Published in final edited form as:
*Nat Rev Neurosci*. 2013 May ; 14(5): 337–349. doi:10.1038/nrn3482.

# Mechanisms of epileptogenesis: a convergence on neural circuit dysfunction

**Ethan M. Goldberg** and **Douglas A. Coulter**
The Children's Hospital of Philadelphia, Division of Neurology, Colket Translational Research Building, 3501 Civic Center Boulevard, Philadelphia, Pennsylvania 19104–4399, USA

## Abstract

Epilepsy is a prevalent neurological disorder associated with significant morbidity and mortality, but the only available drug therapies target its symptoms rather than the underlying cause. The process that links brain injury or other predisposing factors to the subsequent emergence of epilepsy is termed epileptogenesis. Substantial research has focused on elucidating the mechanisms of epileptogenesis so as to identify more specific targets for intervention, with the hope of preventing epilepsy before seizures emerge. Recent work has yielded important conceptual advances in this field. We suggest that such insights into the mechanisms of epileptogenesis converge at the level of cortical circuit dysfunction.

Epilepsy (BOX 1) is a common end point of many forms of acquired brain pathology (such as tumours, infection, stroke and traumatic brain injury). Epilepsy can also be the result of the mutation of a single gene (genetic epilepsy), and it can be one component of a neurodevelopmental disorder. The most common epilepsy syndrome in adults — mesial temporal lobe epilepsy (TLE; see BOX 2) — is typically considered to be an acquired disorder, resulting from injury to a previously normal brain. However, there are also familial (presumed genetic) forms of TLE. Although usually applied to the acquired epilepsies, the concept of epileptogenesis might also be relevant to the genetic epilepsies. An anti-epileptogenic intervention could be used in the context of a broader neurodevelopmental abnormality that includes epilepsy if an underlying defect could be identified and modulated before seizures began. Conversely, there could be potentially modifiable genetic contributions to what are classically considered to be acquired epilepsies, including TLE[1–4]. For example, an individual might have a genetic predisposition to risk factors for epilepsy, such as status epilepticus or hippocampal sclerosis, which in turn drive epileptogenesis.

**Box 1**

**Definitions: from seizure to epilepsy**

**Seizures**

Seizures are abnormal, paroxysmal changes in the electrical activity of the brain. Seizures emerge from and are an inherent property of cerebral cortical structures. However, a



Correspondence to: D.A.C. coulterd@mail.med.upenn.edu.

**Competing interests statement**
The authors declare no competing financial interests.

**FURTHER INFORMATION**
**Douglas A. Coulter's homepage:** http://stokes.chop.edu/web/coulter/ The Children's Hospital of Philadelphia Division of Neurology: http://www.chop.edu/service/neurology/home.html **ALL LINKS ARE ACTIVE IN THE ONLINE PDF**

(single) seizure can occur in an otherwise healthy individual; such individuals are generally not considered to have epilepsy.

**Epilepsy**

Epilepsy, by contrast, is a neurological disorder defined as a brain state that supports recurrent, unprovoked seizures.

**Epileptogenesis**

Epileptogenesis is the process by which the previously normal brain is functionally altered and biased towards the generation of the abnormal electrical activity that subserves chronic seizures. The concept of a 'mechanism of epilepsy' refers to any biological feature of the brain that drives or supports recurrent, unprovoked seizures. Examples of these biological features include molecular, anatomical or circuit level alterations, such as cell death or dysregulation of an ion channel or neurotransmitter receptor. By this definition, epilepsy mechanisms may be the downstream effectors of, but are distinct from, a prior epileptogenic process.

The process of epileptogenesis is classically thought to occur in three phases: first, the occurrence of a precipitating injury or event; second, a 'latent' period during which changes set in motion by the preceding injury act to transform the previously normal brain into an epileptic brain; and third, chronic, established epilepsy. It is during the latent period that the process of acquired epileptogenesis is thought to coalesce, and it is at this point in the process that interventions might be used to prevent the subsequent development of epilepsy.

The distinction between seizures, epilepsy and epileptogenesis is well illustrated by the accumulating data indicating that classical anticonvulsants, which can terminate an ongoing seizure and decrease or prevent the occurrence of future seizures in epileptic individuals, are wholly ineffective in preventing the process of epileptogenesis[169].

## Box 2

### Temporal lobe epilepsy

Mesial temporal lobe epilepsy (TLE; also known as limbic epilepsy), refers to an electroclinical syndrome in which seizures emanate from the temporal lobe. TLE is a prominent and prevalent epilepsy syndrome in children, adolescents and adults. This syndrome is often associated with mesial temporal sclerosis — a pattern of hippocampal damage, including atrophy and gliosis, seen in humans with TLE and in animal models of chronic TLE. TLE is typically considered to be an acquired epilepsy syndrome that is triggered by a precipitating factor, such as head trauma, infection or tumour, or an episode of prolonged, uncontrolled seizure (status epilepticus), although there are known familial forms of mesial TLE[1–4,170,171]. Lateral TLE[172] is a distinct syndrome that will not be discussed in this Review. From a basic science perspective, TLE has generally been considered as an acquired condition; likewise, temporal lobe epileptogenesis has been considered relevant to the acquired epilepsies only.

Animal models of acquired TLE have been a particularly useful tool in the study of epileptogenesis, because the animal model resembles the human condition in many key respects — chronic TLE can be induced in rodents using various types of brain injury that also cause TLE in humans. Previous studies have identified changes in molecular signalling pathways in animal models of acquired TLE, including alterations in immediate-early gene expression and the expression of various ion channels and neurotransmitter receptors. Proposed circuit-level mechanisms include synaptic

reorganization, axonal sprouting and the generation of new, pathological, recurrent excitatory synaptic connections (for example, mossy fibre sprouting). Cellular-level mechanisms include inflammation, neuronal cell death (including the specific loss of mossy cells and vulnerable populations of inhibitory GABAergic interneurons), reactive astrocytosis and dispersion of existing and the generation of new dentate granule cells in ectopic locations. Some of these changes are likely to be involved in and perhaps required for the pathogenesis of epilepsy, whereas others might be reparative of or compensatory for the initial injury and ongoing chronic epileptic condition, and still other changes might simply be epiphenomenal.

In this Review, rather than attempting to provide a broad overview of epileptogenesis, we restrict our discussion to a set of emerging themes. We review the role of specific large-scale molecular cell signalling pathways in epileptogenesis, highlighting the mammalian target of rapamycin (mTOR) and repressor element 1 (RE1)-silencing transcription factor (REST; also known as neuron-restrictive silencer factor (NRSF)) pathways in acquired and genetic epilepsies. We discuss how dysfunction of specific subtypes of neurons might lead to epilepsy, with a focus on the theory of cortical GABAergic interneuron dysfunction as the basis of the infantile-onset epileptic encephalopathy of Dravet syndrome. Finally, we consider the pathology of discrete elements of neuronal circuits in mesial TLE. For a broader overview, see recent general reviews of the mechanisms of acquired epileptogenesis[5,6].

Seizures are network events that require re-entrant activation of populations of neurons embedded within brain circuits; hence, epilepsy is inextricably a circuit-level phenomenon and cannot be understood outside this context. It is likely that multiple and diverse mechanisms of epileptogenesis overlap to various extents among different epilepsy syndromes, both acquired and genetic. However, for a discrete genetic mutation or dysfunction of a molecular signalling pathway or specific cell type to produce epilepsy, it must do so by altering the function of circuits. We suggest that a greater mechanistic understanding of circuit function and circuit-level dysfunction in epilepsy will lead to the development of successful and broadly applicable interventions in epileptogenic processes, which remain a primary unmet need in epilepsy therapy.

## Molecular signalling pathways in epileptogenesis

The possibility that a brain injury or genetic abnormality might lead to the development of epilepsy through maladaptive activity of one or more large-scale molecular signalling pathways is inherently attractive, as this would suggest that directed manipulation of these pathways might be able to modify the epileptogenic process before the appearance of seizures. Research is motivated by an attempt to identify molecular signalling processes that might be involved in driving the formation of the aberrant neuronal circuitry seen in the epileptic brain, including features such as axonal sprouting and cell death that develop following a delay after an initial brain insult. Important potential master regulators of epileptogenesis have emerged in recent years, including brain-derived neurotrophic factor (BDNF)–tropomyosin-related kinase B (TRKB; also known as NTRK2) signalling[7–9], the mTOR pathway[10–12], the REST pathway[13] and other transcriptional regulators[14]. Here, we focus on the roles of mTOR and REST given the recent and rapid accumulation of evidence implicating these pathways in temporal lobe epileptogenesis.

### Mammalian target of rapamycin

mTOR is a serine/ threonine protein kinase that is involved in multiple basic cellular functions, including cell growth, proliferation and survival. mTOR is regulated by pathways

that respond to glutamatergic transmission and the energy state of the cell (FIG. 1). In turn, mTOR regulates synaptic plasticity[15], neuronal structure[16] and the expression of various ion channels and neurotransmitter receptors[17]. The mTOR pathway is strongly associated with epilepsy. Genetic mutations in many components of the mTOR pathway, including tuberous sclerosis complex 1 (*TSC1*) and *TSC2,* STE20-related kinase adaptor-α (*STRADA*), phosphatase and tensin homologue (*PTEN*), the gene encoding the serine/threonine kinase AKT isoform that is predominant in the brain (*AKT3*), phosphatidylinosi-tol-4,5-bisphosphate 3-kinase-α (*PIK3CA*) and *MTOR* itself, produce syndromes that include epilepsy[10,11,18–20] (FIG. 1). In addition, manipulation of the mTOR pathway can modulate acquired epilepsy in animal models.

The role of the mTOR pathway in genetically determined epilepsy[11,18] is typified by tuberous sclerosis, which results from mutations in *TSC1* and *TSC2* (FIG. 1). The mechanisms of seizure generation in tuberous sclerosis are debated[21], but seizures are thought to originate around or within cortical tubers[22], which are foci of abnormal neuronal migration and cellular growth. Intracranial electroencephalographic monitoring in patients with tuberous sclerosis has provided evidence that at least some tubers are intrinsically epileptogenic and that they might in some cases be more epileptogenic than the perituberal cortex[23]. *Tsc1*$^{GFAP}$ conditional knockout mice, in which *Tsc1* is deleted from glia, show astrogliosis and dislamination, with pyramidal cell dispersion in the hippocampus[24]. Glia in these mice show altered expression of gap junction proteins, potassium channels and astrocytic glutamate transporters[25]. Treatment of these mice with the mTOR inhibitor rapamycin suppresses seizures; however, seizures return if rapamycin is withdrawn[26].

Interestingly, selective deletion of the mTOR pathway gene *Pten* from a small subset of dentate granule cells in mice produces TLE[27]. The mechanism by which deletion of *Pten* in a subset of dentate granule cells might lead to epilepsy is unclear, but it caused cellular-level structural abnormalities including increased spine density and granule cell hypertrophy, and this abnormal cell growth might itself be epileptogenic. Mossy fibre sprouting in these mice was the same from wild-type granule cells as from those in which *Pten* had been deleted, which suggests that mossy fibre sprouting might be a consequence rather than a cause of epilepsy, at least in this mouse model.

Somatic mosaicism for gain-of-function mutations in the mTOR pathway components *AKT3, PIK3CA* and *MTOR* are responsible for some cases of the dramatic focal cortical brain malformation that occurs in hemimegalencephaly[28,29]. This condition is almost invariably associated with medically intractable epilepsy. Mice harbouring an activating mutation in *Akt3* have epilepsy and increased brain size[30].

The mTOR pathway might also be involved in acquired epilepsy. This has been investigated using rodent models of chronic acquired TLE after status epilepticus induced by the chemoconvulsants kainate[31] or pilocarpine[32–34] (kainate- or pilocarpine-induced status epilepticus), TLE induced by electrical stimulation of the angular bundle[35] or the amygdala[36], post-traumatic epilepsy after brain injury[37,38] and models of infantile spasms[39] and early hypoxic-ischaemic injury. Rapamycin treatment before kainate-induced status epilepticus prevented the development of spontaneous recurrent seizures in a subset of rats[31]; in those rats that did develop epilepsy, rapamycin increased the latency to seizure development and decreased the frequency of spontaneous seizures. When applied after the induction of status epilepticus, rapamycin decreased cell death, mossy fibre sprouting and aberrant dentate granule cell neurogenesis, all of which are histopathological features associated with TLE. However, in mice, seizure frequency was unchanged by rapamycin treatment given after pilocarpine-induced status epilepticus, although the degree of mossy fibre sprouting was decreased[34].

Mossy fibre sprouting was inhibited by rapamycin given before or after status epilepticus in the kainate- or pilocarpine-induced model of TLE in rats[31–34] and after status epilepticus induced by electrical stimulation of the angular bundle[35] but not in a model of TLE induced by electrical stimulation of the amygdala[36]. However, upon treatment withdrawal, the degree of sprouting returned to the levels seen in untreated epileptic animals[32], and seizures recurred after treatment removal[33]. Hence, whereas manipulation of mTOR signalling in most rodent models of acquired epilepsy appears to modulate seizure frequency and decrease the degree of various measures of TLE neuropathology, such as mossy fibre sprouting and hilar cell loss, mTOR pathway modulation in such models has not been shown to prevent the development of epilepsy permanently. It seems that continued rapamycin administration is required to maintain any anti-epileptogenic effects. Some authors have referred to the effects of rapamycin as 'antiseizure' or 'epileptostatic', given that rapamycin has not been shown to enact a prevention of epilepsy development that persists after rapamycin withdrawal[40,41]. Despite these exciting recent developments and the clear role of mTOR pathway molecules in genetic epilepsy syndromes, the mechanism (or mechanisms) by which manipulation of mTOR signalling after brain injury (such as that due to status epilepticus) might modulate epileptogenesis remain unclear. These mechanisms might include inhibition of mossy fibre sprouting, neuroprotection and maintenance of the integrity of the blood–brain barrier[35].

### Repressor element 1-silencing transcription factor

REST is a transcription factor that binds to DNA-binding motifs called neuron-restrictive silencer elements (NRSEs; also known as RE1 elements). REST negatively regulates the expression of many neuronal genes in non-neuronal cells and neuronal precursor cells. It also regulates neuronal gene expression in mature neurons[42,43]. REST binds to various co-repressors, which recruit his-tone deacetylase 1 (HDAC1) and HDAC2 (FIG. 1); these deacetylases regulate chromatin structure and repress the expression of hundreds of neuronal genes[44]. Almost 2,000 genes have REST-binding motifs[44] (although this number could be much greater[45]), including ~10% of all neuronally expressed genes[46], some of which encode proteins that are key regulators of neuronal excitability and have been independently proposed to be involved in epilepsy mechanisms[47]. Such genes include type A GABA ($GABA_A$) receptor β3 subunit (*GABRB3*), $GABA_A$ receptor δ-subunit (*GABRD*) and the ionotropic AMPA2 glutamate receptor (*GRIA2*), *BDNF* and genes encoding hyperpolarization-activated cyclic nucleotide-gated (HCN) channel subunits 1–4 (*HCN1–HCN4*).

More direct evidence that alterations in REST signalling might be involved in epileptogenesis continues to accumulate[48]. Status epilepticus in animal models increases the expression of REST in the hippocampus and alters the histone acetylation of prominent neuronally expressed genes[42,49]. Status epilepticus induces promoter-specific acetylation of *BDNF* that is inhibited by HDAC inhibitors[49,50]. In the kindling model, inhibition of glycolysis induces the binding of REST to the BDNF promoter, which prevents BDNF upregulation and modulates epileptogenesis, providing an interesting mechanistic link between glycolysis, BDNF–TRKB signalling (see above) and the REST system[51]. The ultimate effector (or effectors) of this signalling cascade remain unknown.

The role of HCN channels in epileptogenesis has been reviewed in REF. 52. Dendritic HCN channels in hippocampal pyramidal cells are dysregulated after prolonged febrile seizures or after kainate- or pilocarpine-induced status epilepticus in rodents, leading to what has been termed an 'acquired HCN channelopathy' (to differentiate this from a genetic channelopathy)[53–55]. This phenomenon might be partly due to REST-mediated transcriptional repression of *Hcn1*[56]. Binding of REST to *Hcn1* is increased after the

induction of status epilepticus in a rodent model of TLE, resulting in decreased HCN1-mediated ionic currents ($I_h$) and altered function of CA1 pyramidal cells. Interference with REST binding to target genes (including *Hcn1*) using an anti-REST decoy oligodeoxynucleotide was associated with decreased seizure frequency in rats after kainate-induced status epilepticus. These examples show that status epilepticus might activate the REST pathway to suppress important regulators of neuronal excitability that have been independently proposed to be involved in pathogenic epilepsy mechanisms. It will be important to determine which gene targets of REST are up- or downregulated by this pathway after status epilepticus or other brain injury. This will prove challenging given the broad activity of this pathway across the genome. If a certain REST target was identified as particularly important in driving epileptogenesis, would it be possible to selectively interrupt status epilepticus-induced, REST-mediated suppression of specific targets? Can the REST system be safely modulated in humans after brain injury (for example, with HDAC inhibitors)? Although long-term treatment with such agents might be accompanied by unacceptable toxicity, perhaps a single treatment or a short course of treatment given immediately after injury would prove safe.

The mTOR and REST pathways are exciting new potential targets for intervention in the epileptogenic process, but specific questions remain. To what extent is activation of the mTOR or REST pathways involved in driving epileptogenesis rather than being the result of this process? What are the circuit-level effects of the manipulation of key molecular signalling cascades such as the mTOR pathway and REST after status epilepticus or other brain injury? What are the relevant molecular effectors of mTOR pathway blockade or HDAC inhibition in the prevention of epileptogenesis in animal models of acquired TLE? And how widespread is the relevance of these signalling cascades across various forms of epilepsy beyond TLE?

Extensive research in animal models and in humans after brain injury using a range of prophylactic anticonvulsants, anti-inflammatory agents and neuroprotective agents has failed to demonstrate anti-epileptogenic effects[5,6]. What hope is there that targeting the mTOR and REST pathways will meet with greater success? As opposed to classical anticonvulsants, for example, which act on one or a few discrete targets, modulation of mTOR or REST would have broad action. As there are likely to be multiple divergent mechanisms of epilepsy development, perhaps polytherapy cocktails that target multiple, parallel pathways will prove effective. Using epileptogenic circuit-level dysfunction (as discussed below) will allow us to place in context the downstream effects of mTOR and/or REST system blockade on epilepsy development and facilitate translational efforts towards therapy development.

## Cell type-specific dysfunction and epilepsy

Previous work studying the basic mechanisms of limbic epilepsy has identified dysfunction at the cellular level that might suggest potential targeted anti-epileptogenic interventions. Here, we discuss specific subtypes of cortical inhibitory GABAergic interneurons for which a mechanistic role in epilepsy is increasingly being recognized. A larger discussion of cortical interneuron subtypes is beyond the scope of this Review and has been addressed recently[57–60]. More general roles of GABAergic inhibition in epilepsy mechanisms have been reviewed in REFS 61–64, including the emerging potential use of GABAergic interneuron transplantation as a therapeutic treatment modality for epilepsy[65–67].

### Somatostatin-positive hilar interneurons

Immuno-reactivity for the neuropeptide somatostatin defines various overlapping subtypes of GABAergic interneurons in the cerebral cortex. In the dentate gyrus, most GABAergic interneuron somata that are immunopositive for somatostatin are located in the hilus[68];

approximately 50% of all hilar GABAergic interneurons are somatostatin-positive[69]. The synaptic terminal fields of somatostatin-positive hilar interneurons are most prominent in the outer molecular layer of the dentate gyrus, where the fibres of the perforant path terminate[70,71]; in addition, the dendrites of these cells are targeted by recurrent collaterals of the granule cell-derived mossy fibres[72]. In turn, somatostatin-positive terminals ramify primarily with granule cell dendrites. Thus, hilar somatostatin interneurons are ideally positioned to modulate the influence of the perforant path on dentate granule cells (FIG. 2).

Selective loss of somatostatin-positive GABAergic hilar interneurons is a well-documented and consistent finding in animal models of TLE[73,74] and is found in the sclerotic hippocampus from humans with TLE[75], indicating that these neurons are highly and selectively vulnerable to excitotoxic and ischaemic damage. In the kainate model of chronic TLE in mice, surviving somatostatin-positive hilar interneurons show axonal sprouting to form new synapses with dentate granule cells[76], which is consistent with increased somatostatin immunoreactivity in the dentate molecular layer in human TLE[77]. This might be a way to restore the compromised levels of inhibition that are produced in part by the loss of inhibitory interneurons. Interestingly, axonal sprouting by somatostatin-positive hilar interneurons is inhibited by rapamycin[78]. The consequences of the loss of somatostatin-positive hilar interneurons and the changes that surviving neurons undergo in the context of hippocampal circuit function are subjects of ongoing investigation.

## Fast-spiking cells and epilepsy

Parvalbumin-positive fast-spiking basket cells are key regulators of inhibition in the cerebral cortex. Fast-spiking cells are the most numerous subtype of cortical interneuron[79,80]. These cells form powerful, proximally localized synapses on the soma and axon initial segment (AIS) of postsynaptic cells, exerting fine control over the timing and probability of target cell discharge. Fast-spiking cells mediate feedforward and feedback inhibition that is recruited by specific spatio-temporal patterns of excitatory activity[81]. In addition, fast-spiking cells form dense interconnected networks through specific gap junctional coupling, which contributes to the function of these cells in the generation of fast oscillations within cortical structures. Modelling studies predict that perisomatically targeted inhibitory cell activity produces gamma oscillations[82]. Optogenetic activation of parvalbumin-positive fast-spiking cells augments gamma-band oscillatory activity in the neocortex[83,84]. In turn, dysfunction of interneuron-mediated gamma-band synchronization has been proposed to contribute to TLE mechanisms[85]. Specific optogenetic activation of parvalbumin-positive interneurons in the hippocampus terminated seizures in a mouse model of TLE[86].

Genetic manipulation of interneuron progenitors in the medial ganglionic eminence (MGE) that are destined to become parvalbumin-positive fast-spiking cells causes epilepsy in mice[87–89]. In addition, genetic mutation or experimental manipulation of various ion channel genes that are prominently or exclusively expressed in fast-spiking cells (including the voltage-gated potassium channel genes *KCNA1* (REFS 90–93) and *KCNC2* (REF. 93)) causes epilepsy in humans and in mouse models.

Mutations of *SCN1A* (which encodes the voltage-gated sodium channel subunit $Na_V1.1$) causes a spectrum of related epilepsy syndromes, including generalized epilepsy with febrile seizures plus (GEFS+) and Dravet syndrome[94–96]. Initial functional studies of *SCN1A* mutations associated with GEFS+ yielded inconsistent results; however, similar studies of the more severe Dravet syndrome phenotype showed a loss of function caused by haploinsufficiency[97]. The loss of function caused by haploinsufficiency was explained by evidence showing that mutation of *SCN1A* generates epilepsy by a selective reduction in the sodium current in cortical inhibitory GABAergic interneurons relative to pyramidal cells, which leads to decreased action potential firing frequency in interneurons[98–103]. This was a

very attractive hypothesis as to the mechanism of Dravet syndrome, given the important role of fast-spiking cells in the regulation of cortical inhibition. The sodium current might be particularly reduced in cerebral cortical parvalbumin-positive fast-spiking interneurons in which $Na_V1.1$ appears to be prominently and perhaps differentially expressed relative to pyramidal cells and parvalbumin-negative interneurons[104]. Further supporting this theory, interneuron-specific deletion of *Scn1a* in the forebrain using *Dlx1/2* Cre transgenic mice causes an epilepsy phenotype that is similar to global heterozygous deletion of *Scn1a*[105]. In addition, pathology in these mice begins to appear at the same time that fast-spiking interneurons acquire a mature electrophysiological phenotype[106,107], which might be the developmental time point at which cortical structures begin to rely on fast-spiking cells as a major source of inhibition. However, a published voltage-clamp recording of sodium currents from interneurons and principal cells in mouse models of Dravet syndrome exists only for dissociated hippocampal neurons in culture[98] and neurons from the visual cortex[104].

Previous data showed that $Na_V1.1$ is expressed at the AIS, soma and dendrites of pyramidal cells[108,109], but more recent findings in adult rats suggest that $Na_V1.1$ expression in the neocortex and hippocampus is restricted to the AIS of parvalbumin-positive interneurons[110]. This unresolved issue complicates the interpretation of the proposed cell type-specific dysfunction in mouse models of Dravet syndrome. Such dysfunction might reflect true cell type-specific differences in $Na_V1.1$ expression or differential compensation by other sodium channel genes in pyramidal cells as compared with interneurons. This compensation might occur differently in the various mouse models of Dravet syndrome (such as *Scn1a* knockout mice, knock-in mice harbouring a nonsense mutation in *Scn1a* or bacterial artificial chromosome (BAC) transgenic mice expressing a mutation from a human patient with Dravet syndrome), although haploinsufficiency seems to be a common theme. Genetic background strain and developmental effects might also affect the phenotype of mouse models of Dravet syndrome. That interneuron-specific *Scn1a* knockout recapitulates the phenotype of the global null mutation shows that *Scn1a* deletion in cortical interneurons is sufficient to produce a Dravet-like phenotype in mice; however, this does not prove that forebrain interneurons are the necessary and exclusive locus of cellular dysfunction in Dravet syndrome or that the absence of $Na_V1.1$ in fast-spiking cells is the mechanism of Dravet syndrome. As mentioned above, many perturbations directed towards fast-spiking cells produce severe epilepsy, at least in mouse models, so it is not surprising that deletion of a major sodium channel that underlies action potential generation in these cells will cause hyperexcitability in cortical circuits with resultant epilepsy. Furthermore, although the *SCN1A* mutation is the most common cause of Dravet syndrome, accounting for approximately 70% of cases[95,96], mutations in other genes can cause a Dravet or Dravet-like phenotype. These genes include *GABRG2* (REF. 111), the gene encoding sodium channel-β1 subunit (*SCN1B*)[112,113], the gene encoding $Na_V1.2$ (*SCN2A*)[114,115] and protocadherin 19 (*PCDH19*)[116], suggesting that multiple genetic mechanisms can produce Dravet or Dravet-like syndrome. Whether these genetic mutations converge on fast-spiking cell dysfunction is unknown.

Dravet syndrome spectrum disorders might represent a point of convergence between acquired and genetic mechanisms of epilepsy. Mutations of *SCN1A* (or other Dravet syndrome spectrum genes) generate a strong genetic predisposition to febrile and afebrile seizures in humans. Patients with Dravet syndrome are not born with epilepsy, just as mice with *Scn1a* (or *Scn1b*[117]) mutations do not exhibit spontaneous seizures until the late second or early third postnatal week. Like patients with Dravet syndrome spectrum disease, mouse models initially show temperature-sensitive seizures before the emergence of spontaneous seizures[98,100,104]. As described above, prolonged febrile and afebrile seizures (particularly status epilepticus) can trigger a secondary cascade of events that might compound the pathology, leading to neuronal plasticity and epileptogenesis[118,119]. Interestingly, focal TLE

has been reported in patients with the *SCN1A* mutation and GEFS+[120,121]. TLE has also been reported in a small group of patients with GEFS+ that is due to the *SCN1B* mutation, including one patient without known history of prolonged febrile seizure or status epilepticus who had TLE with mesial temporal sclerosis (BOX 2) and who underwent successful temporal lobectomy[122]. Of further interest, transgenic mice expressing a gain-of-function *Scn2a* mutation have epilepsy and mesial temporal sclerosis[123]. Although MRI scans of patients with Dravet syndrome are typically normal initially, hippocampal sclerosis develops over time in some patients[124–126]. This concept of epileptogenesis, linking sodium channel mutations, febrile seizures, status epilepticus and TLE, blurs the distinction between the classical mechanisms of epilepsy that are driven either by acquired brain injury or genetic mutation. Hence, it might be possible to intervene medically before the emergence of seizures in at-risk patients, in whom risk is defined by vulnerable genetic background in addition to exposure to brain injury. Possible interventions might include dietary therapies (for example, the ketogenic diet[127]), stem cell-based therapies to replace a dysfunctional cell type or targeted pharmacological or optogenetic agents tailored to a specific neuronal cell type.

The mechanisms by which the *SCN1A* mutation leads to epilepsy are highly controversial and are the subject of intense ongoing investigation. Recent data on *Scn1b* null mice might further complicate the picture. These mice show abnormal neuronal proliferation, neuronal migration and axonal targeting at early developmental time points, before the appearance of seizures[128], suggesting that neurodevelopmental abnormalities might contribute to epilepsy in these mice. As *Scn1b* regulates the physiology, surface expression and subcellular targeting of *Scn1a*, perhaps Dravet syndrome represents a neurodevelopmental disorder of which epilepsy is but one prominent component. That said, the delayed appearance of epilepsy does not necessarily imply an underlying epileptogenic process; it could be that the circuit elements that underlie seizure generation are simply not mature until a particular age (see above). This is clear from the fact that seizures appear days, months or years after birth in Dravet syndrome and in other genetic epilepsies. We suggest that a more complete understanding of these issues will require further investigation of the roles of fast-spiking cells in the normal function of cortical circuits and the mechanisms and consequences of fast-spiking cell dysfunction in circuit operations in *Scn1a* mutant mice.

## Circuit dysfunction in epileptogenesis

Seizures are network-level phenomena, and the incomplete knowledge of neuronal circuit function has left a gap in our understanding of how disruption at a molecular or cellular level generates epilepsy in the intact organism. We argue that bridging this gap will require large-scale, dynamic, circuit-level analysis. Ongoing research in animal models of TLE has been facilitated by recent advances in network-level functional imaging techniques such as voltage-sensitive dye (VSD) and calcium imaging, which allow simultaneous monitoring of the activity of large numbers of neurons at high temporal resolution. The additional use of optogenetic techniques allows researchers to manipulate defined subsets of neurons within these circuits with millisecond precision[86,129,130]. For example, in an animal model of post-stroke epilepsy, HCN channel dysregulation renders thalamic relay neurons hyperexcitable, and optogenetic silencing of these cells interrupts seizures[131]. These techniques are ideally suited for the study of circuit-level phenomena such as seizures and the circuit-level dysfunctions that underlie epilepsy.

Within the temporal lobe, there is an intimate relationship between the hippocampus and entorhinal cortex — two potentially epileptogenic brain regions — formed by multiple re-entrant circuit loops (FIG. 2). These synaptic loops are important for normal hippocampal network operations but also render the temporal lobe particularly vulnerable to the

generation of abnormal electrical activity and underlie the predisposition of temporal lobe structures to epileptogenesis[132,133]. Here, we focus our discussion on two important discrete circuit elements in the temporal lobe, both of which have been proposed to be involved in the pathology of TLE: the dentate gate between the entorhinal cortex and area CA3, and the temporoammonic pathway between the entorhinal cortex and area CA1. Large-scale dynamic imaging has provided insights into the normal operation and dysfunction of these circuit elements in epilepsy.

## The dentate gate in limbic epileptogenesis

The dentate gyrus has been a particular focus in the study of TLE for various reasons. The dentate gyrus is the locus for the classical temporal lobe circuit rearrangement of mossy fibre sprouting, which is seen in human TLE and in animal models[134,135]; sprouting also occurs from CA3 pyramidal cells and mossy cells in the dentate hilus in animal models of TLE[136]. The time course of the appearance of mossy fibre sprouting, which occurs following a delay after brain injury, is concordant with the latent period of temporal lobe epileptogenesis. However, the role of mossy fibre sprouting in epileptogenesis remains controversial[135]. There are additional anatomical alterations in this region in TLE, including dispersion of dentate granule cells, abnormal granule cell neurogenesis, the appearance of granule cells in ectopic locations and the loss of mossy cells and vulnerable populations of inhibitory interneurons in the dentate gyrus and hilar region.

Under normal conditions, the dentate gyrus strongly filters afferent input, relaying only a small component of incoming, synchronous cortical synaptic inputs to its downstream target, hippocampal area CA3. This phenomenon has been termed the ‘dentate gate’[132,133,137]. Dentate gating is thought to allow the dentate gyrus to filter entorhinal cortex input through the perforant path to the hippocampus, selecting inputs and enacting a ‘sparse coding’ scheme that is thought to be important for the computational functions of the dentate gyrus in pattern separation[138]. Dentate granule cells also have roles in the modulation of CA3 place cell behaviour[138] and in cognitive processes such as learning and memory[139,140]. In addition, the gate is thought to protect downstream area CA3, which is vulnerable to metabolic challenges, from excitotoxic damage that could be induced by aberrant hypersynchronous entorhinal cortical activity. A significant contributor to this emergent network phenomenon is the small percentage of dentate gyrus granule cells that are activated by perforant path stimulation (2–5% *in vitro*[141]). A similarly small percentage (2%) of dentate granule cells are active during spatial navigation in animals *in vivo*[142]. For these reasons, the assessment of gating is important as a network-level output measure of dentate gyrus function under normal conditions and of circuit dysfunction in epilepsy. This phenomenon can readily be shown electrophysiologically *in vitro*, where epileptiform activity in the entorhinal cortex induced by pharmacological manipulation does not propagate through the dentate gyrus to area CA3 under control conditions (FIG. 3) but does so, for example, after blockade of GABAergic inhibition (FIG. 3) or in the kindled hippocampus[143]. Dentate gating has also been demonstrated using large-scale VSD imaging of the dentate: stimulation of the perforant path produced an excitatory response in the inner molecular layer of the dentate gyrus that subsequently spread to the granule cell layer of the dentate gyrus as well as the proximal hilus but did not activate area CA3 (REF. 144). Understanding the mechanisms that underlie the breakdown of dentate filter function is a goal of ongoing research, as protecting or bolstering the gate after brain injury or the induction of status epilepticus could theoretically modulate epileptogenesis.

The mechanisms of dentate gating are probably diverse but are likely to include the intrinsic electrophysiological properties of dentate granule cells, unique integrative features of granule cell dendrites[145] and multiple forms of inhibition that operate within the dentate

gyrus. There is potent feedforward and feedback inhibition of perforant path-evoked dentate gyrus granule cell activity (FIG. 2), and granule cells are also subjected to powerful tonic inhibition. Although the granule cells themselves rarely fire action potentials in response to perforant path activation, resident GABAergic interneurons are also targets of perforant path input (for example, parvalbumin-positive fast-spiking basket cells in the dentate gyrus granule cell layer[146] and, perhaps, somatostatin-positive cells in the hilus[147]) and have been shown to activate readily, thereby constraining granule cell activity through feedforward and feedback inhibition.

There is accumulating evidence that transient dentate gate dysfunction develops during the latent phase of TLE in rodent models. Such data suggest that this dysfunction involves $GABA_A$ receptor alterations, dysregulation of the maintenance of chloride gradients and acute loss of GABAergic interneurons in the dentate hilus. The generation of acute brain slices prepared from epileptic rodents after the induction of status epilepticus combined with large-scale, high-speed dynamic optical imaging of population activity has proven to be a powerful approach to the elucidation of circuit-level mechanisms of epileptogenesis.

VSD imaging in rodent brain slices showed network-level dysfunction of the dentate gate during the latent period of chronic TLE[148]. Dentate-mediated filtering of perforant path activation, as measured by the depolarization of upstream area CA3, was compromised during the latent period but recovered during the chronic phase of TLE (FIG. 4). Hence, the dentate gate is 'open' for a brief period of time corresponding to the latent period. One mechanism underlying the breakdown of the dentate gate is a depolarizing shift of the reversal potential for GABA-mediated postsynaptic potentials ($E_{GABA}$) in dentate gyrus granule neurons[148]: $E_{GABA}$ is typically very hyperpolarized in granule cells under baseline conditions but shifts to markedly less hyperpolarized levels 1 week after the induction of status epilepticus, returning again to baseline levels in the chronic phase of TLE. The basis of this transient depolarizing shift in $E_{GABA}$ is decreased expression of potassium/chloride co-transporter 2 (KCC2; also known as SLC12A5), which normally extrudes chloride from cells and sets $E_{GABA}$ at very hyperpolarized levels in granule cells; the result is altered synaptic integration in dentate granule cells and transient pathological hyper-excitability in response to perforant path activation. The developmental 'GABA switch' that renders GABA hyperpolarizing involves the upregulation of KCC2 and the down-regulation of sodium/potassium/chloride co-transporter 1 (NKCC1; also known as SLC12A2)[149,150]. The above observation is interesting in the context of the finding that KCC2 expression can be suppressed by REST[151] (as well as by neurotrophins[152]), possibly linking a molecular signalling cascade with the transient dentate gate dysfunction that is seen during the latent period.

The potential anti-epileptogenic properties of the NKCC1 antagonist bumetanide have been tested in the pilocarpine-induced status epilepticus model of TLE in rats. Bumetanide, when administered alone after status epilepticus, had no effect on the subsequent development and progression of epilepsy, and the addition of bumetanide to a cocktail of diazepam and phenobarbital administered after status epilepticus did not produce any additional effect on seizure frequency beyond that of diazepam and phenobarbital alone[153]. The development of other strategies to protect the dentate gate after status epilepticus are needed to test the hypothesis that maintenance of dentate gate function during the latent period could be anti-epileptogenic.

## Temporoammonic pathway dysregulation in limbic epileptogenesis

Under normal conditions, the excitatory postsynaptic potential (EPSP) generated by temporo-ammonic input to area CA1 is restricted to the apical tuft of CA1 pyramidal cells by GABAergic inhibition[154], does not propagate to the cell soma[154,155] and hence does not

typically elicit action potential firing[154,155]. Instead, the soma primarily responds to temporoammonic pathway activation with an inhibitory postsynaptic potential, which reflects proximally targeted disynaptic inhibition. This response of area CA1 to temporoammonic pathway stimulation is another circuit-level output measure of a component of the hippocampal circuit that can be used to assay large-scale network function (or dysfunction). The mechanisms that underlie this phenomenon include a high density of HCN channels in distal dendrites of CA1 hippocampal pyramidal cells[156,157] and disynaptic feedforward inhibition that might originate from oriens lacunosum-moleculare (O-LM) interneurons. Pharmacologic blockade of GABAergic inhibition results in the loss of this spatial segregation of temporoammonic excitatory input in the stratum lacunosum-moleculare, with subsequent propagation of temporoammonic EPSPs to the stratum radiatum and stratum oriens[154]. Correspondingly, feedback inhibition driven by stimulation of GABAergic interneurons in the stratum oriens further constrains temporoammonic pathway-evoked EPSPs in CA1 hippocampal pyramidal cells.

Temporoammonic pathway dysfunction in rodent models of acquired chronic TLE results from disinhibition owing to acute loss of interneurons after status epilepticus, upregulation of T-type calcium currents ($I_{Ca,T}$) in CA1 hippocampal pyramidal cells and downregulation of dendritic [158,159]. Overall, dendritic inhibition $I_h$ is decreased in CA1 hippocampal pyramidal cells in rodent models of TLE[160]. The A-type potassium current ($I_A$) is downregulated in CA1 hippocampal pyramidal cell dendrites in the pilocarpine model of chronic TLE in rats[53]. In the same model, $I_{Ca,T}$ appears to be upregulated in dendrites of CA1 hippocampal pyramidal cells, which leads to a change in firing mode from tonic to bursting[159,161–163]. It is thought that CA1 hippocampal pyramidal neurons probably possess an intrinsic burst mechanism mediated by $I_{Ca,T}$ that is normally suppressed by $I_A$; however, downregulation of dendritic $I_A$ in combination with upregulation of $I_{Ca,T}$ unmasks and amplifies aberrant bursting in these cells[159].

Among temporal lobe structures, the dentate gyrus is the least prone to the generation of spontaneous epileptiform discharges *in vitro*[164]. Inhibition in the dentate gyrus is enhanced rather than impaired in the chronic phase in the kainate[165] and pilocarpine[166,167] rat models of TLE[165], which is consistent with the results described above. Surprisingly, although dentate gate function is impaired during the latent period, it recovers in the chronic phase of TLE in a rodent model[144] (FIG. 4). In addition, activation of area CA1 is actually decreased in the chronic phase of TLE in response to the stimulation of the Schaffer collateral pathway. However, it is the processing of entorhinal cortex input by area CA1 that is markedly impaired in chronic TLE. Excitation in area CA1 that occurred in response to the stimulation of the temporoammonic pathway was increased tenfold in chronic TLE[144]. Combined, these dynamic changes create a transient breakdown of the dentate gate, which is followed by long-standing pathological hyperexcitability within the temporoammonic pathway that allows aberrant entorhinal cortical input to enter the hippocampus through the disinhibited temporoammonic pathway. Thus, circuit-level analysis allows network excitability to be compared between control and epileptic brains at multiple time points in the epileptogenic process, integrating the multitude of changes that occur during limbic epileptogenesis and generating a large-scale physiological output measure that correlates with changes in the intact organism.

## Conclusions

Here, we have presented a selected review of recent work on the mechanisms of epileptogenesis, citing specific examples chosen from a set of emerging themes in the field, including dysregulation of molecular signalling pathways, cell type-specific dysfunction and pathology of discrete circuit elements. Some of the issues that have been discussed remain

quite controversial. Although we have largely restricted our discussion to mechanisms of TLE and Dravet syndrome, this focus can and should be expanded to other brain areas and models of other epilepsy syndromes, both acquired and genetic. We argue that the roles of individual molecules, particular cell types or a large-scale molecular signalling cascade in epileptogenesis will be best understood in the context of a circuit analyses. This can be seen in the overlap between the themes that we have discussed.

Epilepsy prevention was defined as Benchmark Area I of the 2007 National Institute of Neurological Disorders and Stroke (NINDS) Epilepsy Research Benchmarks[168]. The data highlighted here illustrate new avenues that hold promise of achieving this goal. Theorized mechanisms of epileptogenesis have moved beyond alterations of ion channels and neurotransmitter receptors to include cell type-specific dysfunction such as 'interneuronopathies' as well as dysregulation of master regulatory pathways such as REST and mTOR. We suggest that these various themes all converge at the level of brain circuits, the defining neurophysiological mediators of epilepsy. However, significant questions face the field as we move forward, and we have raised many more questions than we have answered. This is perhaps necessarily the case as we are still without anti-epileptogenic therapies, although we do have new and qualitatively different treatment targets. Future research directions should explore the effects of genetic manipulations, dysfunction of defined cell types and alterations in key cell signalling pathways on circuit function. Such analyses establish a framework within which we can analyse potential molecular and cellular targets for pharmacological and genetic manipulation and therapeutic intervention in the epileptogenic process. This concept provides support for the idea that epileptogenesis is a clinical problem that is amenable to medical intervention and hope for a future in which epilepsy can be prevented.

## Acknowledgments

We thank L. Isom, E. Marsh and three anonymous referees for assistance in the preparation of this manuscript.

## Glossary

| | |
|---|---|
| **Dravet syndrome** | Previously referred to as severe myoclonic epilepsy of infancy, Dravet syndrome is a spectrum of severe infantile-onset epileptic encephalopathy that is due to an underlying genetic cause, most commonly mutation of the voltage-gated sodium channel subunit $Na_V1.1$. Typically, infants are initially normal, then develop febrile and afebrile seizures, progressing to myoclonus and multiple seizure types, and cognitive impairment |
| **Circuits** | Brain elements that are composed of a collection of neurons of various types and are connected to one another to form a network or ensemble, the operation of which is directed towards a particular function or domain of information processing. Circuits receive inputs, process information and produce outputs |
| **Tuberous sclerosis** | An autosomal dominant multisystem disorder that is caused by a mutation of the tumour suppressor genes tuberous sclerosis complex 1 (*TSC1*; encoding hamartin) or *TSC2* (encoding tuberin). It is accompanied by the characteristic brain lesions of cerebral cortical tubers and subependymal |

| | |
|---|---|
| | nodules, and by the neurological sequelae of epilepsy, variable intellectual disability and autism |
| **Mossy fibre sprouting** | A process thought to be triggered by cell death of hilar mossy cells, leading to de-innervation of dentate granule cell dendrites, with subsequent 'sprouting' of mossy fibre collaterals and pathological granule cell autoinnervation. Whether this is a cause or consequence of temporal lobe pathology in temporal lobe epilepsy is controversial |
| **Hemimegalencephaly** | A malformation of cortical development that is characterized by the pathological enlargement of one cerebral hemisphere and is highly associated with developmental delay and intractable epilepsy |
| **Kainate- or pilocarpine-induced status epilepticus** | Standard and widely used models of acquired chronic focal epilepsy in rodents in which status epilepticus is elicited via the administration of a chemoconvulsant, either the glutamate agonist kainic acid (kainate) or the muscarinic acetylcholine receptor agonist pilocarpine |
| **Hyperpolarization-activated cyclic nucleotide-gated (HCN) channel** | An ion channel that is activated by hyperpolarization and mediates a mixed cationic current known as $I_h$. It has multiple important functions in the regulation of neuronal excitability. HCN channels are encoded by four genes (*HCN1–HCN4*) |
| **Kindling** | A phenomenon whereby seizures themselves contribute to epilepsy progression. The kindling model in experimental animals involves the generation of repeated, brief, focal seizures using electrical or chemical stimulation, which subsequently leads to permanently enhanced sensitivity to such stimuli and longer, more severe seizures, and ultimately to chronic epilepsy |
| **GABA switch** | In the developing brain, GABA is an excitatory neurotransmitter and depolarizes neurons. As the brain matures, a shift in the chloride potential of neurons causes the effects of GABA to become hyperpolarizing and hence inhibitory |
| **Stratum lacunosum-moleculare** | A hippocampal layer that is located superficially relative to the stratum radiatum beneath the pial surface and contains the perforant path input onto distal dendritic tufts of CA1 pyramidal cells |
| **Stratum radiatum** | A hippocampal layer located just superficially relative to the stratum lucidum in area CA3 and to the pyramidal cell layer in areas CA2 and CA1 that contains the Schaffer collateral axons from areas CA3 to CA1 and the dendrites of CA1 pyramidal cells |
| **Stratum oriens** | A relatively cell-free layer in the CA subfields of the hippocampus that is located below the pyramidal cell layer that contains the axons of hippocampal pyramidal cells, along with various types of GABAergic interneurons including oriens-lacunosum moleculare interneurons |

## References


1. Helbig I, Scheffer IE, Mulley JC, Berkovic SF. Navigating the channels and beyond: unravelling the genetics of the epilepsies. Lancet Neurol. 2008; 7:231–245. [PubMed: 18275925]
2. Berkovic SF, et al. Familial temporal lobe epilepsy: a common disorder identified in twins. Ann Neurol. 1996; 40:227–235. [PubMed: 8773604]
3. Kobayashi E, Li LM, Lopes-Cendes I, Cendes F. Magnetic resonance imaging evidence of hippocampal sclerosis in asymptomatic, first-degree relatives of patients with familial mesial temporal lobe epilepsy. Arch Neurol. 2002; 59:1891–1894. [PubMed: 12470176]
4. Cendes F, Lopes-Cendes I, Andermann E, Andermann F. Familial temporal lobe epilepsy: a clinically heterogeneous syndrome. Neurology. 1998; 50:554–557. [PubMed: 9484399]
5. Loscher W, Brandt C. Prevention or modification of epileptogenesis after brain insults: experimental approaches and translational research. Pharmacol Rev. 2010; 62:668–700. [PubMed: 21079040]
6. Pitkänen A, Lukasiuk K. Mechanisms of epileptogenesis and potential treatment targets. Lancet Neurol. 2011; 10:173–186. [PubMed: 21256455]
7. McNamara JO, Huang YZ, Leonard AS. Molecular signaling mechanisms underlying epileptogenesis. Sci STKE. 2006; 2006:re12. [PubMed: 17033045]
8. Binder DK. Neurotrophins in the dentate gyrus. Prog Brain Res. 2007; 163:371–397. [PubMed: 17765730]
9. McNamara, JO.; Scharfman, HE. Jasper's Basic Mechanisms of the Epilepsies. 4. Noebels, JL.; Avoli, M.; Rogawski, MA.; Olsen, RW.; Delgado-Escueta, AV., editors. Oxford Univ. Press; 2012. p. 514-531.The classic textbook on basic mechanisms of epilepsy and epileptogenesis, which is now in its recently-released fourth edition
10. Wong M. Mammalian target of rapamycin (mTOR) inhibition as a potential antiepileptogenic therapy: from tuberous sclerosis to common acquired epilepsies. Epilepsia. 2010; 51:27–36. [PubMed: 19817806]
11. Crino PB. mTOR: a pathogenic signaling pathway in developmental brain malformations. Trends Mol Med. 2011; 17:734–742. A prescient review of diverse types of focal brain malformation, suggesting that a spectrum of disorders are due to aberrant mTOR pathway signalling ('TORopathies'). [PubMed: 21890410]
12. Laplante M, Sabatini DM. mTOR signaling in growth control and disease. Cell. 2012; 149:274–293. [PubMed: 22500797]
13. Qureshi IA, Mehler MF. Epigenetic mechanisms underlying human epileptic disorders and the process of epileptogenesis. Neurobiol Dis. 2010; 39:53–60. [PubMed: 20188170]
14. Brooks-Kayal AR, Raol YH, Russek SJ. Alteration of epileptogenesis genes. Neurotherapeutics. 2009; 6:312–318. [PubMed: 19332325]
15. Tang SJ, et al. A rapamycin-sensitive signaling pathway contributes to long-term synaptic plasticity in the hippocampus. Proc Nat Acad Sci USA. 2002; 99:467–472. [PubMed: 11756682]
16. Jaworski J, Spangler S, Seeburg DP, Hoogenraad CC, Sheng M. Control of dendritic arborization by the phosphoinositide-3′-kinase–Akt–mammalian target of rapamycin pathway. J Neurosci. 2005; 25:11300–11312. [PubMed: 16339025]
17. Raab-graham KF, Haddick PCG, Jan YN, Jan LY. Activity- and mTOR-dependent suppression of Kv1.1 channel mRNA translation in dendrites. Science. 2006; 314:144–148. [PubMed: 17023663]
18. Crino PB. Focal brain malformations: seizures, signaling, sequencing. Epilepsia. 2009; 50:3–8. [PubMed: 19761448]
19. Striano P, Zara F. Genetics: mutations in mTOR pathway linked to megalencephaly syndromes. Nature Rev Neurol. 2012; 8:542–544. [PubMed: 22907262]
20. McDaniel SS, Wong M. Therapeutic role of mammalian target of rapamycin (mTOR) inhibition in preventing epileptogenesis. Neurosci Lett. 2011; 497:231–239. [PubMed: 21354266]
21. Wong M. Mechanisms of epileptogenesis in tuberous sclerosis complex and related malformations of cortical development with abnormal glioneuronal proliferation. Epilepsia. 2008; 49:8–21. [PubMed: 17727667]
22. Holmes GL, Stafstrom CE. Tuberous sclerosis complex and epilepsy: recent developments and future challenges. Epilepsia. 2007; 48:617–630. [PubMed: 17386056]

23. Mohamed AR, et al. Intrinsic epileptogenicity of cortical tubers revealed by intracranial EEG monitoring. Neurology. 2012; 79:2249–2257. [PubMed: 23175730]
24. Uhlmann EJ, et al. Astrocyte-specific *TSC1* conditional knockout mice exhibit abnormal neuronal organization and seizures. Ann Neurol. 2002; 52:285–296. [PubMed: 12205640]
25. Zeng LH, et al. Abnormal glutamate homeostasis and impaired synaptic plasticity and learning in a mouse model of tuberous sclerosis complex. Neurobiol Dis. 2007; 28:184–196. [PubMed: 17714952]
26. Zeng LH, Xu L, Gutmann DH, Wong M. Rapamycin prevents epilepsy in a mouse model of tuberous sclerosis complex. Ann Neurol. 2008; 63:444–453. [PubMed: 18389497]
27. Pun RYK, et al. Excessive activation of mTOR in postnatally generated granule cells is sufficient to cause epilepsy. Neuron. 2012; 75:1022–1034. Selective deletion of PTEN from a small subset of dentate gyrus granule cells in adult mice is sufficient to generate TLE. [PubMed: 22998871]
28. Lee JH, et al. *De novo* somatic mutations in components of the PI3K–AKT3–mTOR pathway cause hemimegalencephaly. Nature Genet. 2012; 44:941–945. [PubMed: 22729223]
29. Poduri A, et al. Somatic activation of *AKT3* causes hemispheric developmental brain malformations. Neuron. 2012; 74:41–48. [PubMed: 22500628]
30. Tokuda S, et al. A novel *Akt3* mutation associated with enhanced kinase activity and seizure susceptibility in mice. Hum Mol Genet. 2011; 20:988–999. [PubMed: 21159799]
31. Zeng LH, Rensing NR, Wong M. The mammalian target of rapamycin signaling pathway mediates epileptogenesis in a model of temporal lobe epilepsy. J Neurosci. 2009; 29:6964–6972. [PubMed: 19474323]
32. Buckmaster PS, Ingram EA, Wen X. Inhibition of the mammalian target of rapamycin signaling pathway suppresses dentate granule cell axon sprouting in a rodent model of temporal lobe epilepsy. J Neurosci. 2009; 29:8259–8269. [PubMed: 19553465]
33. Huang X, et al. Pharmacological inhibition of the mammalian target of rapamycin pathway suppresses acquired epilepsy. Neurobiol Dis. 2010; 40:193–199. [PubMed: 20566381]
34. Buckmaster PS, Lew FH. Rapamycin suppresses mossy fiber sprouting but not seizure frequency in a mouse model of temporal lobe epilepsy. J Neurosci. 2011; 31:2337–2347. Rapamycin given to mice after pilocarpine-induced status epilepticus markedly decreased the extent of mossy fibre sprouting, with no effect on the development of epilepsy or on seizure frequency. [PubMed: 21307269]
35. Van Vliet, Ea, et al. Inhibition of mammalian target of rapamycin reduces epileptogenesis and blood–brain barrier leakage but not microglia activation. Epilepsia. 2012; 53:1254–1263. [PubMed: 22612226]
36. Sliwa A, Plucinska G, Bednarczyk J, Lukasiuk K. Post-treatment with rapamycin does not prevent epileptogenesis in the amygdala stimulation model of temporal lobe epilepsy. Neurosci Lett. 2012; 509:105–109. [PubMed: 22227620]
37. Erlich S, Alexandrovich A, Shohami E, Pinkas-Kramarski R. Rapamycin is a neuroprotective treatment for traumatic brain injury. Neurobiol Dis. 2007; 26:86–93. [PubMed: 17270455]
38. Chen S, et al. Alterations in mammalian target of rapamycin signaling pathways after traumatic brain injury. J Cereb Blood Flow Metab. 2007; 27:939–949. [PubMed: 16955078]
39. Raffo E, Coppola A, Ono T, Briggs SW, Galanopoulou AS. A pulse rapamycin therapy for infantile spasms and associated cognitive decline. Neurobiol Dis. 2011; 43:322–329. [PubMed: 21504792]
40. Galanopoulou AS, et al. Identification of new epilepsy treatments: issues in preclinical methodology. Epilepsia. 2012; 53:571–582. [PubMed: 22292566]
41. Galanopoulou AS, Gorter Ja, Cepeda C. Finding a better drug for epilepsy: the mTOR pathway as an antiepileptogenic target. Epilepsia. 2012; 53:1119–1130. [PubMed: 22578218]
42. Palm K, Belluardo N, Metsis M, Timmusk T. Neuronal expression of zinc finger transcription factor REST/NRSF/XBR gene. J Neurosci. 1998; 18:1280–1296. [PubMed: 9454838]
43. Ballas N, Grunseich C, Lu DD, Speh JC, Mandel G. REST and its corepressors mediate plasticity of neuronal gene chromatin throughout neurogenesis. Cell. 2005; 121:645–657. [PubMed: 15907476]

44. Johnson DS, Mortazavi A, Myers RM, Wold B. Genome-wide mapping of *in vivo* protein–DNA interactions. Science. 2007; 316:1497–1502. [PubMed: 17540862]
45. Otto SJ, et al. A new binding motif for the transcriptional repressor REST uncovers large gene networks devoted to neuronal functions. J Neurosci. 2007; 27:6729–6739. [PubMed: 17581960]
46. Mortazavi A, Leeper Thompson EC, Garcia ST, Myers RM, Wold B. Comparative genomics modeling of the NRSF/REST repressor network: from single conserved sites to genome-wide repertoire. Genome Res. 2006; 16:1208–1221. [PubMed: 16963704]
47. Roopra A, Huang Y, Dingledine R. Neurological disease: listening to gene silencers. Mol Interv. 2001; 1:219–228. [PubMed: 14993344]
48. Roopra A, Dingledine R, Hsieh J. Epigenetics and epilepsy. Epilepsia. 2012; 53:2–10. [PubMed: 23216574]
49. Huang Y, Doherty JJ, Dingledine R. Altered histone acetylation at glutamate receptor 2 and brain-derived neurotrophic factor genes is an early event triggered by status epilepticus. J Neurosci. 2002; 22:8422–8428. [PubMed: 12351716]
50. Huang Y, Myers SJ, Dingledine R. Transcriptional repression by REST: recruitment of Sin3A and histone deacetylase to neuronal genes. Nature Neurosci. 1999; 2:867–872. [PubMed: 10491605]
51. Garriga-Canut M, et al. 2-Deoxy-D-glucose reduces epilepsy progression by NRSF-CtBP-dependent metabolic regulation of chromatin structure. Nature Neurosci. 2006; 9:1382–1387. Pharmacologic inhibition of glycolysis during kindling epileptogenesis recruits REST to target genes including *BDNF*, leading to suppression of BDNF expression, reduced neuronal excitability and attenuated progression of kindling. [PubMed: 17041593]
52. Noam Y, Bernard C, Baram TZ. Towards an integrated view of HCN channel role in epilepsy. Curr Opin Neurobiol. 2011; 21:873–879. [PubMed: 21782415]
53. Bernard C, et al. Acquired dendritic channelopathy in temporal lobe epilepsy. Science. 2004; 305:532–535. [PubMed: 15273397]
54. Jung S, Warner LN, Pitsch J, Becker AJ, Poolos NP. Rapid loss of dendritic HCN channel expression in hippocampal pyramidal neurons following status epilepticus. J Neurosci. 2011; 31:14291–14295. [PubMed: 21976514]
55. Chen K, et al. Persistently modified h-channels after complex febrile seizures convert the seizure-induced enhancement of inhibition to hyperexcitability. Nature Med. 2001; 7:331–337. [PubMed: 11231632]
56. McClelland S, et al. Neuron-restrictive silencer factor-mediated hyperpolarization-activated cyclic nucleotide gated channelopathy in experimental temporal lobe epilepsy. Ann Neurol. 2011; 70:454–464. In the kainate model of acquired chronic TLE in rodents, interference with the interaction between REST and target genes using antisense oligodeoxy-nucleotides prevented status epilepticus-induced REST-mediated repression of HCN1 and led to a decrease in spontaneous seizure frequency. [PubMed: 21905079]
57. Freund TF, Buzsáki G. Interneurons of the hippocampus. Hippocampus. 1996; 6:347–470. [PubMed: 8915675]
58. Somogyi P, Klausberger T. Defined types of cortical interneurone structure space and spike timing in the hippocampus. J Physiol. 2005; 562:9–26. [PubMed: 15539390]
59. Ascoli GA, et al. Petilla terminology: nomenclature of features of GABAergic interneurons of the cerebral cortex. Nature Rev Neurosci. 2008; 9:557–568. [PubMed: 18568015]
60. Defelipe J, et al. New insights into the classification and nomenclature of cortical GABAergic interneurons. Nature Rev Neurosci. 2013:202–216. [PubMed: 23385869]
61. Meldrum B. Physiological changes during prolonged seizures and epileptic brian damage. Neuropadiatrie. 1978; 9:213–212. [PubMed: 581397]
62. Bernard C, Cossart R, Hirsch JC, Esclapez M, Ben-Ari Y. What is GABAergic inhibition? How is it modified in epilepsy? Epilepsia. 2000; 41:S90–S95. [PubMed: 10999527]
63. Mann EO, Mody I. The multifaceted role of inhibition in epilepsy: seizure-genesis through excessive GABAergic inhibition in autosomal dominant nocturnal frontal lobe epilepsy. Curr Opin Neurol. 2008; 21:155–160. [PubMed: 18317273]
64. Galanopoulou AS. Mutations affecting GABAergic signaling in seizures and epilepsy. Pflügers Arch. 2010; 460:505–523. [PubMed: 20352446]

65. Sebe JY, Baraban SC. The promise of an interneuron-based cell therapy for epilepsy. Dev Neurobiol. 2011; 71:107–117. [PubMed: 21154914]
66. Anderson, SA.; Baraban, SC. Jasper's Basic Mechanisms of the Epilepsies. 4. Noebels, JL.; Avoli, M.; Rogawski, MA.; Olsen, RW.; Delgado-Escueta, AV., editors. Oxford Univ. Press; 2012. p. 1122-1126.
67. Maisano X, et al. Differentiation and functional incorporation of embryonic stem cell-derived GABAergic interneurons in the dentate gyrus of mice with temporal lobe epilepsy. J Neurosci. 2012; 32:46–61. [PubMed: 22219269]
68. Houser CR. Interneurons of the dentate gyrus: an overview of cell types, terminal fields and neurochemical identity. Prog Brain Res. 2007; 163:217–232. [PubMed: 17765721]
69. Jinno S, Kosaka T. Patterns of expression of neuropeptides in GABAergic nonprincipal neurons in the mouse hippocampus: quantitative analysis with optical disector. J Comp Neurol. 2003; 461:333–349. [PubMed: 12746872]
70. Bakst I, Avendano C, Morrison JH, Amaral DG. An experimental analysis of the origins of somatostatin-like immunoreactivity in the dentate gyrus of the rat. J Neurosci. 1986; 6:1452–1462. [PubMed: 2872280]
71. Katona I, Acsády L, Freund TF. Postsynaptic targets of somatostatin-immunoreactive interneurons in the rat hippocampus. Neuroscience. 1999; 88:37–55. [PubMed: 10051188]
72. Acsády L, Kamondi A, Sík A, Freund T, Buzsáki G. GABAergic cells are the major postsynaptic targets of mossy fibers in the rat hippocampus. J Neurosci. 1998; 18:3386–3403. [PubMed: 9547246]
73. Sloviter RS. Decreased hippocampal inhibition and a selective loss of interneurons in experimental epilepsy. Science. 1987; 235:73–76. [PubMed: 2879352]
74. Buckmaster PS, Dudek FE. Neuron loss, granule cell axon reorganization, and functional changes in the dentate gyrus of epileptic kainate-treated rats. J Comp Neurol. 1997; 385:385–404. [PubMed: 9300766]
75. De Lanerolle NC, Kim JH, Robbins RJ, Spencer DD. Hippocampal interneuron loss and plasticity in human temporal lobe epilepsy. Brain Res. 1989; 495:387–395. [PubMed: 2569920]
76. Zhang W, et al. Surviving hilar somatostatin interneurons enlarge, sprout axons, and form new synapses with granule cells in a mouse model of temporal lobe epilepsy. J Neurosci. 2009; 29:14247–14256. In a mouse model of chronic TLE, surviving somatostatin- positive hilar interneurons exhibit axonal sprouting and form functional inhibitory connections with dentate gyrus granule cells at increased frequency as demonstrated anatomically and physiologically. [PubMed: 19906972]
77. Mathern GW, Babb TL, Pretorius JK, Leite JP. Reactive synaptogenesis and neuron densities for neuropeptide Y, somatostatin, and glutamate decarboxylase immunoreactivity in the epileptogenic human fascia dentata. J Neurosci. 1995; 15:3990–4004. [PubMed: 7751960]
78. Buckmaster PS, Wen X. Rapamycin suppresses axon sprouting by somatostatin interneurons in a mouse model of temporal lobe epilepsy. Epilepsia. 2011; 52:2057–2064. [PubMed: 21883182]
79. Gonchar Y, Wang Q, Burkhalter A. Multiple distinct subtypes of GABAergic neurons in mouse visual cortex identified by triple immunostaining. Front Neuroanat. 2007; 1:3. [PubMed: 18958197]
80. Xu X, Roby KD, Callaway EM. Immunochemical characterization of inhibitory mouse cortical neurons: three chemically distinct classes of inhibitory cells. J Comp Neurol. 2010; 518:389–404. [PubMed: 19950390]
81. Pouille F, Scanziani M. Enforcement of temporal fidelity in pyramidal cells by somatic feed-forward inhibition. Science. 2001; 293:1159–1163. [PubMed: 11498596]
82. Bartos M, Vida I, Jonas P. Synaptic mechanisms of synchronized gamma oscillations in inhibitory interneuron networks. Nature Rev Neurosci. 2007; 8:45–56. [PubMed: 17180162]
83. Cardin JA, et al. Driving fast-spiking cells induces gamma rhythm and controls sensory responses. Nature. 2009; 459:663–667. [PubMed: 19396156]
84. Sohal VS, Zhang F, Yizhar O, Deisseroth K. Parvalbumin neurons and gamma rhythms enhance cortical circuit performance. Nature. 2009; 459:698–702. [PubMed: 19396159]

85. Avoli M, De Curtis M. GABAergic synchronization in the limbic system and its role in the generation of epileptiform activity. Prog Neurobiol. 2011; 95:104–132. [PubMed: 21802488]
86. Krook-Magnuson E, Armstrong C, Oijala M, Soltesz I. On-demand optogenetic control of spontaneous seizures in temporal lobe epilepsy. Nature Commun. 2013; 4:1376. [PubMed: 23340416]
87. Cobos I, et al. Mice lacking *Dlx1* show subtype-specific loss of interneurons, reduced inhibition and epilepsy. Nature Neurosci. 2005; 8:1059–1068. [PubMed: 16007083]
88. Butt SJB, et al. The requirement of *Nkx2-1* in the temporal specification of cortical interneuron subtypes. Neuron. 2008; 59:722–732. [PubMed: 18786356]
89. Batista-Brito R, et al. The cell-intrinsic requirement of *Sox6* for cortical interneuron development. Neuron. 2009; 63:466–481. [PubMed: 19709629]
90. Smart SL, et al. Deletion of the $K_V1.1$ potassium channel causes epilepsy in mice. Neuron. 1998; 20:809–819. [PubMed: 9581771]
91. Eunson LH, et al. Clinical, genetic, and expression studies of mutations in the potassium channel gene *KCNA1* reveal new phenotypic variability. Ann Neurol. 2000; 48:647–656. [PubMed: 11026449]
92. Goldberg EM, et al. $K^+$ channels at the axon initial segment dampen near-threshold excitability of neocortical fast-spiking GABAergic interneurons. Neuron. 2008; 58:387–400. [PubMed: 18466749]
93. Lau D, et al. Impaired fast-spiking, suppressed cortical inhibition, and increased susceptibility to seizures in mice lacking Kv3.2 $K^+$ channel proteins. J Neurosci. 2000; 20:9071–9085. [PubMed: 11124984]
94. Escayg A, et al. Mutations of *SCN1A*, encoding a neuronal sodium channel, in two families with GEFS+2. Nature Genet. 2000; 24:343–345. [PubMed: 10742094]
95. Claes L, et al. *De novo* mutations in the sodium-channel gene *SCN1A* cause severe myoclonic epilepsy of infancy. Am J Hum Genet. 2001; 68:1327–1332. [PubMed: 11359211]
96. Harkin LA, et al. The spectrum of *SCN1A*-related infantile epileptic encephalopathies. Brain. 2007; 130:843–852. [PubMed: 17347258]
97. Ohmori I, Kahlig KM, Rhodes TH, Wang DW, George AL. Nonfunctional *SCN1A* is common in severe myoclonic epilepsy of infancy. Epilepsia. 2006; 47:1636–1642. [PubMed: 17054685]
98. Yu FH, et al. Reduced sodium current in GABAergic interneurons in a mouse model of severe myoclonic epilepsy in infancy. Nature Neurosci. 2006; 9:1142–1149. [PubMed: 16921370]
99. Ragsdale DS. How do mutant Nav1.1 sodium channels cause epilepsy? Brain Res Rev. 2008; 58:149–159. [PubMed: 18342948]
100. Oakley JC, Kalume F, Yu FH, Scheuer T, Catterall WA. Temperature- and age-dependent seizures in a mouse model of severe myoclonic epilepsy in infancy. Proc Nat Acad Sci USA. 2009; 106:3994–3999. [PubMed: 19234123]
101. Mullen SA, Scheffer IE. Translational research in epilepsy genetics: sodium channels in man to interneuronopathy in mouse. Arch Neurol. 2009; 66:21–26. [PubMed: 19139296]
102. Martin MS, et al. Altered function of the *SCN1A* voltage-gated sodium channel leads to γ-aminobutyric acid-ergic (GABAergic) interneuron abnormalities. J Biol Chem. 2010; 285:9823–9834. [PubMed: 20100831]
103. Escayg A, Goldin AL. Sodium channel *SCN1A* and epilepsy: mutations and mechanisms. Epilepsia. 2010; 51:1650–1658. [PubMed: 20831750]
104. Ogiwara I, et al. $Na_V1.1$ localizes to axons of parvalbumin-positive inhibitory interneurons: a circuit basis for epileptic seizures in mice carrying an *Scn1a* gene mutation. J Neurosci. 2007; 27:5903–5914. Knock-in mice harbouring an *Scn1a* nonsense mutation that is also found in patients with Dravet syndrome develop temperature-sensitive and spontaneous seizures resembling the human condition. Immunohistochemistry indicates restricted expression of $Na_V1.1$ protein at the soma and AIS of parvalbumin-positive basket cells; neocortical fast-spiking cells exhibit profoundly abnormal physiology. [PubMed: 17537961]
105. Cheah CS, et al. Specific deletion of $Na_V1.1$ sodium channels in inhibitory interneurons causes seizures and premature death in a mouse model of Dravet syndrome. Proc Nat Acad Sci USA. 2012; 109:14646–14651. [PubMed: 22908258]

106. Jonas P, Bischofberger J, Fricker D, Miles R. Interneuron diversity series: fast in, fast out--temporal and spatial signal processing in hippocampal interneurons. Trend Neurosci. 2004; 27:30–40. [PubMed: 14698608]

107. Goldberg EM, et al. Rapid developmental maturation of neocortical FS cell intrinsic excitability. Cererb Cortex. 2011; 21:666–682.

108. Westenbroek RE, Merrick DK, Catterall WA. Differential subcellular localization of the RI and RII $Na^+$ channel subtypes in central neurons. Neuron. 1989; 3:695–704. [PubMed: 2561976]

109. Gong B, Rhodes KJ, Bekele-Arcuri Z, Trimmer JS. Type I and type II $Na^+$ channel α-subunit polypeptides exhibit distinct spatial and temporal patterning, and association with auxiliary subunits in rat brain. J Comp Neurol. 1999; 412:342–352. [PubMed: 10441760]

110. Lorincz A, Nusser Z. Cell-type-dependent molecular composition of the axon initial segment. J Neurosci. 2008; 28:14329–14340. [PubMed: 19118165]

111. Harkin LA, et al. Truncation of the $GABA_A$-receptor γ2 subunit in a family with generalized epilepsy with febrile seizures plus. Am J Hum Genet. 2002; 70:530–536. [PubMed: 11748509]

112. Patino, Ga, et al. A functional null mutation of *SCN1B* in a patient with Dravet syndrome. J Neurosci. 2009; 29:10764–10778. [PubMed: 19710327]

113. Ogiwara I, et al. A homozygous mutation of voltage-gated sodium channel $\beta_I$ gene *SCN1B* in a patient with Dravet syndrome. Epilepsia. 2012; 53:e200–e203. [PubMed: 23148524]

114. Kamiya K, et al. A nonsense mutation of the sodium channel gene *SCN2A* in a patient with intractable epilepsy and mental decline. J Neurosci. 2004; 24:2690–2698. [PubMed: 15028761]

115. Shi X, et al. Missense mutation of the sodium channel gene *SCN2A* causes Dravet syndrome. Brain Dev. 2009; 31:758–762. [PubMed: 19783390]

116. Depienne C, et al. Sporadic infantile epileptic encephalopathy caused by mutations in *PCDH19* resembles Dravet syndrome but mainly affects females. PLoS Genet. 2009; 5:e1000381. [PubMed: 19214208]

117. Chen C, et al. Mice lacking sodium channel β1 subunits display defects in neuronal excitability, sodium channel expression, and nodal architecture. J Neurosci. 2004; 24:4030–4042. [PubMed: 15102918]

118. McClelland S, Dubé CM, Yang J, Baram TZ. Epileptogenesis after prolonged febrile seizures: mechanisms, biomarkers and therapeutic opportunities. Neurosci Lett. 2011; 497:155–162. [PubMed: 21356275]

119. Shinnar S, et al. MRI abnormalities following febrile status epilepticus in children: the FEBSTAT study. Neurology. 2012; 79:871–877. [PubMed: 22843278]

120. Abou-Khalil B, et al. Partial and generalized epilepsy with febrile seizures plus and a novel *SCN1A* mutation. Neurology. 2001; 57:2265–2272. [PubMed: 11756608]

121. Sugawara T, et al. *$Na_v1.1$* mutations cause febrile seizures associated with afebrile partial seizures. Neurology. 2001; 57:703–705. [PubMed: 11524484]

122. Scheffer IE, et al. Temporal lobe epilepsy and $GEFS^+$ phenotypes associated with *SCN1B* mutations. Brain. 2007; 130:100–109. An interesting case series including a patient with GEFS+ due to the *SCN1B* mutation but no history of febrile or afebrile status epilepticus who developed TLE with classic mesial temporal sclerosis and become seizure-free after unilateral temporal lobectomy. [PubMed: 17020904]

123. Kearney JA, et al. A gain-of-function mutation in the sodium channel gene *Scn2a* results in seizures and behavioral abnormalities. Neuroscience. 2001; 102:307–317. [PubMed: 11166117]

124. Striano P, et al. Brain MRI findings in severe myoclonic epilepsy in infancy and genotype–phenotype correlations. Epilepsia. 2007; 48:1092–1096. [PubMed: 17381446]

125. Siegler Z, et al. Hippocampal sclerosis in severe myoclonic epilepsy in infancy: a retrospective MRI study. Epilepsia. 2005; 46:704–708. [PubMed: 15857436]

126. Guerrini R, Striano P, Catarino C, Sisodiya SM. Neuroimaging and neuropathology of Dravet syndrome. Epilepsia. 2011; 52:30–34. [PubMed: 21463276]

127. Nangia S, Caraballo RH, Kang HC, Nordli DR, Scheffer IE. Is the ketogenic diet effective in specific epilepsy syndromes? Epilepsy Res. 2012; 100:252–257. [PubMed: 22424762]

128. Brackenbury WJ, Yuan Y, Malley HAO, Parent JM, Isom LL. Abnormal neuronal patterning occurs during early postnatal brain development of *Scn1b*-null mice and precedes hyperexcitability. Proc Nat Acad Sci USA. 2013; 110:1089–1094. [PubMed: 23277545]
129. Tye KM, Deisseroth K. Optogenetic investigation of neural circuits underlying brain disease in animal models. Nature Rev Neurosci. 2012; 13:251–266. [PubMed: 22430017]
130. Bernstein JG, Boyden ES. Optogenetic tools for analyzing the neural circuits of behavior. Trend Cog Sci. 2011; 15:592–600.
131. Paz JT, et al. Closed-loop optogenetic control of thalamus as a tool for interrupting seizures after cortical injury. Nature Neurosci. 2013; 16:64–70. [PubMed: 23143518]
132. Heinemann U, et al. The dentate gyrus as a regulated gate for the propagation of epileptiform activity. Epilepsy Res Suppl. 1992; 7:273–280. [PubMed: 1334666]
133. Lothman EW, Stringer JL, Bertram EH. The dentate gyrus as a control point for seizures in the hippocampus and beyond. Epilepsy Res Suppl. 1992; 7:301–313. [PubMed: 1334669]
134. Dudek FE, Sutula TP. Epileptogenesis in the dentate gyrus: a critical perspective. Prog Brain Res. 2007; 163:755–773. [PubMed: 17765749]
135. Buckmaster, PS. Jasper's Basic Mechanisms of the Epilepsies. 4. Noebels, JL.; Avoli, M.; Rogawski, MA.; Olsen, RW.; Delgado-Escueta, AV., editors. Oxford Univ. Press; 2012. p. 416-431.
136. Zhang W, Huguenard JR, Buckmaster PS. Increased excitatory synaptic input to granule cells from hilar and CA3 regions in a rat model of temporal lobe epilepsy. J Neurosci. 2012; 32:1183–1196. [PubMed: 22279204]
137. Coulter DA, Carlson GC. Functional regulation of the dentate gyrus by GABA-mediated inhibition. Prog Brain Res. 2007; 163:235–243. [PubMed: 17765722]
138. Leutgeb JK, Leutgeb S, Moser MB, Moser EI. Pattern separation in the dentate gyrus and CA3 of the hippocampus. Science. 2007; 315:961–966. [PubMed: 17303747]
139. Clelland CD, et al. A functional role for adult hippocampal neurogenesis in spatial pattern separation. Science. 2009; 325:210–213. [PubMed: 19590004]
140. McHugh TJ, et al. Dentate gyrus NMDA receptors mediate rapid pattern separation in the hippocampal network. Science. 2007; 317:94–99. [PubMed: 17556551]
141. Yu EP, et al. Protracted postnatal development of sparse, specific dentate granule cell activation in the mouse hippocampus. J Neurosci. 2013; 33:2947–2960. [PubMed: 23407953]
142. Chawla MK, et al. Sparse, environmentally selective expression of *Arc* RNA in the upper blade of the rodent fascia dentata by brief spatial experience. Hippocampus. 2005; 15:579–586. [PubMed: 15920719]
143. Behr J, Lyson KJ, Mody I. Enhanced propagation of epileptiform activity through the kindled dentate gyrus. J Neurophysiol. 1998; 79:1726–1732. [PubMed: 9535942]
144. Ang CW, Carlson GC, Coulter DA. Massive and specific dysregulation of direct cortical input to the hippocampus in temporal lobe epilepsy. J Neurosci. 2006; 26:11850–11856. [PubMed: 17108158]
145. Krueppel R, Remy S, Beck H. Dendritic integration in hippocampal dentate granule cells. Neuron. 2011; 71:512–528. Unique synaptic integration by dentate gyrus granule cell dendrites relative to other principal cells in the cerebral cortex may contribute to dentate gating. [PubMed: 21835347]
146. Ewell LA, Jones MV. Frequency-tuned distribution of inhibition in the dentate gyrus. J Neurosci. 2010; 30:12597–12607. This paper shows that the dentate gate is subserved at least in part by feedforward inhibition by fast-spiking interneurons onto dentate gyrus granule cells. However, granule cell activity in response to stimulation of the perforant path is frequency-dependent. [PubMed: 20861366]
147. Leranth C, Malcolm AJ, Frotscher M. Afferent and efferent synaptic connections of somatostatin-immunoreactive neurons in the rat fascia dentata. J Comp Neurol. 1990; 295:111–122. [PubMed: 1971287]
148. Pathak HR, et al. Disrupted dentate granule cell chloride regulation enhances synaptic excitability during development of temporal lobe epilepsy. J Neurosci. 2007; 27:14012–14022. [PubMed: 18094240]

149. Ben-Ari Y. Excitatory actions of gaba during development: the nature of the nurture. Nature Rev Neurosci. 2002; 3:728–739. [PubMed: 12209121]

150. Owens DF, Kriegstein AR. Is there more to GABA than synaptic inhibition? Nature Rev Neurosci. 2002; 3:715–727. [PubMed: 12209120]

151. Yeo M, Berglund K, Augustine G, Liedtke W. Novel repression of *Kcc2* transcription by REST–RE-1 controls developmental switch in neuronal chloride. J Neurosci. 2009; 29:14652–14662. [PubMed: 19923298]

152. Rivera C, et al. Mechanism of activity-dependent downregulation of the neuron-specific K-Cl cotransporter KCC2. J Neurosci. 2004; 24:4683–4691. [PubMed: 15140939]

153. Brandt C, Nozadze M, Heuchert N, Rattka M, Löscher W. Disease-modifying effects of phenobarbital and the NKCC1 inhibitor bumetanide in the pilocarpine model of temporal lobe epilepsy. J Neurosci. 2010; 30:8602–8612. [PubMed: 20573906]

154. Ang CW, Carlson GC, Coulter DA. Hippocampal CA1 circuitry dynamically gates direct cortical inputs preferentially at theta frequencies. J Neurosci. 2005; 25:9567–9580. [PubMed: 16237162]

155. Soltesz I. Brief history of cortico-hippocampal time with a special reference to the direct entorhinal input to CA1. Hippocampus. 1995; 5:120–124. [PubMed: 7633513]

156. Tsay D, Dudman JT, Siegelbaum SA. HCN1 channels constrain synaptically evoked $Ca^{2+}$ spikes in distal dendrites of CA1 pyramidal neurons. Neuron. 2007; 56:1076–1089. [PubMed: 18093528]

157. Magee JC. Dendritic $I_h$ normalizes temporal summation in hippocampal CA1 neurons. Nature Neurosci. 1999; 2:508–514. [PubMed: 10448214]

158. Coulter DA, et al. Hippocampal microcircuit dynamics probed using optical imaging approaches. J Physiol. 2011; 589:1893–1903. [PubMed: 21224219]

159. Beck H, Yaari Y. Plasticity of intrinsic neuronal properties in CNS disorders. Nature Rev Neurosci. 2008; 9:357–369. [PubMed: 18425090]

160. Cossart R, et al. Dendritic but not somatic GABAergic inhibition is decreased in experimental epilepsy. Nature Neurosci. 2001; 4:52–62. [PubMed: 11135645]

161. Sanabria ER, Su H, Yaari Y. Initiation of network bursts by $Ca^{2+}$-dependent intrinsic bursting in the rat pilocarpine model of temporal lobe epilepsy. J Physiol. 2001; 532:205–216. [PubMed: 11283235]

162. Su H, et al. Upregulation of a T-type $Ca^{2+}$ channel causes a long-lasting modification of neuronal firing mode after status epilepticus. J Neurosci. 2002; 22:3645–3655. A thorough review of ion channel alterations in epilepsy and other neurological disorders. [PubMed: 11978840]

163. Yaari Y, Yue C, Su H. Recruitment of apical dendritic T-type $Ca^{2+}$ channels by backpropagating spikes underlies *de novo* intrinsic bursting in hippocampal epileptogenesis. J Physiol. 2007; 580:435–450. [PubMed: 17272342]

164. Bear J, Lothman EW. An *in vitro* study of focal epileptogenesis in combined hippocampalparahippocampal slices. Epilepsy Res. 1993; 14:183–193. [PubMed: 8504790]

165. Wu K, Leung LS. Increased dendritic excitability in hippocampal ca1 *in vivo* in the kainic acid model of temporal lobe epilepsy: a study using current source density analysis. Neuroscience. 2003; 116:599–616. [PubMed: 12559115]

166. Gibbs JW, Shumate MD, Coulter DA. Differential epilepsy-associated alterations in postsynaptic $GABA_A$ receptor function in dentate granule and CA1 neurons. J Neurophysiol. 1997; 77:1924–1938. [PubMed: 9114245]

167. Cohen AS, Lin DD, Quirk GL, Coulter DA. Dentate granule cell $GABA_A$ receptors in epileptic hippocampus: enhanced synaptic efficacy and altered pharmacology. Eur J Neurosci. 2003; 17:1607–1616. [PubMed: 12752378]

168. Kelley MS, Jacobs MP, Lowenstein DH. The NINDS epilepsy research benchmarks. Epilepsia. 2009; 50:579–582. [PubMed: 19317887]

169. Temkin NR. Preventing and treating posttraumatic seizures: the human experience. Epilepsia. 2009; 50:10–13. [PubMed: 19187289]

170. Vadlamudi L, Scheffer IE, Berkovic SF. Genetics of temporal lobe epilepsy. J Neurol Neurosurg Psych. 2003; 74:1359–1361.

171. Ribeiro P, et al. Expression profile of *Lgi1* gene in mouse brain during development. J Mol Neurosci. 2008; 35:323–329. [PubMed: 18563303]

172. Michelucci R, Pasini E, Nobile C. Lateral temporal lobe epilepsies: clinical and genetic features. Epilepsia. 2009; 50:52–54. [PubMed: 19469848]

173. Amaral DG, Scharfman HE, Lavenex P. The dentate gyrus: fundamental neuroanatomical organization (dentate gyrus for dummies). Prog Brain Res. 2007; 163:3–22. [PubMed: 17765709]

174. Morgan RJ, Santhakumar V, Soltesz I. Modeling the dentate gyrus. Prog Brain Res. 2007; 163:639–658. [PubMed: 17765743]

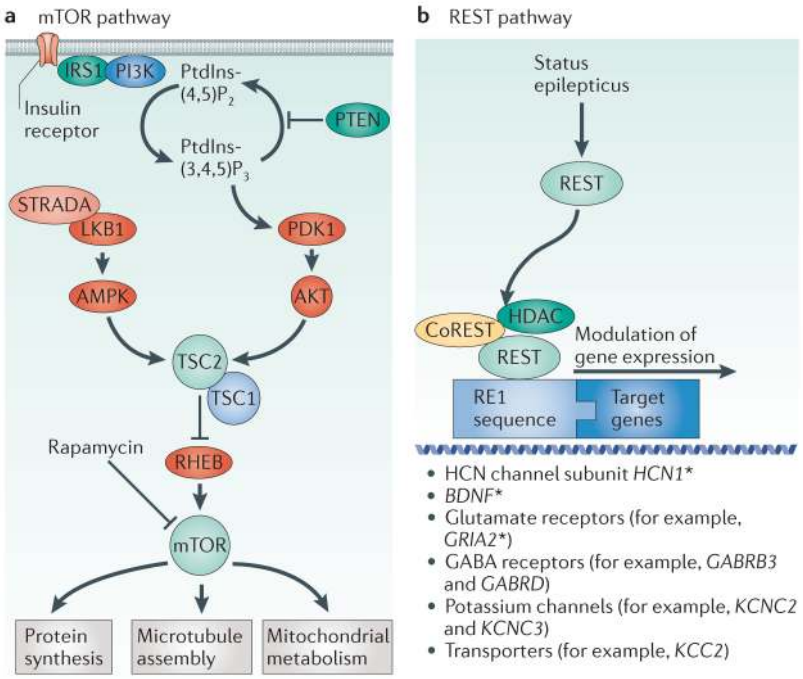


**Figure 1. Mechanistic roles of the mTOR pathway and REST in epileptogenesis**

**a** | A simplified version of the mammalian target of rapamycin (mTOR) pathway, highlighting epilepsy-related molecules. For a more complete illustration of the mTOR pathway, see REF. 12. **b** | A schematic depicting repressor element 1 (RE1)-silencing transcription factor (REST)-mediated gene silencing triggered by status epilepticus. REST co-repressor 1 (CoREST), a component of the histone deacetylase (HDAC) complex, interacts with REST to mediate transcriptional repression. The asterisk indicates genes for which there is specific experimental evidence suggesting regulation by REST or HDAC inhibiton after status epilepticus[49,56]. AMPK, AMP-activated protein kinase; *BDNF*, brain-derived neurotrophic factor; *GABRB3*; type A GABA receptor β3 subunit; *GABRD*; type A GABA receptor δ-subunit; *GRIA2*, ionotropic AMPA 2 glutamate receptor; *HCN1*, hyperpolarization-activated cyclic nucleotide-gated channel subunit 1; IRS1, insulin receptor substrate 1; *KCC2*; potassium/chloride co-transporter 2; *KCNC*, potassium voltage-gated channel subfamily C; LKB1, liver kinase B1 (also known as serine/threonine kinase STK11); mTORC1, mTOR complex 1; PDK1, 3-phosphoinositide-dependent kinase 1; PI3K, phosphoinositide 3-kinase; PtdIns(4,5)$P_2$, phosphatidylinositol-4,5-biphosphate; PtdIns(3,4,5)$P_3$, phosphatidylinositol-3,4,5-triphosphate; PTEN, phosphatase and tensin homologue; RHEB, RAS homologue enriched in brain; STRADA, STE20-related adaptor protein-α; TSC, tuberous sclerosis complex. Part **a** is modified, with permission, from REF. 11 © (2011) Elsevier.

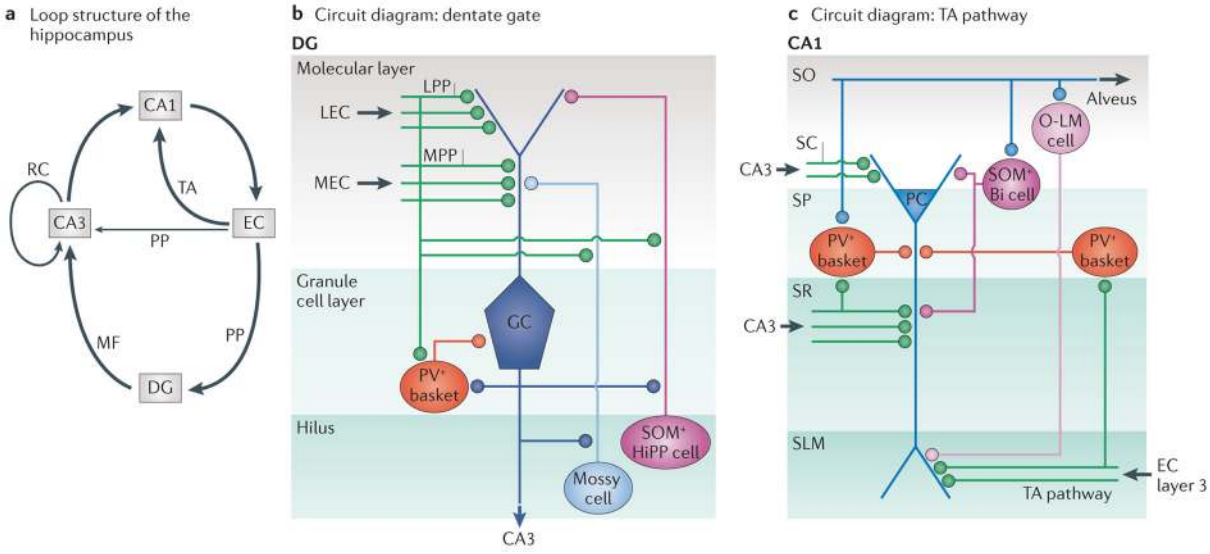


**Figure 2. Embedded loop structures of the temporal lobe**

**a** | The hippocampus receives two inputs, the perforant path (PP) and the temporoammonic (TA) pathway. The entorhinal cortex (EC) is in turn the target of hippocampal output, creating multiple excitatory loops. There is a long loop that originates from the EC and projects to the dentate gyrus (DG) via the PP, from the DG to area CA3 via mossy fibres (MFs), from area CA3 to area CA1 and from area CA1 back to the subiculum (not shown) and/or EC. An intermediate-length loop originates from the EC and projects to area CA3 (via the PP pathway), then to area CA1 and from there to the subiculum and/or EC. A short loop projects from the EC to area CA1 (via the TA pathway) and back to the subiculum and/or EC. Recurrent collaterals (RCs) between CA3 pyramidal cells (PCs) are also shown. **b** | Simplified connectivity within the DG. Many connections and various inhibitory interneuron subtypes are omitted for clarity[58,173,174]. PCs and stellate cells within layer 2 of the medial EC (MEC) and lateral EC (LEC) form the PP to the middle and outer molecular layer, respectively, of the hippocampal DG, as well as to the distal dendrites of PCs in area CA3 (not shown)[173]. In the dentate, lateral PP (LPP) and medial PP (MPP) axons synapse onto dentate granule cells (GCs) and onto GABAergic interneurons (including parvalbumin-positive ($PV^+$) basket cells and somatostatin-positive ($SOM^+$) hilar PP-associated (HiPP) cells) as well as mossy cells in the dentate hilus. **c** | Selected connections of the TA pathway, which proceeds from layer 3 EC pyramidal and spiny stellate neurons (not shown) to the distal dendrites and distal apical tuft dendrites of CA1 neurons in stratum lacunosum-moleculare (SLM) in the short loop mentioned above as well as to the distal dendrites of CA2 (the intermediate-length loop, not shown). Extrinsic excitatory connections are shown in green; excitatory cells and intrinsic excitatory connections are shown in blue; and inhibitory cells and intrinsic inhibitory connections are shown in red and purple. Bi, bistratified cell; O-LM, oriens lacunosum-moleculare. SC, Schaffer collateral; SO, stratum oriens; SR, stratum radiatum.

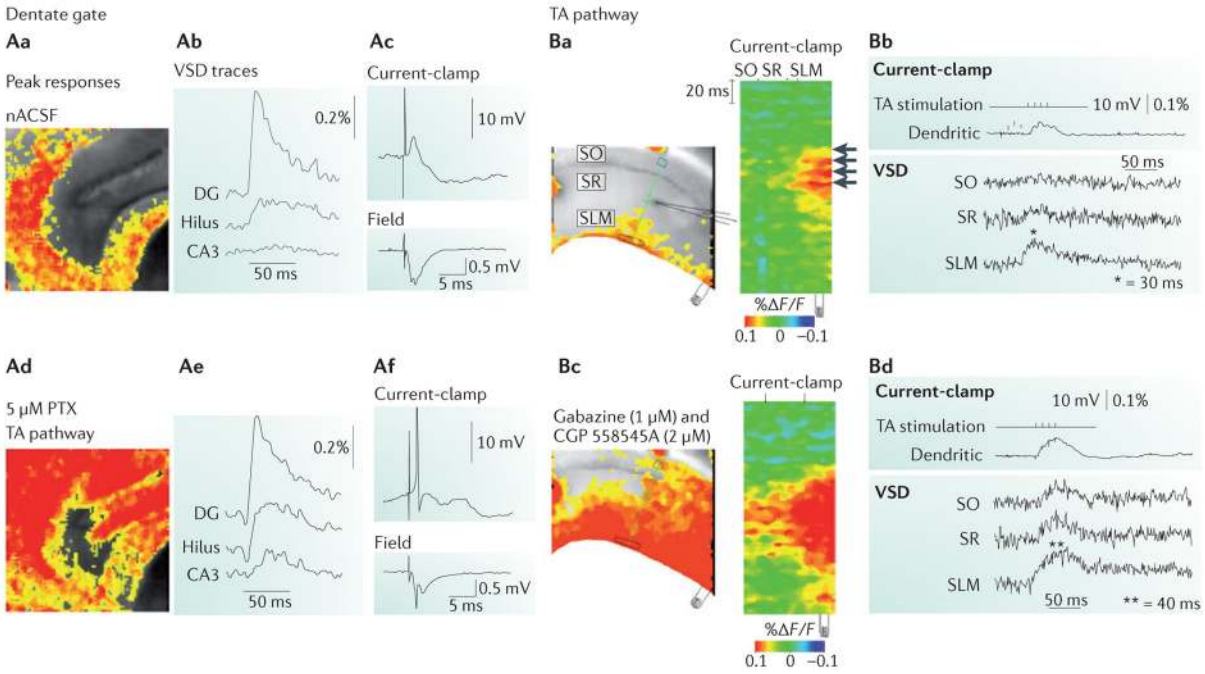


**Figure 3. Temporal lobe circuit elements under normal conditions**

**A |** Dentate gating in the hippocampus under normal conditions (parts **Aa**–**Ac**) and after blockade of GABAergic inhibition (parts **Ad**–**Af**). **Aa |** A voltage-sensitive dye (VSD) image of depolarization that is restricted to the dentate gyrus granule cell layer in response to stimulation of the perforant path in an acute brain slice from the rat hippocampus. **Ab |** The VSD response is quantified in the dentate gyrus, hilus and area CA3. **Ac |** The top panel shows a current-clamp recording from a dentate gyrus granule cell and the botttom panel shows the field potential in the dentate gyrus. **Ad |** The collapse of the dentate after blockade of GABAergic inhibition using 5 μM picrotoxin (PTX; a non-competitive type A ($GABA_A$) receptor antagonist) is illustrated using VSD imaging. **Ae |** The quantification of the VSD response shows collapse of the dentate gate, with activation of upstream area CA3 after GABAergic blockade. **Af |** Blockade of inhibition elicits hyperactivation of dentate granule cells in response to perforant path activation, as shown by the current-clamp recording (top panel) and the field potential (bottom panel). **B |** Temporoammonic (TA) pathway function in area CA1 under normal conditions (parts **Ba**, **Bb**) and following GABAergic blockade (parts **Bc**, **Bd**). **Ba |** Activation within area CA1 in response to stimulation of the TA pathway is spatially restricted to the distal dendrites of CA1 pyramidal cells (indicated by the arrows), which is shown using VSD imaging. **Bb |** The top panel shows a current-clamp recording from a CA1 pyramidal cell dendrite in response to TA pathway activation, and the bottom panel shows the quantification of the VSD response in stratum oriens (SO), stratum radiatum (SR) and stratum lacunosum-moleculare (SLM) 30 ms after extracellular stimulation of the TA pathway, with evoked excitatory responses (indicated by the asterisk) in SLM. **Bc |** A VSD image shows that GABAergic feedforward inhibition after application of the $GABA_A$ antagonist gabazine (1 μm) and the $GABA_B$ antagonist CGP 55845A (2 μm) spatially restricts TA pathway activation to the distal dendrites of CA1 pyramidal cells, with loss of spatial segregation within area CA1. **Bd |** The top panel shows a current-clamp recording from a CA1 pyramidal cell dendrite, and the the bottom panel shows the quantification of the VSD response, with propagation of the response to SR and SO. *F*, fluorescence; nACSF, normal artificial cerebrospinal fluid. Part **A** is reproduced, with

permission, from REF. 137 © (2007) Elsevier. Part **B** is reproduced, with permission, from REF. 154 © (2005) Society for Neuroscience.

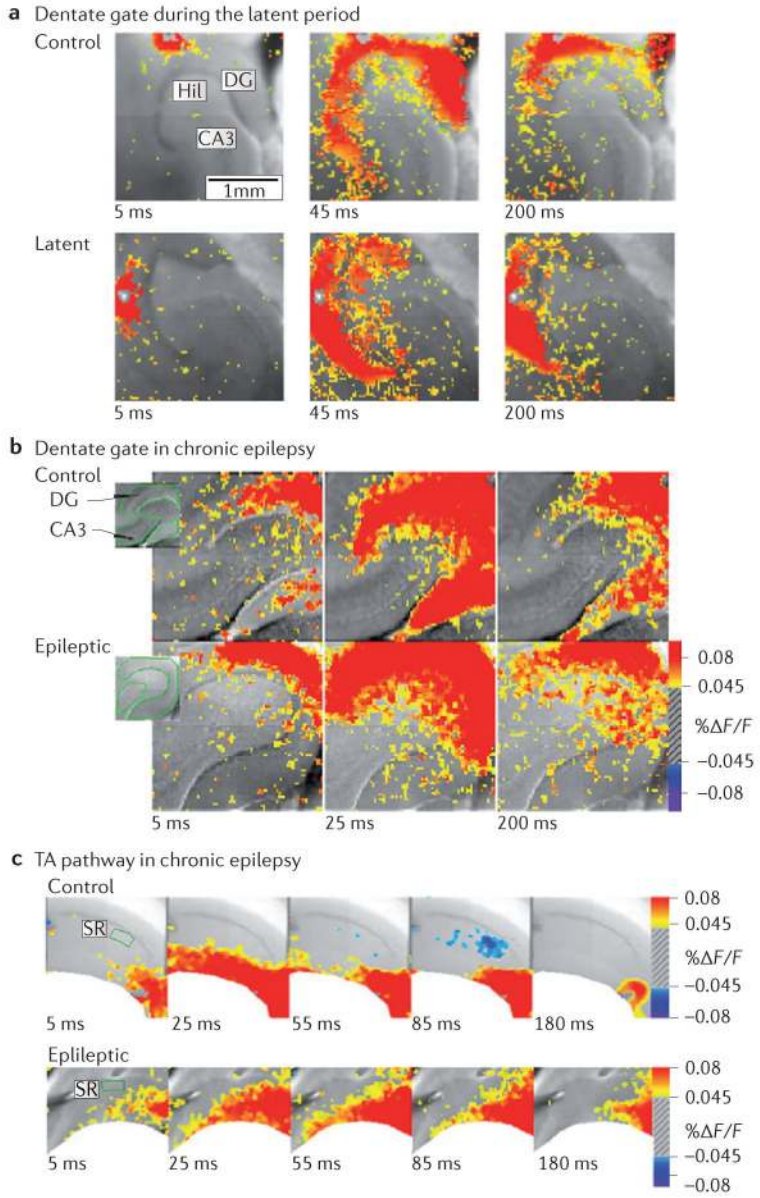


**Figure 4. Circuit dysfunction in temporal lobe epilepsy**

**a** | Dentate gating is impaired during the latent period (1 week after status epilepticus) in an animal model of chronic temporal lobe epilepsy (TLE). The top panel shows voltage-sensitive dye (VSD) recordings of perforant path stimulation under control conditions 5 ms after stimulation (left), at peak (middle) and later (200 ms; right). The top right panel shows restriction of the perforant path-evoked depolarization to the molecular layer of the dentate gyrus (DG). Note that there is minimal area CA3 activation. During the latent period (bottom panel), identical stimulation reveals a delayed peak response as well as >200% increase in activation (as measured by areal pixel activation) in area CA3. **b** | Gating function is retained in the chronic phase of acquired TLE in the rodent model. The two panels show VSD images of the DG 5 ms (left), 25 ms (middle) and 200 ms (right) after stimulation of the perforant path in control rats (top panel) and in chronically epileptic rats (bottom panel). Under both conditions, there is a strong response in the DG that fails to propagate to area CA3. **c** | Temporoammonic (TA) pathway dysfunction in the chronic phase

of acquired TLE. VSD imaging of TA pathway function in control animals (top panel) and epileptic animals (bottom panel) at 5 ms, 25 ms, 55 ms, 85 ms and 180 ms after stimulation of the TA pathway, producing spatially restricted activity in control animals that aberrantly propagates to the strata radiatum and pyramidale in the epileptic condition. Hil, hilus; *F*, fluorescence. Part **a** is reproduced, with permission, from REF. 148 © (2007) Society for Neuroscience. Parts **b** and **c** are reproduced, with permission, from REF. 144 © (2006) Society for Neuroscience.