Epilepsy & Behavior 14 (2009) 16–25

Contents lists available at ScienceDirect

Epilepsy & Behavior

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

Molecular and cellular basis of epileptogenesis in symptomatic epilepsy

Asla Pitkänen [a,b,*], Katarzyna Lukasiuk [c]

[a] A.I. Virtanen Institute for Molecular Sciences, Department of Neurobiology, University of Kuopio, PO Box 1627, FIN-70211 Kuopio, Finland
[b] Department of Neurology, Kuopio University Hospital, Kuopio, Finland
[c] The Nencki Institute of Experimental Biology, Polish Academy of Sciences, Warsaw, Poland

ARTICLE INFO

Article history:
Received 11 August 2008
Revised 18 September 2008
Accepted 18 September 2008
Available online 19 October 2008

Keywords:
Brain
Epilepsy
Gene expression
Epileptogenesis
Microarrays
Posttraumatic epilepsy
Status epilepticus

ABSTRACT

Epileptogenesis refers to a process in which an initial brain-damaging insult triggers a cascade of molecular and cellular changes that eventually lead to the occurrence of spontaneous seizures. Cellular alterations include neurodegeneration, neurogenesis, axonal sprouting, axonal injury, dendritic remodeling, gliosis, invasion of inflammatory cells, angiogenesis, alterations in extracellular matrix, and acquired channelopathies. Large-scale molecular profiling of epileptogenic tissue has provided information about the molecular pathways that can initiate and maintain cellular alterations. Currently we are learning how these pathways contribute to postinjury epileptogenesis and recovery process and whether they could be used as treatment targets.

1. Introduction

Epilepsy is the second most common neurological disorder after stroke [1]. It is estimated that approximately 0.8% of the population is affected by some form of epilepsy. In about 30% of epilepsies, there is an identifiable injury to the brain that triggered the development of epilepsy (symptomatic epilepsies) [2]. Another 30% of patients have "presumed symptomatic epilepsy" (previously called *cryptogenic epilepsy*), in which some brain pathology causing epilepsy is presumed to exist, but has not been identified using current techniques [3]. *Epileptogenesis* refers to the phenomenon in which various kinds of brain insults (e.g., traumatic brain injury, stroke, infection, prolonged febrile seizure) trigger a cascade of events that eventually culminate in the occurrence of spontaneous seizures. An operational definition of epileptogenesis refers to the period between the insult and the occurrence of the first spontaneous seizure (Table 1, Fig. 1).

Our understanding of the epileptogenic process comes largely from animal studies that have investigated status epilepticus (SE)-induced epileptogenesis. The model produces types of alterations similar to those found in resected temporal lobe tissue of patients who have undergone surgery for drug-refractory epilepsy. It has been acknowledged that SE might not be the condition most representative of human epileptogenesis because it is a relatively rare cause of epilepsy in humans, particularly in the adult population, and is often associated with other epileptogenic insults like stroke and traumatic brain injury (TBI). Further, typically the damage produced in animals exceeds that found in humans [4]. Also, data from human surgically operated epileptic tissue represents a relatively small population of patients and, thus, might distort our view of the severity of pathology in most patients with acquired temporal lobe epilepsy (TLE), not even to mention patients with seizure foci in other parts of the brain. These caveats in mind, it is, however, important to acknowledge that these conditions have already provided us with a database of information about molecular and cellular changes in epilepsy that we can use to create testable hypotheses for understanding and discontinuing the epileptogenic process.

2. Molecular basis of cellular alterations in TLE

Bouchet and Cazauvieilh [5] observed that in patients with epilepsy, the hippocampus is hardened and sclerotic. Cellular alterations responsible for sclerosis are multiple and are summarized in Fig. 1.

2.1. Neurodegeneration

Probably the best described of the changes during the epileptogenic process is the neurodegeneration that occurs in the hilus and

* Corresponding author. Address: A.I. Virtanen Institute for Molecular Sciences, Department of Neurobiology, University of Kuopio, PO Box 1627, FIN-70211 Kuopio, Finland. Fax: +358 17 163025.
E-mail address: Asla.Pitkanen@uku.fi (A. Pitkänen).

1525-5050/$ - see front matter © 2008 Elsevier Inc. All rights reserved.
doi:10.1016/j.yebeh.2008.09.023

Table 1
Rat models, in which the initial acquired brain injury leads to the development of spontaneous seizures[a]

Model	Reference
Immature brain	
Hypoxia–ischemia (P7)	
Carotid artery ligation + hypoxia	Williams et al. [126]
Poststroke epilepsy (P12, P25)	
Intrahippocampal endothelin-1	Mátéffyová et al. [127]
Post-SE	
Kainic acid (P14)	Stafström et al. [128]
Li-pilocarpine (P12)	Kubová et al. [129]
Prolonged febrile seizures (P10)	Dube et al. [13]
Mature brain	
Post-SE	
Kainic acid	Nadler et al. [130] Ben-Ari and Lagowska [131]
Pilocarpine	Turski et al. [132]
Li-pilocarpine	Jope et al. [133]
Perforant pathway stimulation	Mazarati et al. [134]
Intrahippocampal stimulation	Lothman et al. [135]
Amygdala stimulation	Nissinen et al. [136]
Post-traumatic epilepsy	
Lateral fluid percussion	D'Ambrosio et al. [137] Kharatishvili et al. [138]
Post-stroke epilepsy	
Cortical photothrombosis	Kelly et al. [139] Karhunen et al. [140]
Cortical endothelin	Karhunen et al. [141]

[a] Most of the currently available molecular and cellular data on the epileptogenic process originate from models in which epileptogenesis is initiated by status epilepticus in adult animals.

CA1 pyramidal cell layer and interneurons, with milder damage in the CA3 and CA2 pyramidal cell layer and granule cells. In addition to the hippocampus, neurodegeneration has also been described in the amygdala and the surrounding entorhinal, perirhinal, and parahippocampal cortices, as well as in many extratemporal areas, including the thalamus and cerebellum [6]. It should be noted that these observations have been made largely in surgically operated temporal lobe tissue, as well as in autopsy tissue from patients with chronic epilepsy of variable etiologies. MRI data, however, suggest that severe hippocampal and cortical atrophy might be present in only a subpopulation of patients at the time of the first seizures [7]. Further, we have very little information on whether patients with different locations of the primary focus or different epileptogenic etiologies present with similar hippocampal and/or extrahippocampal pathology. Currently, the data on whether recurrent seizures contribute to neurodegeneration are controversial. As reviewed by Pitkänen and Sutula [7], there are several experimental and human imaging studies that support the idea that some types of seizures can cause neurodegeneration. Recently, both experimental studies in spontaneous seizure models [8,9] and long-term imaging studies in humans [10,11] have challenged this view.

Neuroprotection has long been believed to be a key in prevention of epileptogenesis. However, as it has been shown, even substantial protection of hippocampal neurons after SE does not prevent epileptogenesis [12]. Further, neurodegeneration is not needed for epileptogenesis, at least in immature brain [13]. However, alleviation of neurodegeneration reduced behavioral consequences after SE [14], and therefore, neuroprotective treatments have a role in enhancement of recovery.

When comparing the molecular profiling data with those obtained using microscopic analysis of neurodegeneration, one can conclude that the information obtained depends on tissue sampling. Sometimes, however, neuronal loss can be predicted after observing downregulation of certain neuronal markers [15].

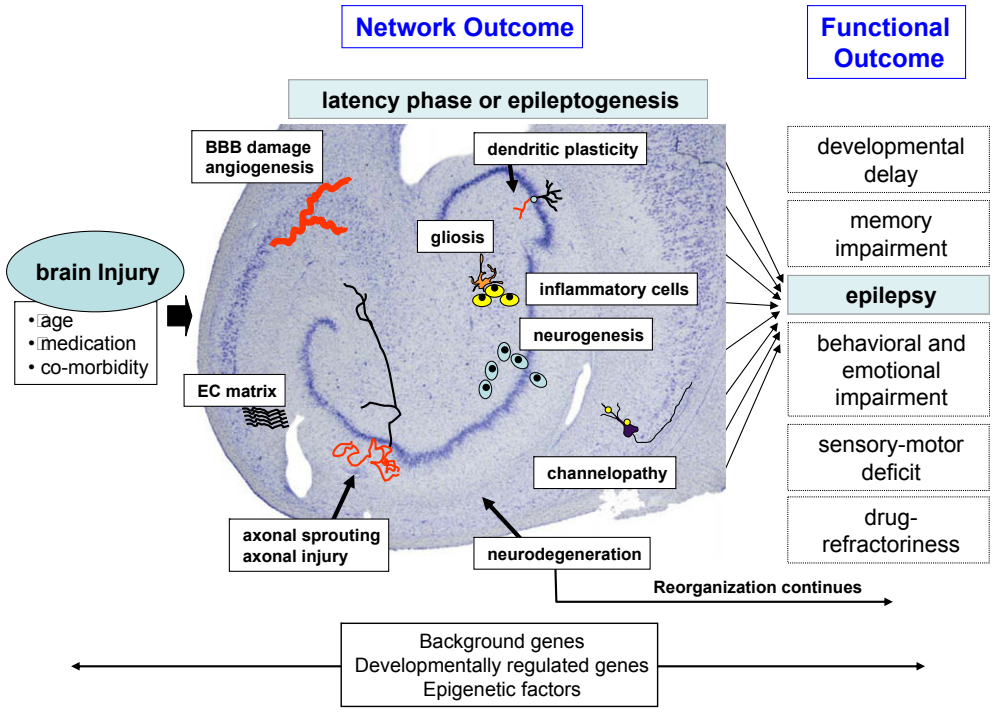

Fig. 1. Cellular alterations occurring during the epileptogenic process. Various types of brain insults such as status epilepticus (SE), traumatic brain injury (TBI), and stroke can trigger the epileptogenic process. Cellular alterations and their temporal distribution are best characterized in the hippocampus, particularly in SE models. These include neurodegeneration, neurogenesis, gliosis, invasion of inflammatory cells, axonal sprouting, axonal injury, dendritic plasticity, angiogenesis, changes in extracellular matrix, and alteration in voltage- and ligand-gated ion channels in individual neurons. These alterations are accompanied by a variety of molecular changes (see Fig. 4). As a consequence, several functional impairments in addition to epilepsy can develop, including developmental delay, memory impairment, emotional impairment, behavioral impairment, somatomotor decline, and drug refractoriness. During the entire epileptogenic process, these alterations are subject to modulation by genetic background, developmentally regulated genetic programs, or epigenetic factors.

Importantly, molecular profiling data extend the histological analysis by providing information about the molecular pathways involved in cell death [16,17].

2.2. Neurogenesis

Occurrence of neurogenesis in human brain was demonstrated by Eriksson and co-workers 10 years ago [18]. Neural precursor cells can be isolated from surgical specimens obtained from patients with intractable TLE and propagated or differentiated into neuronal and glial lineages [19]. Some recent data also suggest that seizures induce neurogenesis in young patients [20]. It is still, however, unclear whether epileptogenic insults or epileptic seizures have an effect on hippocampal neurogenesis in humans [21].

In animals, it has been shown that even brief induced or spontaneous seizures, in addition to SE, can trigger neurogenesis (for review, see [22]). After SE, increased neurogenesis can be detected within a few days and remains elevated for several weeks [23]. In normal animals, newly born neurons receive functional afferent inputs [24] and send functional outputs to postsynaptic neurons [25]. All this occurs within a few months of their birth. In epileptic rodent brain, seizure activity can disturb the migration of newly born neurons, resulting in their ectopic location in the hilus, aberrant connectivity, and, consequently, enhanced excitability [26]. Another line of evidence suggests that newly born cells in an "epileptogenic environment" could actually adapt to the situation and become less sensitive to glutamate and more sensitive to GABA than in the normal hippocampus [27].

It is important to realize that other clinically relevant epileptogenic brain insults such as stroke and TBI can also induce neurogenesis [28,29]. Further, altered neurogenesis has been linked to, for example, learning and memory impairment, as well as depression, which are not uncommon comorbid conditions in patients with epilepsy [30].

Very few studies have investigated the effects of antiepileptic drugs (AEDs) on neurogenesis. Hao et al. [31] showed that chronic treatment with valproate enhances neurogenesis in the dentate gyrus of the normal hippocampus by activating the ERK pathway. Laeng et al. [32] demonstrated that valproate, but not carbamazepine, promotes neurogenesis from embryonic rat cortical stem cells in vitro. A recent study by Jessberger et al. [33] demonstrated that valproic acid blocks seizure-induced neurogenesis in the kainate-induced SE model. Moreover, treatment alleviates hippocampus-dependent learning impairment. Using the in vivo method for measuring hippocampal neurogenesis [incorporation of the stable isotope ^2H into genomic DNA during labeling with ^2H_2O (heavy water)], Shankaran et al. [34] reported enhanced neurogenesis in the hippocampus after exposure to valproate, topiramate, and oxcarbazepine. Carbamazepine, clonazepam, ethosuximide, gabapentin, levetiracetam, phenytoin, primidone, tiagabine, and zonisamide had no effect. Recently, Tung et al. [35] reported that many anesthetics, including isoflurane, propofol, dexmedetomidine, and ketamine, have very little, if any, effect on neural cell proliferation in the adult rat dentate gyrus. This is of interest in that anesthesia is a third-line treatment for SE in humans. Similarly, diazepam, the first-line drug in treatment for SE, was not found to suppress alcohol abstinence-induced hippocampal neurogenesis, even though it effectively suppressed withdrawal symptomatology [36].

In only a few studies have there been therapeutic attempts to regulate neurogenesis and observe whether it affects epileptogenesis. Use of endoneuraminidase (endoN) did not affect epileptogenesis even though it suppressed neurogenesis [37]. The significance of neurogenesis in epileptogenesis has also been challenged in the intrahippocampal kainate model in mouse [38] and in aged rats exposed to kainate [39]. It remains to be explored whether outcomes other than change in seizure threshold could be even more affected by modulation of neurogenesis, and what will be the net outcome of the manipulation of neurogenesis during the epileptogenic process.

Analysis of molecular profiling data of brain tissue following insult frequently reveals changes in expression of genes involved in neurogenesis, neuronal cell differentiation, or maturation [17]. This has been observed in both SE [40,41] and TBI [42] models. Interestingly, for some neurogenesis-related genes, changes in expression can be detected as late as 14 days following the insult [40].

2.3. Gliosis

The four major glial cell types include astrocytes, microglia, oligodendrocytes, and NG2 cells (polydendrocytes). Glial cells can contribute to the epileptogenic process in several ways, including structural support, water and ionic homeostasis (aquaporins), regulation of neurotransmission, inflammatory responses, and neurogenic potential. Most of the data available on glial cells in the epileptogenic process come from astrocytes and microglia.

Astrocytes are presumed to regulate the levels of extracellular potassium, as well as glutamate uptake and synaptic glutamate concentrations, to be active participants in metabolism, and to synthesize various molecules (e.g., proteases) that are needed for the recovery process. Data also point to their role in regulation of synaptic transmission and seizure activity (see [43]). Importantly they also synthesize and release inflammatory mediators during the early postinjury phase, as well as in animals and patients with chronic epilepsy [44]. They can also form a "barrier" that isolates, for example, the cortical lesion from ingrowing axons and blood vessels compromising the self-repair process [45]. A recent finding that some astrocytes acquire stem cell properties after cortical brain injury creates a fascinating scenario that manipulation of the postinjury astrocytic response could be used to generate new neuronal circuits [46]. Whether this would have the potential to favorably modify posttraumatic recovery and epileptogenesis remains to be explored.

Microglia commonly undergo activation in response to the brain insult. Microgliosis can manifest itself as cell proliferation, migration, or secretion of various compounds into the extracellular space. All this is accompanied by characteristic morphological changes. Microglial cells are extremely sensitive to disturbances in brain homeostasis and respond quickly to detected pathology such as damage to neighboring neurons [47]. Increase in the number of microglia and migration toward the place of injury can be detected even in Nissl-stained sections as early as 12–24 h after injury. Microgliosis peaks at about 3–5 days and can remain elevated for several weeks [48,49]. Activated microglia become secretory and release a number of compounds with harmful effects on neurons, such as pro-inflammatory cytokines (interleukin-1, interleukin-6, tumor necrosis factor α) and their receptors, proteases, and nitric oxide [50–53]. On the other hand, activated microglia can secrete transforming growth factor β, brain-derived neurotrophic factor (BDNF), neurotrophin-3, or nerve growth factor, which have a neuroprotective effect and can promote regeneration [54,55]. Another beneficial function of microglia is clearing the dying neurons and glia from the tissue. The net effect of the harmful and protective actions of microglia is controversial. There are some indications that suppression of microglia and the consequent diminishment of its toxic action lead to neuroprotection [56–58]. Whether such neuroprotection can be achieved in models of epilepsy or epileptogenesis and whether this would influence the development or severity of the disease remain to be studied.

Oligodendrocytes are responsible for myelination. Even though white matter damage is often described in human acquired epi-

lepsy [59] and many epileptogenic insults such as TBI are known to damage white matter [60], the contribution of myelin pathology to the epileptogenic process is poorly understood. Whether ongoing seizure activity affects oligodendrocyte function and myelination also needs to be explored. Systematic studies investigating the effects of AEDs on myelin are sparse (see [61]).

NG2 cells, or *polydendrocytes*, are defined as glial cells that express the NG2 proteoglycan and represent a fourth major glial cell population in the mammalian central nervous system. They are morphologically, antigenically, and functionally distinct from mature astrocytes, oligodendrocytes, and microglia. Progenitor cells expressing the proteoglycan NG2 represent approximately 5% of the total cells in the adult brain, and are found in both gray matter and white matter regions, where they give rise to oligodendrocytes. These cells receive synaptic contacts from excitatory and inhibitory neurons, suggesting that they transfer electrical and environmental cues to the myelination process [62]. There are relatively few studies on NG2 cells in epilepsy or seizures. It has, however, been shown that that electroconvulsive seizures induce proliferation of NG2-expressing glial cells both in the rat amygdala and in the hippocampus [63,64].

Molecular profiling data indicate gliosis as one of most pronounced phenomena in the brain following epileptogenic insult. Upregulation of expression of genes coding for markers of glial cells (glial fibrillary acidic protein) and proteins produced by glia (cytokines, chemokines, extracellular matrix proteins) is commonly reported [17].

2.4. Axonal growth

Sprouting of glutamatergic granule cell axons or mossy fibers is the most widely studied form of axonal plasticity in epilepsy. It has been shown in experimental models that many epileptogenic insults trigger mossy fiber sprouting, including SE, TBI, and stroke [65]. This has also been described in the hippocampus of surgical patients with epilepsy of various etiologies [66]. Experimental studies indicate that this sprouting occurs before spontaneous seizures, and is maintained for the lifetime of the epileptic animal [67]. Sprouting occurs also in the axons of pyramidal cells in the CA1 subfield of the rat hippocampus [68], as well as in the human entorhinal cortex [69]. In addition to glutamatergic neurons, GABAergic inhibitory axons have also been shown to sprout in both experimental animal and human TLE [70,71]. Even though one can assume that sprouting is a common response to injury in areas with neurodegeneration, it has not been systematically investigated in other areas of the epileptogenic brain, possibly because of difficulties in detecting sprouting.

The functional role of axonal sprouting in epileptogenesis and ictogenesis is controversial. However, many experimental treatments used to facilitate post-TBI/stroke recovery aim at enhancing axonal sprouting of surviving neurons or providing transplanted cells that form new axonal connections and repair the damaged pathways [72–74]. Recently, Rao et al. [73] suggested that after SE, attraction of mossy fibers that were directed to the inner molecular layer of the dentate gyrus back to the CA3 region using CA3 transplants reduced behavioral seizure frequency for several months [75]. This very interesting observation warrants a video/EEG monitoring study to confirm that the partial seizures are also reduced. Whether enhanced functional improvement (e.g., motor recovery) achieved by new connections is associated with modification of epileptogenesis remains to be explored.

Molecular profiling data reveal changes in the expression of a number of genes involved in axonal plasticity. At early points after insult, changes in the expression of transcription factors (e.g., *Jun*, EGR1, and BHLHB2) that can regulate expression of other plasticity-related genes is observed. Later, changes in the expression of growth factors, components of the synapses, or proteins involved in rearrangement of extracellular matrix are frequently observed [16,17,40,73].

2.5. Axonal injury

Axonal injury is not uncommon after epileptogenic brain insults like TBI and stroke [76]. Histological studies have shown, for example, that after TBI, axonal injury occurs within few minutes to hours, and can continue for up to 1 year (see [29]). Interestingly, no data are available on whether axonal injury contributes to epileptogenesis even though it has significant effect on cognitive and somatomotor outcome [77]. There are some studies suggesting that axonal injury could affect the spread of electrical activity through corpus callosum in traumatized brain [78]. Whether axonal injury affects epileptogenesis and seizure spread (epilepsy phenotype) is unknown.

2.6. Dendritic plasticity

Loss of dendritic spines, changes in spine morphology, and reduced dendritic branching have been described both in experimental TLE models and in persons with TLE [79,80]. It is likely that these alterations could affect the availability of various receptor types as well as their stochiometry and, thus, compromise the information flow from afferent inputs. Ribak and colleagues described the growth of basal dendrites in granule cells of the dentate gyrus during post-SE epileptogenesis [81]. Whether they occur after other epileptogenic insults like TBI and stroke remains to be explored. It has been proposed that basal dendrites could provide an input area to aberrant connectivity being established due to loss of normal targets [82].

2.7. Angiogenesis

Damage to the blood–brain barrier (BBB) and consequent expression of angiogenic factors, proliferation of endothelial cells, and angiogenesis are features common to various epileptogenic insults such as SE, TBI, and stroke, as well as human TLE [83–85], and are most active during the first month after injury. Recent experimental evidence suggests that prolonged seizure activity in animals and humans, as well as the occurrence of brief induced and spontaneous seizures, may be associated with opening of the BBB and angiogenesis [84,86,87]. Disruption of the BBB can trigger epileptiform activity and has been suggested to lead to a vicious cycle that favors ictogenesis [88].

So far, enhancement of angiogenesis has been used to increase post-TBI and poststroke motor recovery. There are no data on whether this would affect epileptogenesis.

Molecular profiling data indicate activation of expression of angiogenic factors after electroconvulsive seizures as well as following status epilepticus [89,90]. Although the activated genes differ between these two conditions, they can participate in overlapping molecular pathways.

2.8. Changes in extracellular matrix

As shown, postinjury epileptogenesis involves a remarkable remodeling of neuronal circuits, including neuronal migration, axonal and dendritic plasticity, and angiogenesis. All these alterations are accompanied by changes in the extracellular matrix (ECM). Only a few families of proteinases have been investigated in epileptogenic tissue (e.g., metalloproteinases, plasminogen activators). Recent molecular profiling studies have pinpointed the regulation of expression of a large number of enzymes contributing to ECM degradation and remodeling. In particular, the plasminogen

system including tissue-type plasminogen activator (tPA), urokinase plasminogen activator (uPA), and their inhibitors TIMP-1 and -2, as well as metalloproteinases, appear to be involved in tissue remodeling following insult [16,17,41].

2.9. Acquired channelopathies

Molecular analysis of ion channels after epileptogenic insults, particularly SE, has revealed that brain injury can result in changes in both ligand-gated and receptor-gated ion channels that are associated with altered function when investigated using single-cell electrophysiology [91–96] (Fig. 2). This phenomenon, called *acquired channelopathy*, has been described in the dendritic, somatic, and axonal channels (Fig. 2). Some of the subunit and functional changes can last even weeks (e.g., [93]) and, thus, can contribute to the establishment of lowered seizure threshold in parallel with other plastic alterations. In the developing brain, channelopathy can occur without any major changes in neuronal networks [13]. Whether this is unique to developing brain remains to be investigated. There is evidence that, for example, changes in $GABA_A$ receptor subunit composition can affect the efficacy of treatment [97]. Whether other channelopathies developing in the early phase of the epileptogenic process contribute to treatment response (e.g., drug refractoriness) remains to be investigated.

Surprisingly, alterations in expression of genes encoding for channels or receptors are rarely detected in molecular profiling experiments, possibly because of the limited sensitivity of the methods used [16,17].

3. Epigenetic mechanisms

The term *epigenetic* refers to changes in genes that occur without directly affecting DNA sequence. This can be achieved by chemical modification of DNA or chromatin, such as DNA methylation and alterations in the methylation or acetylation status of histones. Such modifications strongly influence gene expression and, therefore, are of importance for cell function. There is growing evidence that such phenomena also occur in mature, postmitotic neurons and play a role in psychiatric disorders including addiction, depression, and schizophrenia [98,99] Chromatin modification can be induced by drugs of abuse like cocaine as well as medicines like valproate [99,100]. Even complex stimuli such as maternal care and stress response can be regulated by epigenetic mechanisms [101,102].

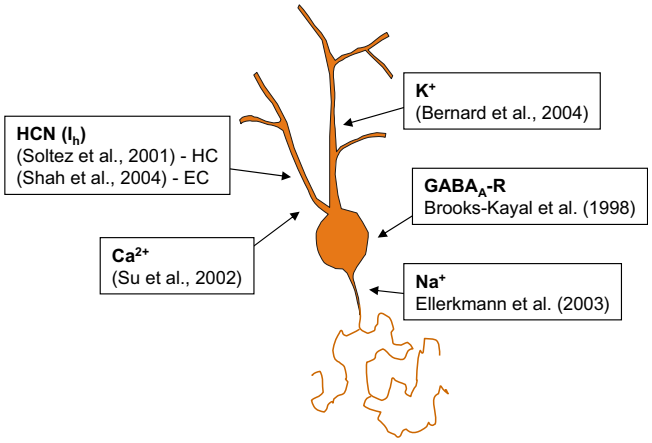

Fig. 2. Acquired channelopathies as investigated by molecular analysis combined with single-cell electrophysiology. As shown, acquired channelopathies can affect dendrites, the perisomatic region, or axons. Whether the same neuron can have multiple channelopathies is not known.

Recent studies have revealed that epigenetic mechanisms may also be relevant to the development of epilepsy and/or seizure control. Seizure-induced [103] or SE-induced [104–106] histone modifications have been reported for promoters of a number of genes, including those involved in neuronal plasticity, such as transcription factors c-fos [103,106], c-jun [107], and CREB [103], the neurotrophin BDNF [103,104], and the glutamate receptor GluR2 [104]. The regulation of transcription by histone modification may be complex. For example, an acute increase in BDNF expression following SE is associated with histone H4 acetylation [103,104], whereas its chronic upregulation may be controlled by acetylation of histone H3 [103]. By influencing the level of growth factor production, the epigenetic mechanism may control the formation of aberrant neuronal connections. At the same time, deacetylation of histones at the GluR2 promoter leads to its decreased expression, which can result in enhanced epileptogenesis [107].

Another clue to the involvement of the epigenetic mechanism in epilepsy was provided by the discovery that the potent antiepileptic drug valproate can act as an inhibitor of histone deacetylases (HDACs) [100] and also can influence gene expression [100,108,109]. Effects of valproate on gene expression could have pronounced consequences for cellular metabolism. It has, for example, been proposed that it can act by inhibition of HDAC-dependent aberrant neurogenesis induced by seizures in the adult hippocampus. Although a recent finding argues against a role for HDAC inhibition in the anticonvulsant activity of valproate [110], it can still be of importance in disease development.

4. Genetic background and focal epilepsy

Investigation of the effect of genetic influence on the epileptogenic process was pioneered by Schauwecker and colleagues

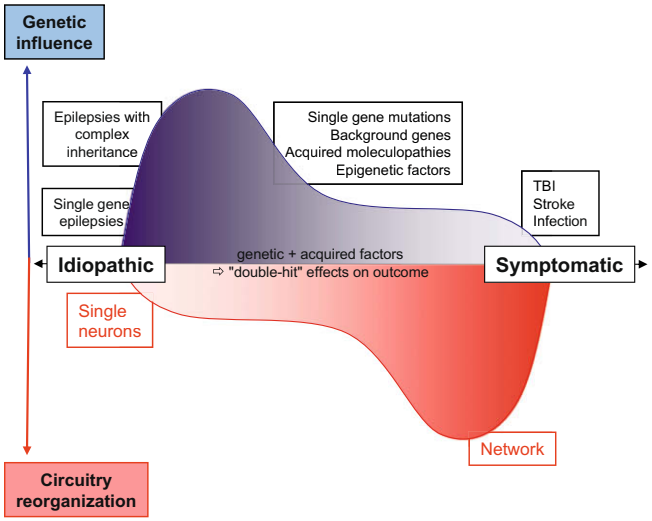

Fig. 3. Combined effects of genetic and acquired factors on epileptogenesis. Highlighted in blue is the effect of genetic influence, which is supposed to be the strongest in idiopathic epilepsies. Highlighted in red is the magnitude of circuitry reorganization, which is most extensive in the acquired symptomatic epilepsies. Between the two extremes are epilepsies, in which the (single) gene mutation(s) can lead to cellular and/or circuitry alterations and brain pathology associated with epilepsy (e.g., by producing proteins that either directly or indirectly affect neuronal excitability and, in parallel, produce disease-specific pathology (Aβ) or by affecting excitability via development of abnormal inhibitory circuitry (uPAR−/−); see text). These mutations can also modify the severity and development of postinjury cellular and/or circuitry alterations and functional outcome (e.g., TNF-α−/−). Background genes can significantly modify the postinjury epileptogenic process. Finally, acquired postinjury changes in gene expression and/or epigenetic factors can modulate and maintain the cellular and circuitry changes associated with the epileptogenic process.

[111]. By investigating the effects of kainate-induced SE on neurodegeneration, axonal plasticity, neurogenesis, and epilepsy phenotype in different mouse strains, they demonstrated the dependence of circuitry alterations on background genes. For example, the "resistant" C57BL/6J mouse shows relatively little neurodegeneration, mossy fiber sprouting, and neurogenesis in the dentate gyrus. These mice also do not develop epilepsy as a consequence of SE. The "sensitive" mouse strains (e.g., FVB/NJ) show circuitry alteration and epileptogenesis. More recently, the background dependence of the aftermath of SE has been demonstrated in the pilocarpine-induced SE model [112,113]. Importantly, the effect of genetic background can, to some extent, be injury type specific. For example, in "resistant" B6 mice, the hippocampal neurodegeneration after controlled cortical injury (TBI) was similar to that in B10 ("sensitive") mice [114]. Even though these studies show that the functional consequences of brain injury can depend on genetic background, more extensive studies are needed on injury-type specificity. Also, there are no studies that have investigated the development of posttraumatic epilepsy in different mouse strains.

5. Single-gene mutations associated with symptomatic epilepsy

Studies in humans have shown that single-gene mutations do not always trigger "generalized" epilepsies, but can also result in focal epilepsies like autosomal-dominant nocturnal frontal lobe epilepsy, familial mesial TLE, familial lateral TLE, and familial partial epilepsy with variable foci (see [115]). Also, data from mice support the idea that single-gene mutations can trigger focal seizures [116]. Importantly, many of the single-gene mutations in epileptic mice are not channelopathies, but encode proteins contributing to cellular migration, transporters, or vesicular proteins. One example is the urokinase-type plasminogen activator receptor (uPAR), which regulates the migration of GABAergic inhibitory neurons in the cortex and the hippocampus during development. Knockout of uPAR results in reduced neuronal numbers in subpopulations of GABAergic neurons (parvalbumin, somatostatin) and the epilepsy phenotype, presumably due to compromised inhibitory circuitry [117]. Another example is the mouse with a mutation that results in encoding of a pathologic protein Aβ (amyloid beta protein) and plaque formation. It was recently shown that this associates with a change in excitability of principal cells and development of epilepsy

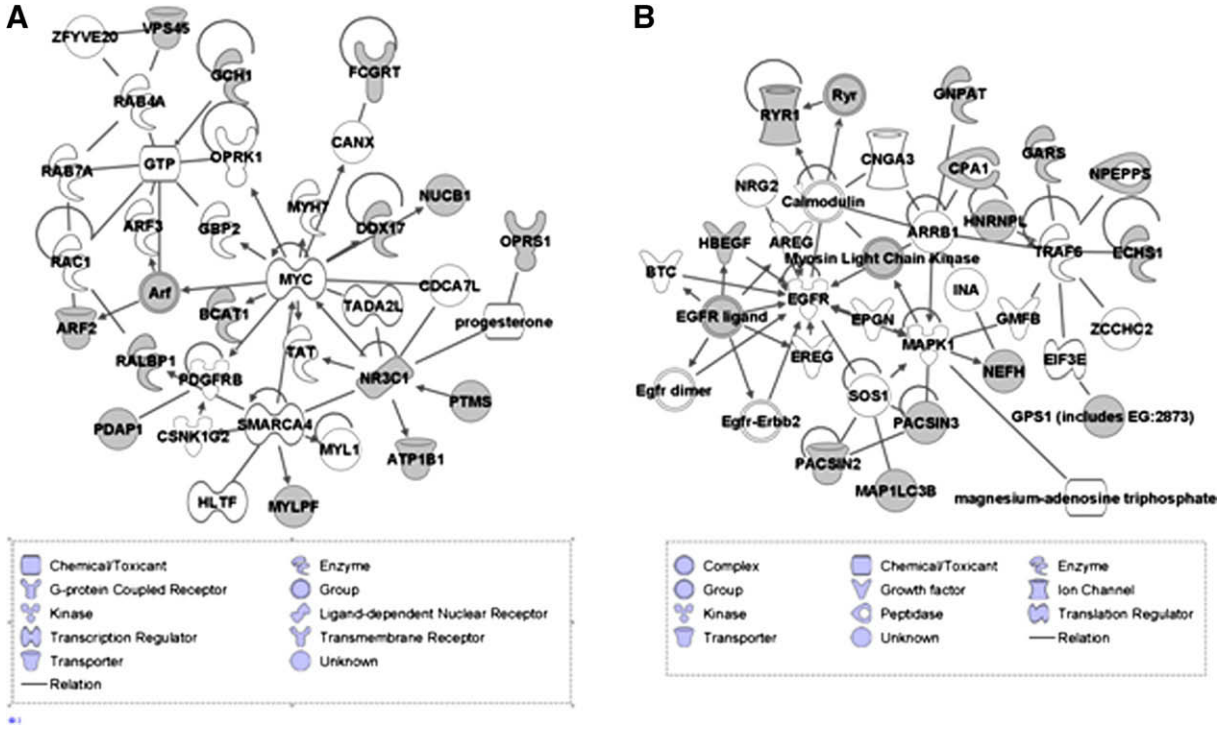

Fig. 4. Examples of molecular networks with a high representation of genes differing in expression level in the hippocampus of epileptic animals as compared with animals undergoing epileptogenesis at 14 days after amygdala stimulation-induced status epilepticus. (A) Cell morphology-related network. Protein products represented in data set of genes with differences in expression levels between epileptogenesis and epilepsy are presented in gray. Only direct relationships between proteins are indicated (line). ARF2, ADP-ribosylation factor 2; ARF3, ADP-ribosylation factor 3; ATP1B1, ATPase, Na^+/K^+ transporting, β1 polypeptide; BCAT1, branched-chain aminotransferase 1; CANX, calnexin; CDCA7L, cell division cycle-associated 7-like; CSNK1G2, casein kinase 1γ2; DDX17, DEAD (Asp–Glu–Ala–Asp) box polypeptide 17; FCGRT, Fc fragment of IgG, receptor α; GBP2, guanylate-binding protein 2, interferon-inducible; GCH1, GTP cyclohydrolase 1; HLTF, helicase-like transcription factor; MYC, v-myc myelocytomatosis viral oncogene homolog; MYH7, myosin heavy chain 7β; MYL1, myosin light chain 1; MYLPF, fast skeletal myosin light chain 2; NR3C1, nuclear receptor subfamily 3, group C, member 1; NUCB1, nucleobindin 1; OPRK1, κ1 opioid receptor; OPRS1, opioid receptor; PDAP1, σ1 PDGFA-associated protein 1; PDGFRB, platelet-derived growth factor receptor, β polypeptide; PTMS, parathymosin; RAB4A, member RAS oncogene family; RAB7A, member RAS oncogene family; RAC1, ras-related C3 botulinum toxin substrate 1; RALBP1, ralA-binding protein 1; SMARCA4, matrix-associated, actin-dependent regulator of chromatin, subfamily a, member 4; TADA2L, transcriptional adaptor 2; TAT, tyrosine aminotransferase; VPS45, vacuolar protein sorting 45 homolog; ZFYVE20, zinc finger, FYVE domain containing 20. (B) Cell cycle-related network. AREG, amphiregulin; ARRB1, arrestin β1; BTC, betacellulin; CaM, calmodulin; CNGA3, cyclic nucleotide-gated channel α3; CPA1, carboxypeptidase A1; ECHS1, enoyl coenzyme A hydratase, short chain, 1; EGFR, epidermal growth factor receptor; EIF3E, eukaryotic translation initiation factor 3, subunit E; EPGN, epithelial mitogen homolog; EREG, epiregulin; GARS, glycyl-tRNA synthetase; GMFB, glia maturation factor β; GNPAT, glyceronephosphate *O*-acyltransferase; GPS1, G-protein pathway suppressor 1; HBEGF, heparin-binding EGF-like growth factor; HNRNPL, heterogeneous nuclear ribonucleoprotein L; INA, internexin neuronal intermediate filament protein α; MAP1LC3B, microtubule-associated protein 1 light chain 3β; MAPK1, mitogen-activated protein kinase 1; NEFH, neurofilament, heavy polypeptide, 200 kDa; NPEPPS, aminopeptidase, puromycin sensitive; NRG2, neuregulin 2; PACSIN2, protein kinase C and casein kinase substrate in neurons 2; PACSIN3, protein kinase C and casein kinase substrate in neurons; RYR1, ryanodine receptor 1; SOS1, son of sevenless homolog 1; TRAF6, TNF receptor-associated factor 6; ZCCHC2, zinc finger, CCHC domain containing 2. IPA, ingenuity pathway analysis, Ingenuity Systems Inc., Redwood City, CA, USA.

[118]. It is important to note that in these mouse epilepsies, the pattern and severity of circuitry reorganization are milder and different from those in the SE-induced epileptogenic process. Whether data from these mouse models imply that a gene mutation resulting in the formation of "an epileptogenic network" (like abnormal inhibitory circuitry) or other brain pathology that is associated with direct or indirect effects of the encoded protein on neuronal excitability is a common concept for epilepsies associated with development or neurodegenerative diseases remains a testable hypothesis (Fig. 3). Both of these mouse models, however, could be considered to model "diseases frequently associated with epileptic seizures" (see [119, Table 6]), that is, a heterogeneous group of epilepsy syndromes, in which the genetic component apparently plays a role. Interestingly, single-gene mutations have also been shown to affect other outcomes associated with the epileptogenic process. For example, knockout of tumor necrosis factor α significantly affected both the severity of TBI-induced neurodegeneration and the consequent motor recovery [120].

6. How much "noise" is there in our modeling and data analysis?

As summarized above, molecular profiling typically shows changes in hundreds of genes depending on the model used, cell type or brain area sampled, time point selected, and analysis platform used. In particular, when using SE models, the cellular alterations are also overwhelming as compared with those in human acquired epilepsy (see [4]). Even though numerous, the gene alterations during epileptogenesis can, however, be linked to known cellular changes. The question arises: Which molecular changes are relevant to epileptogenesis. One way of assessing this would be to look at the genes that are altered in animals that are already epileptic and compare the data with those for animals that are still undergoing epileptogenesis. This analysis revealed differences in gene expression levels between epileptic animals and animals undergoing epileptogenesis (Fig. 4, Table 2). Fourteen days after amygdala stimulation-induced SE, 70 genes were downregulated and 57 upregulated in the hippocampus of epileptic animals as compared with rats undergoing epileptogenesis. In the temporal lobe, the numbers were 7 and 85, respectively. Function is known only for about 70% of these genes, but in many cases the information is limited to sequence-based prediction of function (Table 2). A fraction of these genes can be mapped to known molecular networks (Fig. 4), enabling the formation of hypotheses about their role in epileptogenesis. However, more knowledge about the role of these proteins in cell metabolism, especially in the brain, is

Table 2
Upregulated and downregulated genes in the hippocampus or the temporal lobe in epileptic animals when compared with animals undergoing epileptogenesis at 14 days after amygdala stimulation-induced status epilepticus

Gene function	Gene name
Hippocampus—upregulated	
Enzyme	ATP citrate lyase; arginase I; DEAD box polypeptide; enoyl coenzyme A hydratase; F-box protein 21; glutamate–cysteine ligase; phospholipase D family
Transporter	ATPase β1; solute carrier family 4, member 10; vacuolar protein sorting 45 homolog
Kinase	CDC-like kinase 3; myosin light chain kinase 2; uridine–cytidine kinase 2
Peptidase	Mast cell protease 8; aminopeptidase puromycin sensitive
Transcription regulator	Mediator complex subunit 30
Growth factor	Nerve growth factor
G-protein coupled receptor	Natriuretic peptide receptor B; sigma 1 opioid receptor
Transmembrane receptor	Single immunoglobulin and toll-interleukin 1 receptor
Hippocampus—downregulated	
Transporter	ADP-ribosylation factor 2; α2-macroglobulin; protein kinase C and casein kinase substrate in neurons 2; solute carrier family 6, member 18
Enzyme	Branched-chain aminotransferase 1; cytochrome P450, family 4, subfamily B; cytochrome P450, family 4, subfamily F; defender against cell death 1; glycyl-tRNA synthetase; GTP cyclohydrolase 1; glycerophosphodiester phosphodiesterase 1; glyceronephosphate *O*-acyltransferase; glycogenin 1; lecithin–cholesterol acyltransferase; 5-oxoprolinase; phospholipase A2; ralA-binding protein 1; retinol dehydrogenase 10; steroid sulfatase; thiosulfate sulfotransferase
Peptidase	Carboxypeptidase A1
Transmembrane receptor	Fc fragment of IgG, receptor
Growth factor	Heparin-binding EGF-like growth factor
Ligand-dependent nuclear receptor	Nuclear receptor subfamily 3, group C
Cytokine	Prolactin family 3, subfamily d, member 3
Ion channel	Ryanodine receptor 1; sodium channel, voltage-gated Iβ
Temporal lobe—upregulated	
Peptidase	ADAM 17;
Transporter	ATPase V0 c; LIM and SH3 protein 1; Sec61 α1;
Enzyme	UDP-Gal:βGlcNAc β1,4- galactosyltransferase 3; cytochrome b reductase 1; GNAS complex locus; 3-hydroxy-3-methylglutaryl-coenzyme A reductase; NADH dehydrogenase 1α8; oxoglutarate dehydrogenase; phosphatidylinositol glycan anchor biosynthesis T; RAB40B; retinol dehydrogenase 10; SH3-domain GRB2-like endophilin B1; superoxide dismutase 3; triosephosphate isomerase 1; ubiquitin-conjugating enzyme E2R 2
Growth factor	Bone morphogenetic protein 6;
Kinase	Discoidin domain receptor tyrosine kinase 1; leucine-rich repeat kinase 2
Translation regulator	Eukaryotic translation initiation factor 4A2
Phosphatase	Eyes absent homolog 3
Ion channel	FXYD domain containing ion transport regulator 2; 20 homolog 1
G-protein coupled receptor	Frizzled homolog 6
Transcription regulator	Heat shock factor-binding protein 1; NGFI-A-binding protein 2; ubiquitin-conjugating enzyme E2K
Ion channel	Inositol 1,4,5-triphosphate receptor 1; potassium channel K2
Transmembrane receptor	Opioid-binding protein
Peptidase	Urokinase plasminogen activator
Temporal lobe—downregulated	
Enzyme	Protein disulfide isomerase A6
Transporter	Retinol-binding protein 1; exportin 1

needed before we understand the relevance of the changes in gene expression in injured brain.

Another possibility could be to compare different etiologies associated with hippocampal hyperexcitability but with different degrees of hippocampal cell death and circuitry alterations. Our meta-analysis comparing data on global analyses of gene expression following SE with remarkable hippocampal damage, lateral fluid percussion (relatively mild hippocampal damage), and controlled cortical impact revealed substantial overlap in databases, and indicated a number of genes that commonly change their expression level in response to potentially epileptogenic stimuli [17]. Products of these genes can participate in cell death and survival, neuronal plasticity, or immune response, indicating that these phenomena are crucial for epileptogenesis irrespective of etiology. Although changes in expression of some of the genes in our analysis seem to be etiology related, the data analyzed are not conclusive because of the nature of the data sets available (different microarray platforms and methods of analysis). This would require a well-controlled analysis of transcriptomes collected from tissues of animals (same species, strain, age) that had experienced various epileptogenic insults, and the analysis must be performed using the same platform.

We could also try to investigate what kind of molecular changes are required for the development of a lower seizure threshold with or without cellular damage and circuitry reorganization. One approach would be to compare molecular data obtained in a model with very few, if any, cellular alterations, like a kindling model, and with data from a post-SE model. Such an in-depth comparative study is not available in the literature and cannot be performed on the basis of published data. However, it can be noted that the major functional gene classes modified by kindling are related to second messenger signaling (G-protein signaling, synaptic transmission, Ca^{2+}-dependent kinases) [121], whereas genes related to cell death, apoptosis, or inflammation are barely represented.

Finally, gene expression is under constant change during development. This might be of particular relevance when we consider not only the effects of genetic background, but also the effects of developmentally activated molecular processes, on the development of epilepsy in immature brain (Fig. 1).

7. Conclusion: Where are we and where are we going?

The availability of powerful molecular analysis methods has provided us with a large amount of data that we are just learning to connect to different aspects of the epileptogenic process. An important question remains: How do we optimize animal modeling to reduce "noise" in databases, and, consequently, extract the data that are relevant for understanding the molecular mechanisms of the epileptogenic process, and to find targets for its prevention and modification. Another important aspect is that the same cellular alteration can contribute to various outcomes. For example, axonal sprouting can contribute both to motor recovery and to epileptogenesis. Third, in the design of antiepileptogenic treatment strategies, a key challenge is to answer the question: Is hyperexcitability actually beneficial to the brain's attempts to self-repair? So far, attempts to use AEDs that suppress neuronal activity to prevent epileptogenesis have all failed [122,123]. Should we actually aim at "controlled hyperexcitability" during the epileptogenic process, rather than its suppression, as recent experimental studies using proconvulsants suggest [124,125]?

8. Conflict of interest statement

There are no conflicts of interest.

Acknowledgments

This study was financially supported by the Academy of Finland, the Sigrid Juselius Foundation, and CURE (Citizens United for Research in Epilepsy). Funding organizations had no influence on the contents or on any other aspect of the article.

References

[1] Porter RJ. Classification of epileptic seizures and epileptic syndromes. In: Laidlaw J, Richens A, Chadwick D, editors. A textbook of epilepsy. New York: Churchill Livingstone; 1993.
[2] Hauser WA. Incidence and prevalence. In: Engel Jr J, Pedley TA, editors. Epilepsy: a comprehensive textbook. Philadelphia: Lippincott–Raven; 1997. p. 47–57.
[3] Engel Jr J. Report of the ILAE classification core group. Epilepsia 2006;47:1558–68.
[4] Sloviter RS, Zappone CA, Bumanglag AV, Norwood BA, Kudrimoti H. On the relevance of prolonged convulsive status epilepticus in animals to the etiology and neurobiology of human temporal lobe epilepsy. Epilepsia 2007;48(Suppl 8):6–10.
[5] Bouchet C, Cazauvieilh JB. De l'epilepsie consideree dans ses rapports avec la l'alienation mentale. Arch Gen Med 1825;9:510–42.
[6] Jutila L, Immonen A, Partanen K, et al. Neurobiology of epileptogenesis in the temporal lobe. Adv Tech Stand Neurosurg 2002;27:3–22.
[7] Pitkänen A, Sutula T. Is epilepsy a progressive disease? Prospects for new therapeutic approaches in temporal lobe epilepsy. Lancet Neurol 2002;1:173–81.
[8] Pitkänen A, Nissinen J, Nairismagi J, et al. Progression of neuronal damage after status epilepticus and during spontaneous seizures in a rat model of temporal lobe epilepsy. Prog Brain Res 2002;135:67–83.
[9] Gorter JA, Gonçalves Pereira PM, van Vliet EA, Aronica E, Lopes da Silva FH, Lucassen PJ. Neuronal cell death in a rat model for mesial temporal lobe epilepsy is induced by the initial status epilepticus and not by later repeated spontaneous seizures. Epilepsia 2003;44:647–58.
[10] Liu RS, Lemieux L, Bell GS, et al. The structural consequences of newly diagnosed seizures. Ann Neurol 2002;52:573–80.
[11] Liu RS, Lemieux L, Bell GS, et al. Progressive neocortical damage in epilepsy. Ann Neurol 2003;53:312–24.
[12] Brandt C, Potschka H, Löscher W, Ebert U. *N*-Methyl-D-aspartate receptor blockade after status epilepticus protects against limbic brain damage but not against epilepsy in the kainate model of temporal lobe epilepsy. Neuroscience 2003;118:727–40.
[13] Dubé C, Richichi C, Bender RA, Chung G, Litt B, Baram TZ. Temporal lobe epilepsy after experimental prolonged febrile seizures: prospective analysis. Brain 2006;129(Pt. 4):911–22.
[14] Brandt C, Gastens AM, Sun M, Hausknecht M, Löscher W. Treatment with valproate after status epilepticus: effect on neuronal damage, epileptogenesis, and behavioral alterations in rats. Neuropharmacology 2006;51:789–804.
[15] Becker AJ, Chen J, Paus S, et al. Transcriptional profiling in human epilepsy: expression array and single cell real-time qRT-PCR analysis reveal distinct cellular gene regulation. Neuroreport 2002;13:1327–33.
[16] Lukasiuk K, Pitkanen A. Large-scale analysis of gene expression in epilepsy research: is synthesis already possible? Neurochem Res 2004;29:1169–78.
[17] Lukasiuk K, Dabrowski M, Adach A, Pitkanen A. Epileptogenesis-related genes revisited. Prog Brain Res 2006;158:223–41.
[18] Eriksson PS, Perfilieva E, Björk-Eriksson T, et al. Neurogenesis in the adult human hippocampus. Nat Med 1998;4:1313–7.
[19] Hermann A, Maisel M, Liebau S, et al. Mesodermal cell types induce neurogenesis from adult human hippocampal progenitor cells. J Neurochem 2006;98:629–40.
[20] Blümcke I, Schewe JC, Normann S, Brüstle O, Schramm J, Elger CE, et al. Increase of nestin-immunoreactive neural precursor cells in the dentate gyrus of pediatric patients with early-onset temporal lobe epilepsy. Hippocampus 2001;11:311–21.
[21] Siebzehnrubl FA, Blumcke I. Neurogenesis in the human hippocampus and its relevance to temporal lobe epilepsies. Epilepsia 2008;49(Suppl. 5):55–65.
[22] Parent JM. Adult neurogenesis in the intact and epileptic dentate gyrus. Prog Brain Res 2007;163:529–40.
[23] Parent JM, Yu TW, Leibowitz RT, Geschwind DH, Sloviter RS, Lowenstein DH. Dentate granule cell neurogenesis is increased by seizures and contributes to aberrant network reorganization in the adult rat hippocampus. J Neurosci 1997;17:3727–38.
[24] Van Praag H, Schinder AF, Christie BR, Toni N, Palmer TD, Gage FH. Functional neurogenesis in the adult hippocampus. Nature 2002;415:1030–4.
[25] Toni N, Laplagne DA, Zhao C, et al. Neurons born in the adult dentate gyrus form functional synapses with target cells. Nat Neurosci 2008;11:901–7.
[26] Scharfman HE, Goodman JH, Sollas AL. Granule-like neurons at the hilar/CA3 border after status epilepticus and their synchrony with area CA3 pyramidal cells: functional implications of seizure-induced neurogenesis. J Neurosci 2000;20:6144–58.
[27] Jakubs K, Nanobashvili A, Bonde S, et al. Environment matters: synaptic properties of neurons born in the epileptic adult brain develop to reduce excitability. Neuron 2006;52:1047–59.

[28] Kluska MM, Witte OW, Bolz J, Redecker C. Neurogenesis in the adult dentate gyrus after cortical infarcts: effects of infarct location, *N*-methyl-D-aspartate receptor blockade and anti-inflammatory treatment. Neuroscience 2005;135:723–35.
[29] Pitkänen A, McIntosh TK. Animal models of post-traumatic epilepsy. J Neurotrauma 2006;23:241–61.
[30] Scharfman HE, Hen R. Neuroscience. Is more neurogenesis always better? Science 2007;315:336–8.
[31] Hao Y, Creson T, Zhang L, et al. Mood stabilizer valproate promotes ERK pathway-dependent cortical neuronal growth and neurogenesis. J Neurosci 2004;24:6590–9.
[32] Laeng P, Pitts RL, Lemire AL, et al. The mood stabilizer valproic acid stimulates GABA neurogenesis from rat forebrain stem cells. J Neurochem 2004;91:238–51.
[33] Jessberger S, Nakashima K, Clemenson Jr GD, et al. Epigenetic modulation of seizure-induced neurogenesis and cognitive decline. J Neurosci 2007;27: 5967–75.
[34] Shankaran M, King C, Lee J, Busch R, Wolff M, Hellerstein MK. Discovery of novel hippocampal neurogenic agents by using an in vivo stable isotope labeling technique. J Pharmacol Exp Ther 2006;319: 1172–81.
[35] Tung A, Herrera S, Fornal CA, Jacobs BL. The effect of prolonged anesthesia with isoflurane, propofol, dexmedetomidine, or ketamine on neural cell proliferation in the adult rat. Anesth Analg 2008;106:1772–7.
[36] Nixon K, Crews FT. Temporally specific burst in cell proliferation increases hippocampal neurogenesis in protracted abstinence from alcohol. J Neurosci 2004;24:9714–22.
[37] Pekcec A, Fuest C, Mühlenhoff M, Gerardy-Schahn R, Potschka H. Targeting epileptogenesis-associated induction of neurogenesis by enzymatic depolysialylation of NCAM counteracts spatial learning dysfunction but fails to impact epilepsy development. J Neurochem 2008;105: 389–400.
[38] Kralic JE, Ledergerber DA, Fritschy JM. Disruption of the neurogenic potential of the dentate gyrus in a mouse model of temporal lobe epilepsy with focal seizures. Eur J Neurosci 2005;22:1916–27.
[39] Rao MS, Hattiangady B, Shetty AK. Status epilepticus during old age is not associated with enhanced hippocampal neurogenesis. Hippocampus 2008. [Epub ahead of print].
[40] Elliott RC, Miles MF, Lowenstein DH. Overlapping microarray profiles of dentate gyrus gene expression during development- and epilepsy-associated neurogenesis and axon outgrowth. J Neurosci 2003;23:2218–27.
[41] Lukasiuk K, Kontula L, Pitkänen A. CDNA profiling of epileptogenesis in the rat brain. Eur J Neurosci 2003;17:271–9.
[42] Li HH, Lee SM, Cai Y, Sutton RL, Hovda DA. Differential gene expression in hippocampus following experimental brain trauma reveals distinct features of moderate and severe injuries. J Neurotrauma 2004;21:1141–53.
[43] Wetherington J, Serrano G, Dingledine R. Astrocytes in the epileptic brain. Neuron 2008;58:168–78.
[44] Vezzani A, Baram TZ. New roles for interleukin-1 beta in the mechanisms of epilepsy. Epilepsy Curr 2007;7:45–50.
[45] Itoh T, Satou T, Nishida S, Hashimoto S, Ito H. Immature and mature neurons coexist among glial scars after rat traumatic brain injury. Neurol Res 2007;29:734–42.
[46] Buffo A, Rite I, Tripathi P, et al. Origin and progeny of reactive gliosis: a source of multipotent cells in the injured brain. Proc Natl Acad Sci USA 2008;105:3581–6.
[47] Hailer NP. Immunosuppression after traumatic or ischemic CNS damage: it is neuroprotective and illuminates the role of microglial cells. Prog Neurobiol 2008;84:211–33.
[48] Jørgensen MB, Finsen BR, Jensen MB, Castellano B, Diemer NH, Zimmer J. Microglial and astroglial reactions to ischemic and kainic acid-induced lesions of the adult rat hippocampus. Exp Neurol 1993;120:70–88.
[49] Hailer NP, Grampp A. NitschR. Proliferation of microglia and astrocytes in the dentate gyrus following entorhinal cortex lesion: a quantitative bromodeoxyuridine-labelling study. Eur J Neurosci 1999;11:3359–64.
[50] Benveniste EN. Inflammatory cytokines within the central nervous system: sources, function, and mechanism of action. Am J Physiol 1992;263(1, Pt 1): C1–C16.
[51] Nathan C. Nitric oxide as a secretory product of mammalian cells. FASEB J 1992;6:3051–64.
[52] Kreutzberg GW. Microglia: a sensor for pathological events in the CNS. Trends Neurosci 1996;19:312–8.
[53] Allan SM, Tyrrell PJ, Rothwell NJ. Interleukin-1 and neuronal injury. Nat Rev Immunol 2005;5:629–40.
[54] Elkabes S, DiCicco-Bloom EM, Black IB. Brain microglia/macrophages express neurotrophins that selectively regulate microglial proliferation and function. J Neurosci 1996;16:2508–21.
[55] Miwa T, Furukawa S, Nakajima K, Furukawa Y, Kohsaka S. Lipopolysaccharide enhances synthesis of brain-derived neurotrophic factor in cultured rat microglia. J Neurosci Res 1997;50(6):1023–9.
[56] Tokime T, Nozaki K, Kikuchi H. Neuroprotective effect of FK506, an immunosuppressant, on transient global ischemia in gerbil. Neurosci Lett 1996;206:81–4.
[57] Buki AD, Okonkwo O, Povlishock JT. Postinjury cyclosporin A administration limits axonal damage and disconnection in traumatic brain injury. J Neurotrauma 1999;16:511–21.
[58] Zawadzka M, Kaminska B. A novel mechanism of FK506-mediated neuroprotection: downregulation of cytokine expression in glial cells. Glia 2005;49:36–51.
[59] Lee JW, Andermann F, Dubeau F, et al. Morphometric analysis of the temporal lobe in temporal lobe epilepsy. Epilepsia 1998;39: 727–36.
[60] Gale SD, Johnson SC, Bigler ED, Blatter DD. Nonspecific white matter degeneration following traumatic brain injury. J Int Neuropsychol Soc 1995;1:17–28.
[61] Pitkänen A. Neuroprotection with antiepileptic drugs: state of evidence. In: Trimble M, editor. Seizure freedom: clinical, research and quality of life perspectives. London: Clarion; 2007. p. 9–27.
[62] Kukley M, Kiladze M, Tognatta R, et al. Glial cells are born with synapses. FASEB J 2008;22:2957–69.
[63] Wennström M, Hellsten J, Tingström A. Electroconvulsive seizures induce proliferation of NG2-expressing glial cells in adult rat amygdala. Biol Psychiatry 2004;55:464–71.
[64] Wennström M, Hellsten J, Ekdahl CT, Tingström A. Electroconvulsive seizures induce proliferation of NG2-expressing glial cells in adult rat hippocampus. Biol Psychiatry 2003;54:1015–24.
[65] Pitkänen A, Kharatishvili I, Karhunen H, et al. Epileptogenesis in experimental models. Epilepsia 2007;48(Suppl. 2):13–20.
[66] Mathern GW, Babb TL, Mischel PS, et al. Childhood generalized and mesial temporal epilepsies demonstrate different amounts and patterns of hippocampal neuron loss and mossy fibre synaptic reorganization. Brain 1996;119(Pt. 3):965–87.
[67] Nissinen J, Lukasiuk K, Pitkänen A. Is mossy fiber sprouting present at the time of the first spontaneous seizures in rat experimental temporal lobe epilepsy? Hippocampus 2001;11:299–310.
[68] Cavazos JE, Jones SM, Cross DJ. Sprouting and synaptic reorganization in the subiculum and CA1 region of the hippocampus in acute and chronic models of partial-onset epilepsy. Neuroscience 2004;126: 677–88.
[69] Mikkonen M, Soininen H, Kälviänen R, et al. Remodeling of neuronal circuitries in human temporal lobe epilepsy: increased expression of highly polysialylated neural cell adhesion molecule in the hippocampus and the entorhinal cortex. Ann Neurol 1998;44:923–34.
[70] Davenport CJ, Brown WJ, Babb TL. Sprouting of GABAergic and mossy fiber axons in dentate gyrus following intrahippocampal kainate in the rat. Exp Neurol 1990;109:180–90.
[71] Mathern GW, Babb TL, Leite JP, Pretorius K, Yeoman KM, Kuhlman PA. The pathogenic and progressive features of chronic human hippocampal epilepsy. Epilepsy Res 1996;26:151–61.
[72] Marklund N, Bakshi A, Castelbuono DJ, Conte V, McIntosh TK. Evaluation of pharmacological treatment strategies in traumatic brain injury. Curr Pharm Des 2006;12:1645–80.
[73] Rao MS, Hattiangady B, Shetty AK. Fetal hippocampal CA3 cell grafts enriched with FGF-2 and BDNF exhibit robust long-term survival and integration and suppress aberrant mossy fiber sprouting in the injured middle-aged hippocampus. Neurobiol Dis 2006;21:276–90.
[74] Nudo RJ. Postinfarct cortical plasticity and behavioral recovery. Stroke 2007;38(2, Suppl.):840–5.
[75] Hattiangady B, Rao MS, Shetty AK. Grafting of striatal precursor cells into hippocampus shortly after status epilepticus restrains chronic temporal lobe epilepsy. Exp Neurol 2008;212:468–81.
[76] Coleman MP, Perry VH. Axon pathology in neurological disease: a neglected therapeutic target. Trends Neurosci 2002;25:532–7.
[77] King Jr JT, Carlier PM, Marion DW. Early Glasgow Outcome Scale scores predict long-term functional outcome in patients with severe traumatic brain injury. J Neurotrauma 2005;22:947–54.
[78] Baker AJ, Phan N, Moulton RJ, et al. Attenuation of the electrophysiological function of the corpus callosum after fluid percussion injury in the rat. J Neurotrauma 2002;19:587–99.
[79] Von Campe G, Spencer DD, de Lanerolle NC. Morphology of dentate granule cells in the human epileptogenic hippocampus. Hippocampus 1997;7:472–88.
[80] Isokawa M. Remodeling dendritic spines of dentate granule cells in temporal lobe epilepsy patients and the rat pilocarpine model. Epilepsia 2000;41(Suppl. 6):S14–7.
[81] Spigelman I, Yan XX, Obenaus A, Lee EY, Wasterlain CG, Ribak CE. Dentate granule cells form novel basal dendrites in a rat model of temporal lobe epilepsy. Neuroscience 1998;86:109–20.
[82] Thind KK, Ribak CE, Buckmaster PS. Synaptic input to dentate granule cell basal dendrites in a rat model of temporal lobe epilepsy. J Comp Neurol 2008;509:190–202.
[83] Chopp M, Li Y, Zhang J. Plasticity and remodeling of brain. J Neurol Sci 2008;265:97–101.
[84] Rigau V, Morin M, Rousset MC, et al. Angiogenesis is associated with blood–brain barrier permeability in temporal lobe epilepsy. Brain 2007;130(Pt. 7):1942–56.
[85] Morgan R, Kreipke CW, Roberts G, Bagchi M, Rafols JA. Neovascularization following traumatic brain injury: possible evidence for both angiogenesis and vasculogenesis. Neurol Res 2007;29:375–81.
[86] Hellsten J, West MJ, Arvidsson A, et al. Electroconvulsive seizures induce angiogenesis in adult rat hippocampus. Biol Psychiatry 2005;58: 871–8.

[87] Van Vliet EA, da Costa Araújo S, Redeker S, van Schaik R, Aronica E, Gorter JA. Blood–brain barrier leakage may lead to progression of temporal lobe epilepsy. Brain 2007;130(Pt. 2):521–34.
[88] Seiffert E, Dreier JP, Ivens S, et al. Lasting blood–brain barrier disruption induces epileptic focus in the rat somatosensory cortex. J Neurosci 2004;24:7829–36.
[89] Hunsberger JG, Bennett AH, Selvanayagam E, Duman RS, Newton SS. Gene profiling the response to kainic acid induced seizures. Brain Res Mol Brain Res 2005;141:95–112.
[90] Newton SS, Collier EF, Hunsberger J, et al. Gene profile of electroconvulsive seizures: induction of neurotrophic and angiogenic factors. J Neurosci 2003;23:10841–51.
[91] Brooks-Kayal AR, Shumate MD, Jin H, Rikhter TY, Coulter DA. Selective changes in single cell GABA(A) receptor subunit expression and function in temporal lobe epilepsy. Nat Med 1998;4:1166–72.
[92] Chen K, Aradi I, Thon N, Eghbal-Ahmadi M, Baram TZ, Soltesz I. Persistently modified h-channels after complex febrile seizures convert the seizure-induced enhancement of inhibition to hyperexcitability. Nat Med 2001;7:331–7.
[93] Su H, Sochivko D, Becker A, 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;22:3645–55.
[94] Ellerkmann RK, Remy S, Chen J, et al. Molecular and functional changes in voltage-dependent Na^{+} channels following pilocarpine-induced status epilepticus in rat dentate granule cells. Neuroscience 2003;119:323–33.
[95] Bernard C, Anderson A, Becker A, Poolos NP, Beck H, Johnston D. Acquired dendritic channelopathy in temporal lobe epilepsy. Science 2004;305:532–5.
[96] Shah MM, Anderson AE, Leung V, Lin X, Johnston D. Seizure-induced plasticity of h channels in entorhinal cortical layer III pyramidal neurons. Neuron 2004;44:495–508.
[97] Goodkin HR, Joshi S, Kozhemyakin M, Kapur J. Impact of receptor changes on treatment of status epilepticus. Epilepsia 2007;48(Suppl. 8):14–5.
[98] Tsankova N, Renthal W, Kumar A, Nestler EJ. Epigenetic regulation in psychiatric disorders. Nat Rev Neurosci 2007;8:355–67.
[99] Renthal W, Nestler EJ. Epigenetic mechanisms in drug addiction. Trends Mol Med 2008;14:341–50.
[100] Göttlicher M, Minucci S, Zhu P, et al. Valproic acid defines a novel class of HDAC inhibitors inducing differentiation of transformed cells. EMBO J 2001;20:6969–78.
[101] Weaver IC. Epigenetic programming by maternal behavior and pharmacological intervention. Nature versus nurture: let's call the whole thing off. Epigenetics 2007;2:22–8.
[102] Graff J, Mansuy IM. Epigenetic codes in cognition and behaviour. Behav Brain Res 2008;192:70–87.
[103] Tsankova NM, Kumar A, Nestler EJ. Histone modifications at gene promoter regions in rat hippocampus after acute and chronic electroconvulsive seizures. J Neurosci 2004;24:5603–10.
[104] Huang Y, Doherty J, 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;22:8422–8.
[105] Sng JC, Taniura H, Yoneda Y. Inhibition of histone deacetylation by trichostatin A intensifies the transcriptions of neuronal c-fos and c-jun genes after kainate stimulation. Neurosci Lett 2005;386:150–5.
[106] Sng JC, Taniura H, Yoneda Y. Histone modifications in kainate-induced status epilepticus. Eur J Neurosci 2006;23:1269–82.
[107] Sanchez RM, Koh S, Rio C, et al. Decreased glutamate receptor 2 expression and enhanced epileptogenesis in immature rat hippocampus after perinatal hypoxia-induced seizures. J Neurosci 2001;21:8154–63.
[108] Milutinovic S, D'Alessio AC, Detich N, Szyf M. Valproate induces widespread epigenetic reprogramming which involves demethylation of specific genes. Carcinogenesis 2007;28:560–71.
[109] Phiel CJ, Zhang F, Huang EY, Guenther MG, Lazar MA, Klein PS. Histone deacetylase is a direct target of valproic acid, a potent anticonvulsant, mood stabilizer, and teratogen. J Biol Chem 2001;276:36734–41.
[110] Hoffmann K, Chapp M, Löscher W. Increase in antiepileptic efficacy during prolonged treatment with valproic acid: role of inhibition of histone deacetylases? Epilepsy Res, in press.
[111] Schauwecker PE. Role of genetic influences in animal models of status. Epilepsia 2007;48(Suppl. 8):21–3.
[112] Mohajeri MH, Madani R, Saini K, Lipp HP, Nitsch RM, Wolfer DP. The impact of genetic background on neurodegeneration and behavior in seizured mice. Genes Brain Behav 2004;3:228–39.
[113] Winawer MR, Makarenko N, McCloskey DP, et al. Acute and chronic responses to the convulsant pilocarpine in DBA/2J and A/J mice. Neuroscience 2007;149:465–75.
[114] Witgen BM, Lifshitz J, Grady MS. Inbred mouse strains as a tool to analyze hippocampal neuronal loss after brain injury: a stereological study. J Neurotrauma 2006;23:1320–9.
[115] Andermann F, Kobayashi E, Andermann E. Genetic focal epilepsies: state of the art and paths to the future. Epilepsia 2005;46(Suppl. 10):61–7.
[116] Noebels JL. The biology of epilepsy genes. Annu Rev Neurosci 2003;26: 599–625.
[117] Powell EM, Campbell DB, Stanwood GD, Davis C, Noebels JL, Levitt P. Genetic disruption of cortical interneuron development causes region- and GABA cell type-specific deficits, epilepsy, and behavioral dysfunction. J Neurosci 2003;23:622–31.
[118] Palop JJ, Chin J, Roberson ED, et al. Aberrant excitatory neuronal activity and compensatory remodeling of inhibitory hippocampal circuits in mouse models of Alzheimer's disease. Neuron 2007;55:697–711.
[119] Engel Jr J, for the International League Against Epilepsy (ILAE). A proposed diagnostic scheme for people with epileptic seizures and with epilepsy: report of the ILAE Task Force on Classification and Terminology. Epilepsia 2001;42:796–803.
[120] Scherbel U, Raghupathi R, Nakamura M, et al. Differential acute and chronic responses of tumor necrosis factor-deficient mice to experimental brain injury. Proc Natl Acad Sci USA 1999;96:8721–6.
[121] Gu J, Lynch BA, Anderson D, et al. The antiepileptic drug levetiracetam selectively modifies kindling-induced alterations in gene expression in the temporal lobe of rats. Eur J Neurosci 2004;19:334–45.
[122] Pitkänen A, Kubova H. Antiepileptic drugs in neuroprotection. Expert Opin Pharmacother 2004;5:777–98.
[123] Temkin NR. Antiepileptogenesis and seizure prevention trials with antiepileptic drugs: meta-analysis of controlled trials. Epilepsia 2001;42:515–24.
[124] Pitkänen A, Narkilahti S, Bezvenyuk Z, Haapalinna A, Nissinen J. Atipamezole, an alpha(2)-adrenoceptor antagonist, has disease modifying effects on epileptogenesis in rats. Epilepsy Res 2004;61:119–40.
[125] Chen K, Neu A, Howard AL, et al. Prevention of plasticity of endocannabinoid signaling inhibits persistent limbic hyperexcitability caused by developmental seizures. J Neurosci 2007;27:46–58.
[126] Williams PA, Dou P, Dudek FE. Epilepsy and synaptic reorganization in a perinatal rat model of hypoxia–ischemia. Epilepsia 2004;45:1210–8.
[127] Mátéffyová A, Otáhal J, Tsenov G, Mares P, Kubová H. Intrahippocampal injection of endothelin-1 in immature rats results in neuronal death, development of epilepsy and behavioral abnormalities later in life. Eur J Neurosci 2006;24:351–60.
[128] Stafstrom CE, Thompson JL, Holmes GL. Kainic acid seizures in the developing brain: status epilepticus and spontaneous recurrent seizures. Brain Res Dev Brain Res 1992;65:227–36.
[129] Kubová H, Mares P, Suchomelová L, Brozek G, Druga R, Pitkänen A. Status epilepticus in immature rats leads to behavioural and cognitive impairment and epileptogenesis. Eur J Neurosci 2004;19:3255–65.
[130] Nadler JV, Perry BW, Cotman CW. Intraventricular kainic acid preferentially destroys hippocampal pyramidal cells. Nature 1978;271:676–7.
[131] Ben-Ari Y, Lagowska J. Epileptogenic action of intra-amygdaloid injection of kainic acid. C R Acad Sci Hebd Seances Acad Sci D 1978;287:813–6.
[132] Turski WA, Cavalheiro EA, Schwarz M, Czuczwar SJ, Kleinrok Z, Turski L. Limbic seizures produced by pilocarpine in rats: behavioural, electroencephalographic and neuropathological study. Behav Brain Res 1983;9:315–35.
[133] Jope RS, Morrisett RA, Snead OC. Characterization of lithium potentiation of pilocarpine-induced status epilepticus in rats. Exp Neurol 1986;91:471–80.
[134] Mazarati AM, Wasterlain CG, Sankar R, Shin D. Self-sustaining status epilepticus after brief electrical stimulation of the perforant path. Brain Res 1998;801:251–3.
[135] Lothman EW, Bertram EH, Bekenstein JW, Perlin JB. Self-sustaining limbic status epilepticus induced by 'continuous' hippocampal stimulation: electrographic and behavioral characteristics. Epilepsy Res 1989;3:107–19.
[136] Nissinen J, Halonen T, Koivisto E, Pitkänen A. A new model of chronic temporal lobe epilepsy induced by electrical stimulation of the amygdala in rat. Epilepsy Res 2000;38:177–205.
[137] D'Ambrosio R, Fairbanks JP, Fender JS, Born DE, Doyle DL, Miller JW. Post-traumatic epilepsy following fluid percussion injury in the rat. Brain 2004;127(Pt. 2):304–14.
[138] Kharatishvili I, Nissinen JP, McIntosh TK, Pitkänen A. A model of posttraumatic epilepsy induced by lateral fluid-percussion brain injury in rats. Neuroscience 2006;140:685–97.
[139] Kelly KM, Kharlamov A, Hentosz TM, et al. Photothrombotic brain infarction results in seizure activity in aging Fischer 344 and Sprague Dawley rats. Epilepsy Res 2001;47:189–203.
[140] Karhunen H, Bezvenyuk Z, Nissinen J, Sivenius J, Jolkkonen J, Pitkänen A. Epileptogenesis after cortical photothrombotic brain lesion in rats. Neuroscience 2007;148:314–24.
[141] Karhunen H, Nissinen J, Sivenius J, Jolkkonen J, Pitkänen A. A long-term video-EEG and behavioral follow-up after endothelin-1 induced middle cerebral artery occlusion in rats. Epilepsy Res 2006;72:25–38.