystems.\(^{43}\) Beyond adolescence, in a lifespan sample of 586 individuals, Petrician and colleagues\(^{44}\) found that intra-network connectivity profiles for ventral attention and auditory networks showed the expected increase in connectivity from childhood to young adulthood. However, such connectivity began to decrease in late middle age. They also found that connectivity between intrinsic networks associated with cognitive control and attention (i.e., cingulo-opercular, fronto-parietal, salience, and ventral attention networks) showed increased inter-network connectivity strength through adolescence. Consistent with other findings of DMN flexibility, the DMN showed different inter-network connectivity trajectories with subcortical and dorsal attention networks with increasing age.

**Development of language networks.** The specific networks/subnetworks that mediate language are particular to the specific language process or task, and distinct networks have different developmental trajectories. Yet, overall, lateralization of language systems develops in early childhood, and is in place by five years of age\(^{45}\). Activation of a widespread network of nodes associated with semantic processing involved in naming has been found to increase gradually with age\(^{45}\), as has structural connectivity within a fronto-temporal network implicated in syntactic processing\(^{46}\). In five-year-olds, the ability to understand syntactically complex sentences has been associated with long-range functional connectivity in this syntactic network.\(^{47}\) Qi and colleagues\(^{48}\) examined structural covariance, or the degree to which gray
matter properties are similar across widely distributed nodes, between fronto-temporal regions within the syntactic network. Here the authors compared covariance of cortical thickness in syntactic network nodes in preschoolers (5-year olds), school-aged children (9-13 year olds), and young adults (18-33 year olds). This study found that covariance patterns developed from being relatively localized in preschoolers to more widespread in adults, and this change was linked to maturation in white matter tracts.\textsuperscript{48} Thus, language networks show a consistent pattern of lateralization over early childhood as well as overall similar maturation patterns of increased within network connectivity that develop at a pace commensurate with language abilities.

\textit{Development of emotion-recognition networks.} Again, development of networks that facilitate social emotional processes varies depending on the specific process or task. Relevant to emotion-recognition deficits in epilepsy, reliable networks are evoked in healthy adults during facial emotion recognition tasks, and show a predictable healthy developmental trajectory. A recent big-data study of 759 participants aged 8-23 identified three subnetworks activated in emotion recognition, which overlapped with components of canonical salience and default mode networks.\textsuperscript{49} This subnetwork structure was observed by the age of eight years and remained relatively stable across later ages. Key hubs for emotion perception were identified in the perigenual anterior cingulate cortex (a key node of the salience network) and the ventromedial prefrontal cortex and dorsomedial prefrontal cortex (nodes of the DMN). This study reported age-related \textit{increases} within and between subnetworks, as well as age-related \textit{decreases} within two of the subnetworks. In the midline (overlapping with DMN) subnetwork, connectivity increased in response to viewing faces expressing anger and fear in a way that mirrored the increase in identification accuracy for those emotions with age.

\textit{EEG synchrony measures of network development.} Another approach to examining the development of network synchronization has been to use EEG to examine patterns of phase synchrony in specific frequency bands. These have found that oscillation patterns continue to shift and gain precision through adolescence, but the trajectory can be non-linear. One study of 68 healthy children and adolescents (aged 8-21 years) found gradual increase of low frequency synchrony until age 15, followed by a destabilization of this pattern, associated with a dip in cognitive performance, which in turn was followed by a later reorganization from 18-21 years.\textsuperscript{50} The reorganization period was characterized by both increased long-range theta synchronization and an increase in both amplitude and frequency within the gamma band. These changes, which the authors describe as a shift from more widespread to more focal distribution, occurred in parallel with improvements in working memory and other aspects of cognitive control.\textsuperscript{50}

\textit{Evidence for late sensitive periods.} Such a reorganization period as the one described above may constitute a sensitive period in adolescence for improvement of high-order cognitive and social-emotional processes.\textsuperscript{51,52} Researchers point to the importance of precise timing of oscillations in tuning synaptic connections for network development.\textsuperscript{53} For example, long-term depression (LTD) and long-term potentiation (LTP) are forms of spike timing dependent plasticity that enable learning. At lower frequencies, stimulation at the peak or trough of the theta cycle can produce LTP/LTD respectively, and the same is true for other frequency bands (beta & gamma). So theta timing must be precise for learning to occur. Developmental shifts in gamma oscillatory activity coincide with the opening of CPs in both rodents and humans, and have been hypothesized to underlie enhanced plasticity during these periods.\textsuperscript{54} Higher-frequency gamma changes are proposed to reflect a change in GABA-ergic transmission (e.g., shift from $a_2$ to $a_1$...
receptors) in regions of dorsolateral prefrontal cortex that play a role in working memory and high-level rule-based decisions. This is important because GABA-ergic interneurons play a crucial role in establishing patterns of synchrony in local circuitry. For example, they can synchronize a whole population of pyramidal neurons and the duration of the inhibitory postsynaptic potential (IPSP) can determine the dominant frequency of local oscillations. With destabilization and reorganization occurring in both theta and gamma activity in mid-adolescence, frequent seizures or spikes could potentially disrupt reorganization; however, for those who experienced early-onset seizures it could also potentially reopen plasticity to offer a second chance for intervention.

**Pre-existing networks constrain network reconfiguration when perturbed**

To understand how spatially distributed networks respond to aberrant neuronal firing, we need to first understand how networks can predictably change with stimulation. Khambhati and colleagues examined how local and global graph network measures changed with neurostimulation in a cohort of 94 adults undergoing intracranial EEG monitoring for refractory epilepsy, and examined how baseline connectivity mediated these differences. Baseline coherence at the stimulation site in the low frequencies (5-15 Hz) was correlated with pre- and post-stimulation increases in node strength, suggesting that previously developed networks constrain the response to stimulation. Thus, altered development of networks associated with TLE may have compounding effects on later seizures, creating a cascading trajectory of altered activation over the long term. We next review network alterations that have been observed in TLE in adults and over development.

**Network alterations in temporal lobe epilepsy**

**Medial temporal lobe connectivity.** Given the locus of TLE, alterations to connectivity within the medial temporal lobes would be expected. A body of research by Bartolomei and colleagues using stereotaxic EEG (sEEG) monitoring in individuals with temporal lobe epilepsy (for review see) found that ictal discharges were primarily limited to medial “limbic” structures (amygdala and hippocampus). At seizure onset, no significant connectivity was observed between these medial structures and lateral temporal regions; however, a rapid discharge was observed over the temporal cortex at seizure onset. Examination of connectivity in medial temporal lobe structures found a significant decrease in power spectrum density from sEEG in the theta frequencies in a medial temporal lobe epilepsy (MTLE) group compared to a control group of epilepsy patients without medial temporal structure involvement. Non-linear correlations between the medial temporal structures were higher for the MTLE patients than the non-MTLE patients. Together these findings suggest an increase in signal interdependencies and reinforced aberrant functional connectivity in MTLE.

**Global connectivity.** At a larger scale, another result of epilepsy may be a less efficient overall brain network organization. The earlier the onset, the more the healthy development of network coordination and efficiency may be disrupted. A recent meta-analysis of 12 studies examining global network organization in adults and children found evidence of longer path lengths and larger clustering coefficients in participants with temporal lobe epilepsy, indicating a more regular pattern of organization. A further shift toward increased regularity has also been observed during (relative to pre) seizures, suggesting that both inter-ictal and ictal patterns of regularity may reinforce each other and possibly maintain a propensity toward seizures.
and colleagues\textsuperscript{62} found global network changes in fMRI BOLD signal also suggesting increased connectivity within the contralateral hemisphere relative to the epileptic hemisphere in 18 of 21 patients. This suggests that long range connectivity is generally reinforced at the expense of more local network connectivity patterns. Yet, it is important to note that in three patients the opposite pattern was observed, and inter-hemispheric connectivity was reduced relative to intra-hemispheric connectivity. These three patients had EEG localization of the seizure onset zone to parieto-occipital regions, rather than frontal or temporal regions, suggesting that different network patterns may emerge depending on localization of the epileptogenic zone. Thus, evidence suggests that alterations in global connectivity, while largely reducing network efficiency, can be manifest in multiple and divergent patterns of global to local connectivity.

\textit{Intrinsic networks.} Beyond global activity, canonical intrinsic network activity is also affected by TLE. Individual studies in adults have observed that DMN and salience network activity are altered in TLE relative to controls\textsuperscript{63-65}, although exact patterns of alteration vary — most likely due to divergent analysis methods and small sample sizes. Convergent evidence indicates coherence between nodes of the DMN is largely decreased in those with TLE, but some nodes may also be more highly connected.\textsuperscript{65} There is also evidence of less efficient, more distributed, and less segregated structure in networks associated with high order cognitive processes in TLE.\textsuperscript{66} Figure 1 shows an example of differing task-related canonical network structures based on seizure frequency for language and mental rotation networks. A child with more frequent seizures (Figure 1b) shows stronger functional BOLD connectivity across tasks and greater connectivity with non-canonical regions than does a child with less frequent seizures (Figure 1a). In this case, the child with greater seizure frequency and connectivity also showed a mild generalized learning disability. This example is consistent with the literature suggesting greater canonical network atypicality is generally associated with a higher incidence of cognitive impairment. For example, pre-surgery patterns of DMN activity in TLE patients have been linked to post-surgical capacity to remember faces and words\textsuperscript{63}. Although small sample sizes require cautious interpretation, functional connectivity patterns between the hippocampus and amygdala and nodes of the DMN\textsuperscript{67}, as well as altered connectivity between the hippocampus (healthy and focal sides) and a number of other brain regions, have also been reported.\textsuperscript{68} As with global connectivity, intrinsic network activity is altered by TLE, but the pattern of alteration depends on the network, the measures used, and the population examined.

\textit{Language networks.} Consistent with language impairments observed behaviorally, alteration in language networks has been observed in adults with TLE (for review see\textsuperscript{69}), with an overall pattern of reduced long-range connectivity\textsuperscript{70,71}. The most commonly reported language impairments involve naming, auditory language comprehension, and language fluency.\textsuperscript{69} One study used machine learning with graph theory indices to examine the relationship between white matter structure and naming ability in 24 adults with medication resistant TLE.\textsuperscript{72} Here researchers measured both local connectivity and whole brain topology, and found that a measure of local connectivity in a network that included anterior and central temporal regions predicted 60% of naming performance. Yet, whereas local subnetwork configuration was predictive of naming impairment, global configuration was not. Another study of 16 adults with
right and 16 with left TLE also took a graph theoretic approach – in this case to examining resting state connectivity between established nodes of language networks as well as relevant DMN nodes (hippocampus and posterior cingulate cortex). They found that more integration of the inferior frontal gyrus, a key language node, with the rest of the network predicted better language, and better integration of the hippocampus on the healthy (non-focal) side of the brain the better the outcome after surgery. Finally, a relatively well-powered study of 59 adults with TLE and 32 controls found that stronger functional connectivity between nodes activated by a naming task predicted naming performance across all participants. Importantly, for participants with left-hemisphere seizure foci, earlier age of onset and longer duration of epilepsy predicted lower connectivity levels. Thus, within language networks, local connectivity patterns have been most closely linked to outcomes, connectivity reductions resulting from left-hemisphere foci may be most linked to poorer naming performance.

Social-emotional networks. Motivated by behavioural findings of reduced capacity for emotion expression in TLE, correlated BOLD activation between regions associated with viewing emotionally expressive faces (relative to landscapes) has also been examined. Here again reduced connectivity has been observed, particularly in those with right lateralized foci, and this pattern was more extreme with earlier age of onset (though due to small sample sizes correlational findings should be treated with caution).

**Putative network perturbation disrupts the formation of developing networks**

*Alterations in brain network activity in pediatric epilepsy and effects of age.* If existing brain networks constrain aberrant network formation in adults, and brain network systems show different developmental trajectories, what is the impact of childhood epilepsy? In children with epilepsy, disruption of the global trend toward increasing within-network integration and between-network segregation with age has been observed. Ibrahim and colleagues found that children with focal epilepsy rather than TLE specifically (in children, foci are more varied and less likely to be temporally located) showed impaired development of regional hubs in the intrinsic salience and default mode network. These were characterized by greater inter- and less intra-network connectivity compared to propensity-score matched controls. Moreover, there were significant interactions between epilepsy duration and network development, suggesting that prolonged epilepsy during childhood may cause progressive alterations in intrinsic network connectivity. Again, this suggests some disruption of the network segregation that occurs in healthy development.

A number of studies examining network patterns in children with benign rolandic (centrotemporal) epilepsy may provide insight into how more intractable temporal lobe epilepsy and associated inter-ictal discharges disrupt the development of brain networks. Several studies of rolandic epilepsy in children report alterations in DMN activity, with decreased connectivity observed between all or some DMN nodes. In temporal lobe epilepsy, lateralization of the epileptogenic zone may play a role in intrinsic network development as well. In a study examining default network connectivity in TLE using magnetoencephalography, Hsiao and colleagues found functional increases in connectivity between the default mode network and the medial temporal lobes in the delta frequency band that were consistent with lateralization of the epileptogenic zone: the right medial temporal lobe was more connected in right lateralized epilepsy, while bilateral medial temporal regions showed increased connectivity in left lateralized
epilepsy. In the theta band, the authors found increased connectivity of the anterior cingulate cortex with the default mode network; however, no lateralization effect was found. Changes in network architecture are also seen outside of resting-state intrinsic connectivity networks and more specifically within task-related networks such as the language and socio-emotional networks.

Disruption of language networks in childhood epilepsy. A number of studies report disruption of activity in language networks, which is associated with problems with semantic comprehension and word generation. Again, it is important to note that sample sizes in these studies are typically too low to reliably observe reliable correlations with age or age of onset. One study compared patterns of activation during a language task (matching a verbal description to an object) in 29 children and adolescents (aged 9-19 years) with focal epilepsy and 20 controls. This study, also using graph theory measures, found that whereas controls used fairly segregated subnetworks to perform the task, the epilepsy group used the whole brain, indicating reduced efficiency during task performance. This measure of activation extent was used effectively to predict group membership (epilepsy vs. control). The epilepsy group also showed a relatively more random topology than controls, and there was evidence of reorganization of left-lateralized language networks.

Within language networks, children with epilepsy have been found to have more diffuse, more randomly organized networks than healthy propensity-matched controls during an auditory discrimination task. Moreover, children with epilepsy showed greater hub reorganization ipsilateral to the seizure onset zone. Within the epilepsy patients, three different language network patterns were identified: (1) typically organized language network (35/58 patients), (2) a less typically organized, left dominant, language network, and (3) an atypically organized, right dominant activation pattern with activations in the homologous regions to the typical, left lateralized language network (8/58 patients). By contrast, only four of the 64 control subjects showed any degree of atypical language lateralization. Overall, this data suggests that children with epilepsy show alterations in the development of their language networks that may reflect a less efficient cognitive strategy.

Language networks, which are typically left lateralized, are known to show atypical lateralization relative to normal controls in children with epilepsy, with increasing levels of atypical lateralization associated with earlier age of seizure onset. Earlier epilepsy likely disrupts lateralization, which is associated with tighter network integration, as well as altering patterns of intrinsic network connectivity.

Effects of inter-ictal discharges (IEDs) as mechanism of network alterations. Major seizures are not the only pattern of epileptic brain activity that can shape networks. Inter-ictal discharges (IEDs) are sub-threshold seizure-like spikes that can occur between seizures. The remote inhibition hypothesis proposes that IEDs disrupt distributed network activity. This hypothesis is supported by findings that IEDs alter activity in several canonical networks in TLE, including the
DMN and networks sensitive to emotional salience and reward\textsuperscript{89} There is also evidence that, beyond interrupting communication between nodes of canonical networks, IEDs can generate their own networks\textsuperscript{90}, which we’ll refer to as spike networks, through repeated iteration of spike-specific co-activation patterns. Such spike networks may in turn play a role in systematically disrupting healthy network development.

One study in children with rolandic epilepsy found reduced DMN activity ONLY in children who experienced IEDs, indicating a key role for IEDs in disrupting DMN activity,\textsuperscript{91} consistent with the remote inhibition hypothesis. Moreover, DMN coherence has been observed to fracture just prior to an inter-ictal spike and is subsequently enhanced during the spike, indicating direct effects of IEDs in DMN activity in real time.\textsuperscript{64} Another study in a population with rolandic epilepsy found altered patterns of structural connectivity between the DMN and language networks to be associated with IEDs.\textsuperscript{92} In this EEG/fMRI study, researchers examined the spike network during IEDs, operationalized as regions showing synchronized activity with rolandic regions thought to be the source of spikes. Over repeated IEDs they observed a network showing greater connectivity between the rolandic opercula and left inferior frontal gyrus, supramarginal gyrus and inferior parietal lobe. They concluded that, because the spike network activated by the IEDs overlapped with canonical network regions that showed reduced connectivity, spike networks likely play a key role in disrupting healthy patterns of network connectivity.\textsuperscript{78,90}

Another study exploited the high temporal resolution of magneto encephalography (MEG) alongside the spatial resolution of fMRI to examine effects of IEDs on oscillatory activity in canonical intrinsic networks (DMN, salience network, DAN, and motor network) in 26 children and adolescents (7-17 years) with intractable focal epilepsy.\textsuperscript{83} Using a graph theoretic approach, these researchers used MEG to dynamically characterize changes in path length and clustering coefficients before and after the onset of IEDs. Overall, they observed greater clustering (more segregation) and reduced path length (greater integration) before and after IEDs at frequencies below 10hz, which is a shift toward a more regular topology. The degree of this change was used to index the ‘vulnerability’ (more change) or ‘resilience’ (less change) of networks. The more resilient a participant’s score was, the stronger the pattern of connectivity in canonical networks and the more canonical hubs contributed to the map. In contrast, the more vulnerable the profile, the more additional regions contributed to canonical maps, a finding interpreted as reflecting a less robustly segregated network. Importantly, the more resilient the participant, the higher their overall IQ score, and resilience accounted for much more variance in IQ than simply the number of IEDs experienced by each participant. The authors conclude that large-scale network changes precede as well as follow inter-ictal discharges, suggesting that reorganization of oscillatory processes within intrinsic networks to a pattern that is more regular may play a causal role in generating IEDs. Reduced differentiation of canonical networks observed in those with higher vulnerability scores runs counter to healthy developmental trajectories characterized by greater within-network organization with age. This study provides preliminary evidence that the degree to which this trajectory is disrupted is directly related to cognitive capacity. Future large-sample studies should examine the degree to which age of onset or duration of epilepsy influence the strength of spike networks and the degree to which canonical networks are disrupted. It should also probe time of onset in relation to estimated sensitive periods in the development of specific network hubs in relation to resultant performance on precise measures of social cognition (e.g., language capacity, emotion recognition, theory of mind).
Balance of inhibition/excitation regulates plasticity

We now will review evidence that the timing of sensitive periods in brain development involves the maturation of inhibitory (GABAergic) networks, which are disrupted in epilepsy. Inhibitory networks also play a pivotal role in setting oscillatory timing. Based on this evidence we will argue that impairments in inhibitory functioning may trigger a manifold disruption to the development of cortical networks, altering the timing of local sensitive periods as well as interrupting inter-regional oscillatory communication.

Brain development occurs through the complex interaction of genetic and environmental elements and processes, leading to reliable functioning in a changing world. Critical periods during development occur when a given brain system is highly plastic in response to environmental influence (Figure 2). Similarly, sensitive periods are windows during which a given brain region or circuit shows elevated plasticity in response to environmental information, but is still malleable in response to environmental change later in life, to a lesser extent. The importance of these periods for fine tuning neuronal responses has long been appreciated in primary sensory areas, including those mediating visual, auditory and somatosensory processing.

Critical periods for more complex behaviors, involving the integration of information across multiple domains, have also been observed, including multimodal integration in barn owls, filial imprinting in geese, and the generation of vocal behavior in humans and songbirds. More recently, evidence for a sensitive period has been shown in the development of memory persistence, providing further support for the idea that brain development involves the interaction of carefully orchestrated critical/sensitive periods during which different systems are calibrated to the environment.

Perhaps the clearest example of the importance of sensitive periods in human development comes from the language domain. Language acquisition is regulated by a number of overlapping critical/sensitive periods that attune the brain to the particular properties of an infant’s language environment. Even in the womb infants have access to the low pass filtered speech signal, an experience which drives newborns’ acoustic preferences. Over the first several years of life, an infant’s perception is further shaped by their language environment, influencing their perception of rhythm, phonetic contrasts, phonotactic rules and visual speech. Perceptual narrowing to the native phonetic repertoire is perhaps the most well studied from a critical period perspective. At 6-8 months old, infants discriminate phonetic contrasts from across the world’s languages, even if that contrast is not used phonemically in the infant’s language environment, or perceived by adult speakers of that language. By 11 months of age, the infant’s perception narrows to match that of the phoneme repertoire of their native language — that is, discrimination of native sound contrasts improves, and discrimination of non-native sound contrasts declines. The stabilization of phonetic categories supports early word learning and is correlated with later language outcomes.

That phonetic attunement occurs within a critical period that is regulated by similar neuronal mechanisms to those elucidated in animal models is supported by several convergent lines of evidence. First, there seems to be a maturational floor before which experience does not alter perception. Despite having three months of additional environmental input, premature infants lose the ability to perceive non-native phonetic contrasts at the same age—from conception as full-term infants. Second, bilingual infants maintain the ability to perceive non-
native sound contrasts longer than their monolingual peers.\textsuperscript{114} This extension of the normal period of perceptual narrowing has been hypothesized to arise because of the relatively reduced input bilingual infants receive in each language, which creates a deprivation situation possibly akin to dark rearing in rodent models.\textsuperscript{104} Third, international adoption studies have found a lasting influence of the birth language environment, even if it is never encountered again, following adoption as early as 6 months of age.\textsuperscript{115} Conversely, lack of exposure even in the first few months and years results in lower levels of both auditory and visual language discrimination in fully bilingual adults.\textsuperscript{116,117} Finally, the critical period for phonetic attunement can be shifted by pharmacological exposures known to alter critical period plasticity in rodent models.\textsuperscript{118}

In the visual/social domain, a similar perceptual narrowing or attunement has also been described for face processing. At 6-months of age, infants not only discriminate between the faces of individual humans, but also discriminate the faces of individual monkeys\textsuperscript{119-121} and sheep.\textsuperscript{122} By 9-months, infants lose the ability to discriminate members of other species. A similar effect has been observed for race, with young infants able to discriminate equally well between members of their own and other races. However, by 9-months, an infant’s performance discriminating other race faces declines.\textsuperscript{123} Studies on premature infants, to directly compare the role experience and maturation play in controlling the opening of the sensitive period for face processing, are lacking. Yet it is established that visual experience in infancy is required for the development of normal face processing abilities in adulthood, with deprivation leading to deficits in both holistic processing and face memory.\textsuperscript{124,125} Additionally, delayed exposure to faces regulates the closure of the sensitive period for species-specific narrowing. In a variation on the visual and auditory deprivation studies, monkeys were raised without exposure to either human or primate faces.\textsuperscript{126} Upon their first exposure to faces, monkeys raised in these conditions were able to discriminate human and primate faces equally well. Following a month of normal experience with either monkeys or humans, however, monkeys’ perception narrowed to the exposed species.

Face processing abilities continue to improve throughout childhood into adolescence. Sensitivity to different aspects of faces, such as identity, emotion and gaze perception, follow different developmental trajectories.\textsuperscript{127} Gaze following and perception, for example, is one of the earliest social/communicative developmental indices. In contrast, improvements in face identification, emotional expression, and facial memory can be seen through adolescence.\textsuperscript{128} It is still unclear whether these improvements are driven by a gradual accumulation of face experience with age, an improvement in general cognitive abilities, or whether there are additional sensitive period windows.\textsuperscript{52} There is some evidence for all three of these accounts, and indeed they are not mutually exclusive. Certainly, experience with faces even in adulthood impacts perception. Exposure to faces where the ‘face space’ (i.e., the distance between and size of different facial features) has been altered systematically affects adults’ identity determination and attractiveness ratings.\textsuperscript{129} Other aspects of facial processing seem to be constrained in a sensitive period manner. The perceptual narrowing that occurs for one’s own race (i.e., the other race effect) can be reversed or mitigated by early exposure to other race faces.\textsuperscript{130-132} However, this experience is most effective in childhood, and even extensive exposure in adulthood appears unable to negate the other race effect.\textsuperscript{132}

More recently, some of the mechanisms known to be involved in regulating critical periods in sensory domains have been found to be involved in hippocampal development as well, leading
to the proposal that developmental changes in memory may reflect critical or sensitive periods in development of the hippocampus and amygdala. Studies in rodents have found that early memory formation drives hippocampal activity, leading to an experience-dependent maturation of hippocampal circuits that is similar to that seen in primary sensory areas. This maturation pattern has been proposed to underlie the phenomenon of infantile amnesia, which is the typical paucity in adults of memories of the first few years of life. In essence, the very act of memory formation at these young ages alters the circuit in an experience-dependent fashion that subsequently renders these memories inaccessible.

There are additional developmental changes in memory that extend beyond infantile amnesia. Memories in early life, once formed, tend to be over generalized, such that until around 10 years of age memories can be recalled using different cues from those present during memory encoding. This overgeneralization can be seen both in fear conditioning paradigms in rodents and humans, as well as explicit memory tasks in humans. This may be related to the finding that, in fear conditioning paradigms, extinction training in juvenile animals appears to overwrite the original fear memory. This is in contrast to adult fear conditioning memories, which can be reinstated even after extensive extinction training. Thus, adults form specific memories that are updated but not erased with new information, while juveniles seem to form less specific memories that are overwritten with new information. In rodents, the resistance of fear memories to extinction is driven by the accumulation of perineuronal nets around parvalbumin containing cells (PV+) in the amygdala, a process that has been linked to critical period closure in a number of different models, and is discussed in detail below. Pharmacological dissolution of these nets prior to memory encoding enables full erasure of the fear memory in adult mice, similar to that seen in juvenile mice. Interestingly, dissolution of the perineuronal nets following memory formation does not result in successful extinction of the memory, suggesting that the state of PV+ cells during memory encoding is what renders the memory susceptible to erasure, as opposed to the state of PV+ cells during relearning. A similar mechanism may be involved in the encoding of memory in the hippocampus. Indeed, the formation of new memories is regulated by the current state of the PV+ cell network. Developmental changes in PV+ cells would therefore be predicted to alter memory encoding in the hippocampus.

The juvenile period during which memories are more easily overwritten seems to be followed by a period of enhanced memory stability. Researchers have observed a propensity for individuals to preferentially recall life events that occur between the ages of 10-30 years old, termed the reminiscence bump. This phenomenon has been documented across different cultures and for autobiographical memories as well as for preferences in music and books. Such findings have given rise to the idea that this juvenile window may represent a late sensitive period for memory formation.

Work in animal models has elucidated the cellular and molecular mechanisms regulating the timing at which critical/sensitive windows occur during development (Figure 2). The opening of these windows is controlled by the maturation of inhibitory circuitry in the brain. The cortical inhibitory circuit is made up of a diverse array of GABAergic neuronal subtypes, defined by their morphology, circuit localization and gene expression. The majority of cortical interneurons fall within four main classes, expressing either parvalbumin (PV), somatostatin (SST), 5HT3aR and vasoactive intestinal peptide or 5HT3aR and reelin. Accumulating evidence points to a subclass of fast-spiking inhibitory interneurons expressing the Ca2+ binding protein parvalbumin (PV+) as
a critical hub in regulating critical period plasticity. However, the other inhibitory cell types also play a role, either by modulating PV+ cell activity or directly altering pyramidal cell excitability. Several studies point to an important role for somatostatin cells, although it has been argued that they may play a larger role in cortical plasticity in adulthood. In this review we focus on the role of PV+ cells, with numerous studies pointing to a pivotal role for this cell type in regulating the timing of CP plasticity.

Inhibitory cell maturation lags behind the development of excitatory cells, with migration from the ganglionic eminences continuing for several months postnatally in the human brain. Both PV+ and SST+ cells are born in the medial ganglionic eminence, and integrate into cortical circuits following a similar developmental timeline. PV+ cells integrate into the cortical circuit and gradually mature, with changes in their intrinsic membrane properties, increased PV and GABA content, and shifts in receptor composition and excitatory input leading to their precise, fast-spiking behavior. PV+ cells are gradually ensheathed in perineuronal nets, extracellular matrix proteins that regulate the accumulation of molecular factors supporting PV+ cell maturation, including orthodentical homeobox 2 (Otx2), semaphorin3A, and NARP. The timing of PV cell maturation is staggered across the cortex, and in primates continues into adolescence in prefrontal cortical regions. The course of this development is regulated by the complex interplay between an intrinsic maturational program and environmental input, regulating both the opening of plasticity and the circuit’s eventual stabilization and critical period closure.

Alterations in the trajectory of inhibitory cell maturation shift the timing of critical periods during development. Subtle genetic disruptions in mice have enabled the careful dissection of the contribution of inhibitory maturation to critical period plasticity. Disruption of the GABA synthetic enzyme GAD65 results in a slight reduction in inhibitory signaling, which does not impair gross brain development. However, the opening of critical period plasticity is indefinitely delayed in these mice. This phenotype is replicated through the specific deletion of α1-containing GABA receptors, which are preferentially expressed on PV+ basket cells. Treatment with pharmacological agents that boost inhibitory signaling, such as benzodiazepines, rescue plasticity in these mice and also trigger a precocious plasticity in young wild type mice. Promoting inhibitory cell maturation either directly (e.g., through early removal of the permissive morphogenetic factor polysialic acid (PSA) or infusion of Otx2) or indirectly (through environmental enrichment) accelerates the onset of critical periods. In contrast, sensory deprivation delays both the maturation of inhibitory circuitry and the closure of the critical period. This delay can be prevented with brain-derived neurotrophic factor (BDNF) treatment, which triggers the maturation of inhibitory cells in the absence of environmental experience.

Ultimately, full maturation of the PV+ cell circuit appears to dampen plasticity in adult animals. Neurotrophic factors involved in development also serve to actively maintain the excitatory/inhibitory balance of the cortical circuit in adult mice. Neuregulin-1 regulates the level of excitatory input to PV+ cells, and disruption of the neuregulin/ErbB4 signaling pathway reduces PV+ cell activation, leading to a reactivation of critical period like plasticity in adult animals. Interestingly, neuregulin-1 treatment during the CP prevents plasticity in response to eye occlusion and prematurely closes the CP. This work highlights the fact that interventions that alter inhibitory signaling in the cortex may enhance or disrupt plasticity.
depending on the initial state of the inhibitory circuit. Whereas boosting inhibition triggers a precocious CP in juvenile mice, treatments such as environmental enrichment and fluoxetine treatment, which reopen plasticity in adult animals, decrease PV+ cell strength.\textsuperscript{170,171,172} Thus, PV+ cells appear to go through a ‘goldilocks moment’ during development, where inhibition and excitation are optimally balanced to support plasticity.

Whereas the levels of various molecular factors mentioned above are controlled by environmental experience, and altered by exposures such as dark rearing and environmental enrichment, evidence suggests that a cell’s autonomous maturational clock also controls the timing of plasticity. Transplantation of immature inhibitory neurons from the ganglionic eminence into the visual cortex of an adult mouse reopens visual cortical plasticity.\textsuperscript{148,173} The timing of this reopening occurs approximately one month after transplant, once the transplanted cells reach an age that corresponds with the opening of critical period plasticity in the visual system of the donor. Reopening of the CP is supported by the transplantation of either PV+ or SST+ cells from the medial ganglionic eminence, further suggesting that both cell types play a role in regulating CP plasticity. This developmental program is modified by the hosts’ cortical environment, where transplanted cells are innervated and rapidly enwrapped in perineuronal nets.\textsuperscript{174}

In adulthood, plasticity is actively suppressed by a number of molecular brakes.\textsuperscript{104} Disruption of these brakes resets the excitatory/inhibitory balance, reopening plasticity. As discussed briefly above, PV+ cells are gradually enwrapped by perineuronal nets as they develop. Environmental input drives the release of developmental regulators such as Otx2, a cell-extrinsic factor that serves to both mature and maintain perineuronal net integrity.\textsuperscript{154} Through a positive feedback loop, Otx2 drives the gradual enrapuretion of PV+ cells with perineuronal nets. In turn, these nets support Otx2 uptake from the cortical milieu. Removal of either perineuronal nets or Otx2 in adulthood resets PV+ cells to a juvenile state, transiently reinstating critical period levels of plasticity.\textsuperscript{175-178} Insulin like growth factor 1 (IGF-1) is another factor that supports PNN formation, and may serve as one path through which environmental experiences impact inhibitory circuit maturation.\textsuperscript{179} Proteins associated with myelin, including the Nogo receptor and paired immunoglobulin-like receptor B complex, also actively limit plasticity, and their disruption enables heightened plasticity beyond the closure of the normal critical period.\textsuperscript{180,181}

The elucidation of these molecular mechanisms has led to proposals that critical period plasticity may be disrupted in developmental disorders.\textsuperscript{182,183} Of particular interest for this review, the co-morbidity of autism and epilepsy is cited as evidence for an imbalance between excitatory and inhibitory signaling in the brains of these individuals.\textsuperscript{184} This in turn has inspired theories that critical period plasticity may be disrupted in autism spectrum disorder, and that this disruption may contribute to the etiology of autism.\textsuperscript{182,185,186} However, how this imbalance between excitatory and inhibitory signaling alters brain development in pediatric epilepsy, specifically in the context of critical periods, has received surprisingly little attention.

It has been hypothesized that the cognitive deficits observed in pediatric epilepsy patients, especially in the realms of language development or social cognition, may be driven not by damage to the cortical tissue, but by disruption of the normal developmental of cortical regions or network hubs.\textsuperscript{187} Thus, seizures may perturb normal attunement of the system to environmental input. Many of the key molecular players regulating the maturation of inhibitory circuitry in the brain and the timing of critical periods are disrupted by epileptogenesis. Whether
triggered by the initial seizure activity or as a homeostatic response, early life seizures have been linked to a shift in glutamatergic receptor composition, an increase in BDNF expression, perineuronal net degradation, and increased microglial activity.\textsuperscript{187-189}

To date, only one study has directly investigated whether early life seizure activity alters critical period plasticity. The tonotopic organization of primary auditory cortex is refined during an early postnatal critical period, and passive tone exposure during this time alters thalamocortical input, resulting in an over-representation of the exposed tone. This plasticity is dependent on a maturational shift from ‘silent’, NMDA-containing thalamocortical synapses to functional synaptic connections following the insertion of AMPA receptors. Pharmacologically induced seizures prior to this critical period trigger precocious ‘unsilencing’ of NMDA-only synapses, preventing normal plasticity in response to environmental change.\textsuperscript{190} Importantly, seizure activity did not disrupt normal tonotopic map organization, instead it occluded the reorganization in response to environmental change. It is interesting to consider whether seizure activity completely suppressed critical period plasticity in these mice, or whether it instead shifted the timing of this window (Figure 2). Given the fact that seizure activity sped up the typical maturational unsilencing of NMDA only synapses, it is possible that the normal critical period was similarly shifted earlier instead of being completely disrupted. This has important implications for the impact of seizure activity on brain development in humans, as altering the timing of cortical plasticity as opposed to preventing it all together may result in different developmental outcomes.

Additionally, there is evidence that perinatal brain injury, one of the major underlying causes of neonatal seizures,\textsuperscript{191} alters critical period plasticity. Cortical subplate neurons are particularly vulnerable to early hypoxia-ischemic injury.\textsuperscript{192} Pharmacological ablation of this population of cells in mice disrupts the maturation of cortical inhibitory cells, lowering KCC2 expression and preventing the typical switch of GABA induced currents from depolarizing to hyperpolarizing.\textsuperscript{193} Disrupting this switch drives a paradoxical plasticity in the visual cortex, in which experience during the critical period now results in the expansion of territory for the deprived as opposed to the non-deprived eye. Neonatal hypoxia-ischemia in rats produced similar results, leading to the death of subplate neurons, cortical inhibitory cell impairment, and disrupted critical period plasticity.\textsuperscript{194,195}

We now review evidence that disruption of the KCC2 chloride transporter, resulting in loss of GABAergic inhibition, plays a critical role in generating epileptic foci and disrupts the normal development of GABA receptors.\textsuperscript{196,197}

**KCC2 and GABA.** We have seen how maturation of inhibitory GABA circuitry triggers the onset of plasticity in neural circuits across the brain by achieving an optimal excitation-inhibition balance, beginning an experience-sensitive period of development, and also how this plasticity is halted by a variety of molecular “brakes.” A mature and well-functioning GABA system is thus critical for the development of functional brain circuits in every region of the brain. Thus, any factor that affects excitation-inhibition balance might affect the onset, offset, and outcomes of critical periods in a particular brain area, and as well have cascading effects on the development of connected brain regions. In this section, we will describe a molecular mechanism that is clearly involved in epilepsy and that might be especially important during the development of GABAergic circuitry.
Our discussion here will focus on a fact about GABA synapses that has been fairly recently discovered: in immature neurons GABA<sub>A</sub> synapses are excitatory rather than inhibitory.\textsuperscript{197-199} That is, when the GABA<sub>A</sub> receptor binds a1-GABA, the result is a depolarization of the pyramidal neuron, causing its intracellular potential to move closer to its spiking threshold (around -50mV), and thus increasing the probability that it will produce a spike potential given other synaptic input. As the neuron matures normally, the effects of GABA gradually become inhibitory. Clearly, unless the GABA circuitry is inhibitory, the net effect of any input to such neurons will be excitatory. This could have at least two important effects: (1) it can disrupt normal plasticity during critical/sensitive periods,\textsuperscript{193} and (2) it will promote excitatory pyramidal neurons sending axonal spike potentials to other neurons to which they are connected, including to those in other brain areas to which they are structurally connected, thus exciting them — potentially triggering ictal activity. It is thus important to understand how this mechanism works and how it can be impaired.

When in development could these effects become important? Apparently all immature neurons have this character, whenever they appear - i.e. if the hippocampus is continuously generating new neurons\textsuperscript{200} they would be of this type for a time period. Any (most) neurons in newborns are immature and would mature over various time periods but typically would enter the mature stage fairly soon depending on brain area – for early sensory critical periods this would be early – typically within the first year. But, if maturity is delayed for some reason, then they could continue to exhibit excitatory GABA into even adulthood – as when there are dysplasias, ectopias, etc. The hippocampus would be expected to be most vulnerable to late-emerging neurons that have excitatory GABA, as one of the only regions in the brain where new neurons continue to be incorporated into the circuit throughout life (but see Kempermann et al., 2018 for renewed debate on the presence of hippocampal neurogenesis in humans\textsuperscript{201}). Synaptogenesis and pruning occur at high rates early and diminish later but still occur at significant rates into the twenties.\textsuperscript{202} To the extent that GABA immaturity affects synaptic processes it could have effects even this late.

The Cl<sup>-</sup> ion is critical to the normal functioning of a GABA<sub>A</sub> synapse. Typical neurons at equilibrium maintain a negative charge of about -70 mV inside the neuron relative to the outside. This is the result of different amounts of various ions and some electrically charged proteins inside and outside the neuron. Because opposite charges attract, there is an electrostatic pressure for positive ions to enter the cell, and for negative ions to exit. In addition, when there are fewer ions of a particular type inside than outside, or vice versa, there is a chemical pressure for ions to travel along the gradient from higher to lower concentration. A neuron at equilibrium has more K+ ions and negatively charged proteins inside the cell, and more Na+ and Cl<sup>-</sup> ions outside. Ions can move through the cell membrane via various ion channels, whereas the proteins cannot. Although there are leaks of Na+ and K+ ions into and out of the cell, the most important mechanism to maintain the respective electro-chemical gradients of the Na+ and K+ ions is the ubiquitous energy consuming NaK-ATPase pump, which transports 3 Na+ ions out of the cell for every 2 K+ ions it transports in.

A mature neuron with GABA<sub>A</sub> synapses has its internal Cl<sup>-</sup> concentration maintained by several co-transporter mechanisms, especially NKCC1 and KCC2 (Figures 3 and 4).\textsuperscript{203} The NKCC1 co-transporter uses the Na<sup>+</sup> electro-chemical gradient to bring Na+, K+ and Cl<sup>-</sup> ions into the neuron, whereas the KCC2 co-transporter uses the K+ electro-chemical gradient to transport K+.
and Cl- ions out of the neuron. In a mature neuron, the KCC2 co-transporter is much more active than the NKCC1, maintaining intracellular Cl- at around 7mM, much lower than the extracellular concentration, which averages around 140 mM.\textsuperscript{198} This large difference in concentration means that the chemical Cl- gradient is sufficiently large to cause Cl- to flow into the neuron against the electrical gradient when GABA\textsubscript{A} receptors open. This tends to hyperpolarize the neuron, reducing its potential below the resting level, and making it more difficult for excitatory potentials at other synapses to cause it to fire: i.e., inhibition is the result.

In the immature neuron, in contrast, KCC2 expression is low and NKCC1 expression is dominant, meaning that the intracellular Cl- concentration is maintained at around 20-25 mM.\textsuperscript{69} The smaller difference between intracellular and extracellular concentration of Cl- is insufficient to force Cl- into the neuron against the electrical gradient pushing it out, and thus Cl- flows out of the neuron when GABA\textsubscript{A} receptors open, causing a depolarization, and thus increasing excitability. As the neuron matures, the normal expression of the SLC12A2 (NKCC1) and SLC12A1 (KCC2) genes reverses, and KCC2 comes to dominate the Cl- concentration, making inhibition the typical effect of GABAergic stimulation. This effect is caused by activity-dependent GABA release at these synapses. Importantly, the depolarizing function of early-developing GABAergic synapses is useful, for example these synapses are very plastic and can affect synaptic transmission, but only when depolarizing. And, although the maturational trajectory of GABA is complex, we will point to one particular factor in this trajectory that seems to be of some importance: the abundance of BNDF. This protein is activity-dependent and likely controls the maturation of inhibitory circuits, at least in V1 via an interaction with KCC2 expression. We will see later that this factor affects how GABA\textsubscript{A} synapses function at pyramidal neurons. If, however, for some reason the KCC2 proteins are not expressed, or are malfunctioning in some way, then the inhibitory effect of GABA will not develop – the Cl- level inside the neuron will remain high and GABA synapses will continue to act to depolarize the cell.

With respect to epilepsy, this constitutes a possible mechanism by which an excitatory neural cascade can be initiated and sustained: KCC2 malfunctioning results in a higher than normal concentration of Cl- ions inside of some subset of neurons, causing them to fire more when stimulated by activity at GABAergic synapses, instead of being damped by the activity at those synapses. This excitatory effect then cascades across the circuits in which those neurons are involved, resulting in epileptiform activity or even a seizure. Indeed, several studies in both animals and humans have found that KCC2 malfunctioning is closely associated with epileptiform neural activity and with epilepsy itself. We now briefly review those studies.

\textbf{Animal models.} A number of animal studies have implicated the KCC2, Cl-, excitatory GABA complex in induction of epileptic seizures. Rivera et al.\textsuperscript{204} reported that, both in rat hippocampus cultures and in acute slices, administration of brain-derived neurotrophic factor (BDNF; TrkB is its receptor) induced a TrkB-mediated fall in KCC2 mRNA and protein, and an accompanying impairment in neuronal Cl- extrusion capacity. When they induced seizures by kindling in mice, expression of KCC2 was decreased in hippocampus in the same places and over the same times as expression of TrkB and BDNF were increased. Thus, it seems that BDNF/TrkB – induced decreases in KCC2 activity, leading to suppression of chloride-dependent fast GABAergic inhibition, are closely associated with seizure genesis. Later, Rivera, et al.\textsuperscript{205} found that sustained IED-like activity in hippocampal slices decreased KCC2 mRNA and protein expression in CA1
pyramidal neurons, again reducing capacity for neuronal Cl⁻ extrusion. As in their earlier study, this decrease in KCC2 expression was mediated by an increase in BDNF. Nabekura, et al.²⁰⁶ found that axotomy (cutting a neuron’s axon) led to a decrease of KCC2 expression, followed by elevation of intracellular Cl⁻, and an excitatory response to GABA. Thus, down regulation of KCC2 may be a general early response involved in various kinds of neuronal trauma. This could be a mechanism whereby brain injury can lead to seizures.

Importantly, the way in which KCC2 responds in different models and to different insults may vary. For example, Eftekhari, et al.²⁰⁷ induced seizures in a rat model by injecting pilocarpine into the hippocampus. This increased the expression of NKCC1 and decreased that of KCC2 in epileptic hippocampi compared to intact controls. Application of BDNF, however, actually increased KCC2 expression in epileptic hippocampi, and decreased that of NKCC1. Thus, the role of BDNF in modulating KCC2 remains unclear.

More evidence that down-regulation of KCC2 is involved in seizure genesis comes from a study by Chen, et al.²⁰⁸ They directly (chemically with Cyclothiazide, CTZ) down-regulated hippocampal KCC2 in a rat model, causing epileptiform activity in vitro and seizures and epileptic EEG in vivo. Notably, the KCC2 down-regulation occurred before the seizure-like activity in the hippocampal slices, indicating a causal role for KCC2. Dephosphorylation of pS940 KCC2 residue (destabilizing the KCC2 protein) was also caused by the treatment with CTZ, further decreasing the ability of the neurons to extrude Cl⁻. In this model, NKCC1 level was low and not modulated by the CTZ treatment. On the other hand, overexpression of KCC2, caused by introduction of KCC2-pIRES2-EGFP plasmid to cultured hippocampal cells, protected those cells from CTZ-induced epileptiform bursting. Finally, Chen et al. also used viral vectors to down-regulate KCC2 expression in dentate gyri of living rats. This treatment also resulted in spontaneous seizures in the KCC2 deficient rats.

Interestingly, Goutierre, et al.²⁰⁹ showed that chronic KCC2 knockdown in the in vivo rat hippocampus, induced by a viral vector containing suppressive RNA, has little direct effect on GABA signalling. Instead it seems that lack of KCC2 in hippocampal neurons enhances neuronal excitability and increases network activity by down-regulating membrane expression of Task-3 leak (always open) K⁺ channels, thus changing the amount of K⁺ in the cell. Although the reversal potential for GABA was more depolarized in affected neurons, so was the neurons’ resting potential (unexpectedly), indicating that steady-state (at least) GABA transmission was not significantly affected by the chronic suppression of KCC2. This mechanism is different from the canonical one described earlier, and it remains to be seen if this is the only mechanism by which GABA is rendered excitatory or if it just one of several.

Finally, Campbell, et al.²¹⁰ injected human-derived glioma cells into mice brains and allowed them to grow into gliomas (malignant tumours of the glia). These mice then generated generalized tonic-clonic seizures spontaneously. They found in cortical slices from these mice that there was a loss of parvalbumin-positive GABAergic inhibitory interneurons in the vicinity of the glioma, and the ones that remained had elevated levels of intracellular Cl⁻, and thus depolarizing, excitatory GABA responses. KCC2 membrane expression also was found to be significantly decreased in these neurons. Accompanying these changes in GABAergic interneurons were deficits in spontaneous and evoked inhibitory neurotransmission near the tumour. Thus, it is possible that “…GABAergic disinhibition renders peritumoral neuronal networks hyper-excitable and susceptible to seizures triggered by excitatory stimuli” (p. 23).
Human studies. There exist two recent, comprehensive reviews of the role of KCC2 in epilepsy and other neurodevelopmental disorders, Moore, et al. and Di Christo, et al. These reviews (and the review by Chamma et al.) also describe the many functions of KCC2 in much more detail than we can here, including the mechanisms that regulate KCC2 activity (especially total amount of protein expressed and phosphorylation at the S940 site on the KCC2 protein), KCC2 genetics, interaction with other proteins, and other roles played by KCC2 membrane proteins. One of these roles is of particular importance in the present context, that of participating in the genesis and maintenance of dendritic spines where AMPA and NMDA receptors are located. Using genetic manipulations, animal work has found that acceleration of spine maturation leads to a shortening of the CP for auditory tonotopic map formation, and seizure driven precocious unsilencing of ‘silent’ synapses, which contain only NMDA receptors, disrupts plasticity. They also review some of the human and animal studies. We recommend consulting these reviews for more details. Here we describe a few of the studies that directly implicate KCC2 in human epilepsy.

One of the earliest studies to implicate KCC2 in human epilepsy was done by Huberfeld, et al. They showed that tissue excised from patients with mesial temporal lobe epilepsy, associated with hippocampal sclerosis, generated interictal-like activity and revealed depolarizing GABA receptors. Notably, mRNA for KCC2 was absent from about 30% of pyramidal cells where the interictal activity was generated. They found that, during interictal events, all of the hyperpolarizing neurons were immunopositive for KCC2, whereas the majority of depolarized cells were immunonegative. Interestingly, in this slice preparation, blocking NKCC1 shifted GABA reversal potential to hyperpolarizing and suppressed the interictal activity. So in this case the relative activity of NKCC1 and KCC2 proteins seems to have played a role in generating interictal events.

In a similar study, Munoz, et al. used tissue excised from humans with mesial temporal lobe epilepsy associated with hippocampal sclerosis to prepare slices that in vivo had generated epileptiform activity. They immunolabeled both NKCC (1 and 2) and KCC2 in the slices at the transition between the subiculum and sclerotic regions of CA1 and in the latter regions. They found that about 20% of the NKCC-immunoreactive neurons did not express KCC2. They too suggested that changes in the relative expression of NKCC1 and KCC2 may contribute to epileptiform activity, in this case in the subicular regions adjacent to sclerotic areas of the hippocampus.

More recently, Pallud, et al. studied postoperative tissue from areas of neocortex around gliomas in epileptic patients. They found that the tissue from 69% of patients spontaneously generated high-frequency, oscillating, synchronized interictal-like activity in layers of neocortex near areas of glioma infiltration. Further studies of the tissue slices revealed that the interictal-like events depended both on glutamatergic AMPA receptor-mediated transmission and on depolarizing GABAergic signaling. Importantly, GABA released by interneurons depolarized 65% of pyramidal cells. In these neurons Cl⁻ homeostasis was perturbed because expression of KCC2 was reduced by 42% and expression of NKCC1 increased by 144%. They could induce ictal-like activity using convulsant stimuli in these specific epileptogenic areas but not in other unaffected areas of the specimens.

Jansen, et al. did a post-mortem comparison of frozen specimens from epileptic (focal dysplasia or focal gliosis) and non-epileptic children of a range of ages. They assayed the
specimens for neocortical GABA\textsubscript{A}, NKCC1 and KCC2. The results for the non-epileptic children were similar to those found in animal studies: three subunits of GABA\textsubscript{A} receptors changed over development, KCC2 increased with age through adolescence whereas NKCC1 decreased, and the NKCC1/KCC2 ratio decreased drastically by age 2. This pattern was absent in the specimens from the epileptic children, who typically displayed idiosyncratic levels of various receptor subunits and co-transporters. This result implies that brain areas in which the epileptic children suffered focal lesions related to their epilepsy had different GABA functionality from normal, quite possibly excitatory rather than inhibitory.

Interestingly, in this study epileptic infants less than one year of age did show normal (a\textsubscript{1}) or enhanced (g\textsubscript{2}) expression of GABA receptor subunits that mediate fast, phasic, inhibition, as well as increased levels of KCC2, relative to non-epileptic controls, whereas children greater than one year of age showed decreased expression of all of these relative to controls. This result implies that the older epileptic children might have experienced less of this form of inhibition, whereas the younger infants might have experienced more of it. This is in direct contrast to the normal pattern of excitatory GABA effect in immature neurons and inhibitory effect in mature neurons. This is consistent with some animal results, in which immature rats subjected to status epilepticus developed increased expression of a\textsubscript{1} in the hippocampus, whereas mature rats subjected to the same insult demonstrated decreased a\textsubscript{1} expression. As the expression of the various GABA receptor subunits can be changed by prolonged seizure activity, the distribution of GABA receptor subtypes mediating both phasic and tonic inhibition could be quite different from normal in epileptic children. This would mean that not only would the normal excitatory function of GABA in immature neurons be distorted, with effects on synaptic growth and change, but also the normal inhibitory function of GABA in mature neurons would also be distorted, resulting in GABA enhancing rather than damping excitatory activity in these neurons.

One important aspect of this study for our approach here is that the tissue examined was from gliomas and dysplasias in the children’s brains—implying that when such abnormalities are present, the normal course of development is disrupted. It’s not clear as to when the abnormalities developed in the older children—or on how long is required for an earlier abnormality to trigger seizures that would be treated later. Yet clearly if the abnormality produces excitatory rather than inhibitory activity when the latter is required, this could disrupt critical periods at any age, as the KCC2 mechanism would be dysfunctional during the entire period when the dysplasia or glioma was present. Gliomas are cancers of the glia, which could develop at any age. Dysplasias such as ectopias typically develop early—in utero or in the first year while brain growth is happening rapidly. Thus, it is possible that early critical periods would be more likely to be disrupted when ectopias are present, whereas sensitive periods at any age could be disrupted by the presence of a glioma or other later-developing abnormality such as sclerosis.

Ectopias, abnormal location of granule cells in the dentate gyrus, and other types of microdysgenesis, are associated with TLE as well as with other abnormalities, including dyslexia.\textsuperscript{216-219} In one important study, induction of febrile seizures in a rat model induced abnormal granule cell migration in the dentate gyrus that was precipitated by hyperactivation of excitatory GABA receptors, resulting in ectopias that persisted into adulthood. Interestingly, knockdown of NKCC1 rescued this model from ectopia development\textsuperscript{217}. Moreover, chemical inhibition of NKCC1 activity after the febrile seizures prevented the ectopia from developing, and
also prevented limbic seizures and the development of epilepsy in this model. These results imply that dysplasias, like ectopias and other types of developmental abnormalities, might be involved with generating hyperexcitable foci in TLE. Importantly, febrile seizures, which happen often in infants, can themselves trigger ectopias that could be involved in generating such foci at a later time, with consequences for sensitive periods occurring at the later time.

In a similar lifespan human post-mortem study, Hyde, et al. assayed prefrontal cortex and hippocampal specimens for the expression of genes for GAD1 (GAD25, GAD67), SLC12A2 (NKCC1) and SLC12A1(KCC2). The switch from GAD25 to GAD67 promotes GABA synthesis and GAD25/GAD67 ratio dropped over first 10-15 years of life, which implies that GABA increased during these years. They also found that the NKCC1/KCC2 ratio dropped over the first 10 years then stayed steady until the 50s-60s, although both proteins tended to increase in absolute amounts over the first 10 years of life. This implies that there is more active transport of Na+, K+ and Cl- ions with increasing age.

From these two post-mortem studies it is clear that the complex of genes and proteins that involve KCC2, NKCC1, and GABA function undergo significant changes over the first 2 years of life, and more gradually over at least the first 10 years. Thus, in general, GABA function does not depend on an all-or-nothing switch, and different children will have different trajectories. Gene activations both intrinsically and environmentally (including experience) controlled, determine the trajectory for any particular individual, but there is a lot that is not known. In addition, brain insults or gene mutations also can significantly affect this trajectory.

Finally, genetic studies are beginning to establish links between specific KCC2 gene variants and various forms of epilepsy. Kahle et al. screened the DNA of hundreds of families in which one member had severe idiopathic generalized epilepsy (IGE). They discovered two non-synonymous functional variants in human KCC2 genes, R952H and R1049C, that exhibited a statistical association with IGE. These variants exhibit significantly impaired Cl- extrusion capacities, and impair KCC2 stimulatory phosphorylation at S940. At the same time, Puskarjov, et al. reported that their screening study had revealed one of the same variants (R952H) in an Australian family with febrile seizures. They also showed that this variant reduces neuronal Cl- extrusion and has a compromised ability to induce dendritic spines in vivo and in vitro. Further biochemical analyses indicated reduced surface expression of R952H, which they argued contributes to the functional deficits. In another study, Stodberg, et al. reported recessive loss-of-function SLC12A5 (the gene for KCC2) mutations in four affected children from two unrelated families. The children had a severe infantile-onset pharmaco-resistant epilepsy syndrome: epilepsy of infancy with migrating focal seizures. In vitro studies of cells transfected with the mutant KCC2 genes showed decreased KCC2 surface expression, reduced protein glycosylation and impaired Cl- extrusion.

All of these findings, and others detailed in the reviews cited above, strongly support the theory that precisely regulated KCC2 activity is required for effective synaptic inhibition in humans and other vertebrates. It seems clear that impairment of KCC2 function, whether through injury, developmental misfortune, or genetic abnormalities, through its influence on GABAergic inhibition, can influence the likelihood that a person will develop an epileptic syndrome. Moreover, as excitation/inhibition balance is centrally involved in onset and termination of critical periods, KCC2 impairment likely plays a dual role in the network abnormalities found in epileptic patients.
Conceptual framework

We now propose a novel conceptual framework, based on a formal model of seizure generation\textsuperscript{223,224} outlining processes by which seizure-related activity prevents the establishment of neurotypical excitatory-inhibitory balance. In real-time, slight variations in a parameter of the formal model can determine the frequency of seizures and interictal events. Such epileptiform activity can then disrupt the developmental timing of large-scale networks and establish new networks defined by and dedicated to epileptogenic brain activity.

Computational Modeling. We begin with a brief survey of computational models of epilepsy, including a description of a recent model upon which we will base our conceptual framework. Epilepsy is complex at multiple levels, as the review above has made clear. Computational models can potentially bridge across levels of organization, from the molecular to the systemic, and also help to establish causality by suggesting experiments that can then test the model\textsuperscript{225}(Figure 5). There are many types of models but we will focus on only a couple here. Progress in computational modeling of epilepsy, as well as methods for doing so, up until 2011, can be found in the edited book by Soltesz and Staley\textsuperscript{226}. The book has chapters describing a number of examples of computational modeling of epilepsy, including a discussion of model scale factors and also multi-causality. We will review a couple of the foundational models here, as well as the most recent model to focus on the dynamics of epilepsy.

Wendling et al.\textsuperscript{227} produced an early model of a hippocampal neuron population with pyramidal neurons, excitatory interneurons, and two classes of inhibitory interneurons: those that project to dendrites of pyramidal and those that project to somas of pyramidal. In the model, all interneurons are excited by pyramids, and the soma-projecting interneurons are inhibited by the dendrite-projecting ones. The time constants of the various feedback loops differ, with the standard somatic inhibition being 10 times faster than the slower dendritic inhibition, and about 5 times faster than the excitatory loop. The synaptic gains of the loops on the pyramidal also varied, with the standard gain of the slow inhibition being about half that of the fast inhibition, but three times that of the excitation. The model was implemented in a system of ordinary differential equations and solved numerically, while varying the gain parameters to produce six different types of EEG-like signals, all of which mimicked signals recorded in epileptogenic brain areas. Importantly, in the model the path from normal brain activity to ictal activity via interictal activity was triggered by two successive reductions in slow dendritic inhibition followed by an increase in slow dendritic inhibition accompanied by a decrease in fast somatic inhibition. Thus, the model predicts that changes in inhibition magnitude are critically important in the genesis of seizures.

In another early modeling paper, Lopes da Silva and colleagues\textsuperscript{228} introduced the idea of state transitions between normal and abnormal (seizure) dynamics, each characterized by an attractor in phase space separated by a separatrix, with the attractors closer together in the epileptic brain (Figure 6). This closeness in the epileptic brain allows the system to transition from the normal attractor to the “seizure attractor” more easily, caused by fluctuations in the
critical parameters or in the initial conditions, something that the same type of fluctuations would seldom cause in the normal brain.

Another important idea in computational models of epilepsy is that of the connectome: the structure of connectivity between various brain regions (e.g., \(^{229}\)). The brain is often construed as a small-world network, one in which there are many short-range connections and a smaller number of longer-range connections. Nethoff, et al.\(^ {230}\) used this idea to create a small world population model for epilepsy. They found that the more long-range connections were present, the more likely a seizure was, and also that the seizure activity was different between CA1 and CA3 neuron models because of the different numbers of recurrent synaptic connections in the two regions (more in CA3).

The model that will inform our conceptual framework was developed by Jirsa and colleagues\(^ {224}\). It is a model of the dynamics of epileptic activity based on a characterization of seizure dynamics from a variety of experimental model sources, including human intracranial EEG. They begin with an immature mouse hippocampus slice model, characterizing seizures by a beginning (onset), sequences of fast discharges and spike and wave events (SWEs), and an end (offset). They found similar patterns in a zebrafish model and in human epileptic iEEG, including a noticeable DC shift at the onset of the seizures, and a logarithmic decline in frequency of SWEs as the seizures ended. Based on these dynamics, they wrote a set of five differential equations that together reproduce sequences of “normal” and epileptic activity closely resembling that seen in their experimental models:

\[
\begin{align*}
\frac{dx_1}{dt} &= y_1 - f_1(x_1, x_2) - z + I_{\text{rest1}} \\
\frac{dy_1}{dt} &= y_0 - 5x_1^2 - y_1 \\
\frac{dz}{dt} &= \frac{1}{\tau_0} (4(x_1 - x_0) - z) \\
\frac{dx_2}{dt} &= -y_2 + x_2 - x_2^3 + I_{\text{rest2}} + 0.002g(x_1) - 0.3(z - 3.5) \\
\frac{dy_2}{dt} &= \frac{1}{\tau_2} (-y_2 + f_2(x_1, x_2))
\end{align*}
\]

In this model, termed the “Epileptor,” the first two equations generate fast oscillations, and the fourth and fifth generate SWEs. The third equation, representing what Jirsa et al. call the “permittivity,” \(z\), operates on a slower time scale than the other four, inducing transitions between the two other regimes. Not shown here is the Gaussian noise that is added into each equation, so that they are not deterministic but rather stochastic. This means that they embody fluctuations as well as deterministic trends.

The sum of \(x_1\) and \(x_2\) variables represents the local field potential in a brain region and reproduces the typical EEG or iEEG trace of fast discharges and SWEs. The variables \(y_1\) and \(y_2\)
interact with $x_1$ and $x_2$ to create oscillations. Jirsa et al. tentatively associate $x_1$ and $y_1$ and the first two equations with glutamatergic activity, and $x_2$ and $y_2$ and the fourth and fifth equations with GABAergic activity, although it’s not that simple, as $x_1$ appears in all of the equations, and $x_2$ appears in the first equation. Nonetheless, they confirmed several predictions from the simple association in their experimental immature mouse hippocampus model, so it is at least plausible as a first approximation. Among these was the prediction that GABAergic activity would be greater during SWEs than during fast discharges, and also that there would be a barrage of GABA activity to the hippocampus just before the onset of seizure-like events. So GABA activity is closely associated with seizures and SWEs both in the computational model and in the experimental model. Similarly, although the biophysical specification of the permittivity is not clear, its slow time course is similar to those of variables like oxygenation, extracellular ion (e.g., K+) concentration, and metabolism during or preceding seizures.

The functions $f_1$, $f_2$, and $g$ are somewhat complicated nonlinear functions that serve to connect the fast discharge system with the SWE system. Notice that the permittivity equation involves only $x_1$, $x_0$, and $t_0$, thus affecting the likelihood of the system to generate fast discharges or SWEs based on what is happening in the fast discharge system as well as what was previously happening in the permittivity. Also note that the time scale of the permittivity, $t_0$, is assumed to be much longer than $t_2 \gg t_1$ ($t_1 = 1$ here). In these equations the constants and initial values were calibrated based on fits to experimental field potential data with $x_1 + x_2$ representing the field potential in the model.

The complexity of the Epileptor, and the number of ways in which the underlying biophysics of the brain can affect the parameters of the model, imply that there are many routes to seizures (Figure 5). Jirsa et al. also computed the “normal” fixed point and “seizure” limit cycle attractors in state space, including also identifying their separatrix. They described how the model system moved through state space as the two interacting subsystems were affected by the permittivity variable, and also discussed the consequences of the system being held close to the separatrix, for example by a particular value of the permittivity, thus increasing the probability that chance fluctuations (neural noise) would force the system across the separatrix from the normal to the seizure attractor. Finally, they emphasized the importance of considering propagation of the fast discharges (and SWEs as well) away from the focal area and into a wider network, thus disrupting neural activity across the brain.

In another important paper, Jirsa and colleagues showed how to use the Epileptor to generate models of the seizure network for individual patients. To define the network, Jirsa et al. defined a spatial map of epileptogenicity where each node is modeled by a set of Epileptor equations coupled to the other nodes via the permittivity equation, and each is characterized by an excitability value, $x_0$, that represents the ability of the Epileptor to trigger a seizure at that node. A value, $x_{0c}$, is the critical value above which the Epileptor can spontaneously trigger or propagate a seizure. Notice that $x_0$ is a parameter in equation 3 of the Epileptor model, so that it influences the permittivity, which in turn influences where the Epileptor is in phase space, i.e., the likelihood of a seizure. The coupling of the models of the separate nodes through equation 3 of the model also influences the permittivity of each node, with the same effect. Each node in the network has its own value of $x_0$. The areas in which $x_0 \gg x_{0c}$ are termed the “epileptogenic zone” (EZ), those where $x_0 > x_{0c}$ but not too much greater, and to which seizures can propagate because of this, belong to the “propagation zone” (PZ), and those where $x_0 < x_{0c}$, so that the
seizure cannot propagate there, are outside the seizure network. Figure 7 depicts one example of such a network, where the epileptogenic zone consists of one hippocampus and the PZ consists of a variety of regions in other task positive networks (TPNs). When the PZ includes regions that are parts of other networks as in Figure 7, which is nearly certain, then the activity in these networks will be disturbed by the seizure, and other interictal events, potentially affecting their functioning, including during development.

Conceptual Framework. As stated in the previous sentence, seizures potentially disturb the functioning of large-scale intrinsic networks even during non-seizure periods. In exploiting this observation we codify it into seven principles based on the literature we have reviewed — and especially on the Jirsa et al. computational model reviewed above.

1) The EZ is characterized by a value of $X_0 >> X_c$, so that seizures can be spontaneously triggered there when the permittivity variable forces the system across the separatrix into the ictal zone. The high value of $X_0$ (an excitability variable) in EZ, and also possibly in PZs, likely arises from malfunctioning of the KCC2 transporter in those areas, resulting in excitatory rather than inhibitory responses to GABAergic input to a subset of pyramidal and interneurons.

1a) The lack of normal inhibitory GABA function in key network nodes might change the sensitive period timing of that area, causing critical circuits to fail to develop normally in response to external input. Lack of KCC2 expression will also affect development of dendritic spines containing AMPA and NMDA receptors, which could also affect sensitive period timing because of fewer excitatory synapses available for network wiring (see Principle 6).

2) The barrage of spikes and oscillations that constitute ictal activity, and the briefer interictal epileptiform spikes and waves, contain no useful information about either the external world or the internal world; they merely signal that the EZ is active. Thus they comprise excessive “noise” in the brain, which is disruptive, rather than facilitative.

3) Ictal and interictal epileptiform activity propagate outward from the EZ to the PZ, thus not only disrupting the operation of the EZ itself, but also the operation of other connected nodes that happen to have $X_0$ values that are greater than $X_c$.

4) The EZ and PZ are both typically nodes in large-scale brain networks, and thus the epileptiform activity disrupts activity in networks during the seizure, depending on where the EZ is located.

5) The normal course of development of large-scale brain networks primarily involves an overall pattern of increased dissociation between functionally separate networks and increased integration within each network.

6) Because neurons that “fire together wire together” (Hebbian principle), epileptiform activity will cause “meaningless” wiring changes within the EZ and PZs, and also between them, potentially connecting nodes that should be dissociating and dissociating nodes that should be connecting (Figure 6). This will be especially likely during sensitive periods of brain development, when synapses are being formed and dissolved at a high rate.

6a) In an alternative scenario, lack of normal GABA inhibition and dendritic spine formation resulting from KCC2 abnormalities causes a delayed sensitive period within a given node, so that critical circuits between it and others in a network either develop differently or do
not develop at all, perhaps being replaced by connections generated by ictal activity.

(7) The meaningless and wrongly directed connectivity within and between large-scale brain networks will impair their functioning during tasks, even when ictal activity is absent, leading to observed perceptual and cognitive deficits.

To make this framework more concrete, consider a typical example of a sclerosed hippocampus (Figure 7). This area will constitute the EZ, as has been amply demonstrated in the literature. The sclerosis might have been caused by a tumor, or by infantile febrile seizures. In and around the sclerosed region KCC2 transporters will be malfunctioning, leading to highly excitable tissue, easily triggered to a seizure. When a seizure, or even a brief epileptiform event, occurs, meaningless brain activity will radiate out along structural connections to other brain areas, and the ones with values of $X_0 > X_c$ will also seize, generating their own ictal activity. Not only will this activity disrupt other meaningful activity, such as learning language, or remembering facts or spatial arrangements, but it will also cause wiring changes (synaptic connectivity) that are unrelated to the current situation, and thus meaningless to present or future tasks. For example, the hippocampus is part of the medial temporal lobe component of the DMN, which includes: 1) ventral-medial prefrontal cortex, a region that also plays an important role in context sensitive valuation of objects or actions and regulation of subcortical responses to emotionally salient events; 2) posterior midline regions associated with recall of autobiographical memory detail, and 3) the parahippocampus, a region of the visual cortex sensitive to large-scale spatial scenes. The hippocampus is also a key node in networks involved in sequencing and encoding sensory information in space and time and in episodic memory retrieval. Moreover, it participates with the amygdala in laying down and retrieving emotionally fraught memories. Links between the hippocampus and the dorsal anterior cingulate cortex play a role in generalized fear memory and networks that include anterior cingulate regions and the hippocampus are nodes of a network altered in major depression. Finally, the hippocampus has been implicated in forms of semantic recall. Developmental disruption of segregation and integration by hippocampal sclerosis of networks implicated in mediating social-emotional valuation, emotional self-regulation, and memory may be an important factor in the extremely high comorbidity of epilepsy and depression, and in challenges to social-emotional cognition observed in epilepsy. For example, due to sub-optimal integration and segregation of networks subserving encoding and remembering emotional salient events, children may not learn to attend to the facial features that signify specific emotional expressions and the consequences associated with encountering them. This in turn may result in impaired emotional expression recognition often observed in epilepsy. A similar process may underlie language deficits. Imagine a child trying to think of the word for an object to tell their mom about it. Typically, the perceptual information about the object would interact with the hippocampus and left-hemisphere association areas to retrieve the desired word. But, if the hippocampal memory circuits are not properly connected to the incoming sensory/perceptual circuits, the desired word will not be forthcoming, possibly leading to the language production deficits seen in epileptic children.

Conclusion.

Temporal lobe epilepsy is associated with a number of negative outcomes linked to challenges in communication ability, and the earlier the onset the greater the challenges. But the
processes by which TLE seizures disrupt the developing brain have been under examined. In this paper, we tackled the question of how and why age matters by synthesizing literature ranging in scale from molecular processes to whole-brain networks. We first reviewed literature on healthy brain network development and sensitive periods in regional brain development, drawing on studies conducted using a range of methods in humans and non-human animals. We then reviewed what is known about how network development is altered by epileptic seizures and Interictal activity. We reviewed evidence that, overall, brain network activity becomes more articulated and efficient through childhood and adolescence, but the sculpting of connections within and between networks can be disrupted in favor of connections shaped by epileptic activity. We further reviewed patterns of GABA-ergic inhibition implicated in windows of plasticity associated with temporally circumscribed sensitive periods, and described some ways in which processes involved in sensitive period plasticity may interact with seizure activity to alter developmental trajectories. At the molecular level, we described how altered KCC2 function may influence brain network development both during sensitive windows and over more sustained developmental periods. Finally, we drew on a computational model of seizure activity, the Epileptor model, as the basis of an original theoretical framework outlining key ways in which age of onset matters to the course of brain development. Here we proposed that high excitability of the epileptogenic zone, stemming from malfunction of the KCC2 transporter, results in excitatory rather than inhibitory response, rendering the region susceptible to seizures. Over time, ictal and interictal activity add excessive “noise” to the process of network development, and lack of normal GABA inhibition also may cause a mistimed sensitive period within a given node, disrupting typical network development. Mis-wiring within and between large-scale brain networks will impair their functioning during tasks, even when ictal activity is absent, leading to perceptual and cognitive deficits. This framework provides an explanatory basis for why, in epilepsy, age of onset matters so much, with earlier onset exacerbating challenges in linguistic and social-emotional communication through multiple routes.

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Figures

Figure 1. FMRI BOLD language (warm colors) and mental rotation (cool colors) task-based functional connectivity networks in two children with epilepsy. Activations shown are from multiple barycentric discriminant analysis employed to find network activation patterns that best discriminate the tasks (Abdi et al., 2012). This figure illustrates the bootstrap ratios (akin to Z-scores) for the first dimension, which represents the separation between the language network and the mental rotation networks. The network pattern in (a) is from a child who suffers more than 12 seizures per year. The network pattern in (b) is from a child who suffers more than 52 seizures per year, and who also has a mild generalized learning disability. The task-based language network for the child with greater seizure frequency shows greater connectivity (the deeper orange and red colors) and connectivity to more regions than the child with fewer
seizures. The task-based mental rotation network also shows greater connectivity to more regions for the child with greater seizure frequency.
Figure 2. Theoretical alterations to critical period plasticity. A) A schematic showing the rise and fall of plasticity across different brain regions in a series of cascading critical periods. Critical period timing is staggered across development, and controlled by the maturation of key inhibitory circuits in these different systems. The color code reflects brain regions of interest, showing the relative ordering of critical periods across these different systems, however the timing is not meant to be exact. Disruption of inhibitory circuits may lead to either an alteration in the level of plasticity (B) or shift the timing with which the critical period window for a given domain occurs during development (C).
Figure 3. NKCC1 and KCC2 actions from Moore et al. (2017) with permission. In the immature neuron the NKCC1 transporter is more active, resulting in a high intracellular concentration of Cl- ions, and resulting in the transport of Cl- out of the cell when the GABA receptor is activated. In the mature neuron the KCC2 transporter is more active, decreasing the intracellular concentration of Cl- ions to a level where activation of the GABA receptor will result in an influx of Cl- ions and inhibition.
Figure 4. KCC2 and phosphorylation sites; from Moore et al. (2017) with permission. Notice that the KCC2 protein is interwoven into the cell membrane, and that there are several intracellular sites where the addition of a phosphoryl group results in a change in the transport of the Cl⁻ ion. Particularly important is the S940 site, where phosphorylation results in upregulation of Cl⁻ ion transport out of the neuron by increasing the membrane stability of KCC2.

Figure 5. The river of epilepsy, exemplifying its multi-level, multi-causal nature. Computational models can include any or all of these factors. Modified (Cl⁻ NKCC1-KCC2 channels added) from Lytton (2008) with permission.
Figure 6. Normal and seizure attractors and their separatrices in normal and epileptic brains. Notice that the trajectories in phase space in the normal brain are farther from the separatrix than are those in the epileptic brain, so that small fluctuations are more likely to cause transitions from normal to ictal activity in the epileptic brain. Based upon Lopes da Silva et al. (2003).
Figure 7. An epileptic network, with an epileptogenic zone (EZ) (red), where seizures can be initiated, propagation zones (PZ) (blue), to which the seizures can spread but not be initiated, and other nodes (white) whose excitability is low enough that the seizure doesn’t propagate into them. See Jirsa et al. (2017). Regarding the proposed framework, development of abnormal connectivity between red and blue nodes, the epileptic network, could disrupt functioning of the TPNs during non-seizure periods. A=amygdala, EC=entorhinal cortex, H=hippocampus.