Late-Onset Epileptogenesis and Seizure Genesis: Lessons From Models of Cerebral Ischemia

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Patients surviving ischemic stroke often express delayed epileptic syndromes. Late poststroke seizures occur after a latency period lasting from several months to years after the insult. These seizures might result from ischemia-induced neuronal death and associated morphological and physiological changes that are only partly elucidated. This review summarizes the long-term morphofunctional alterations observed in animal models of both focal and global ischemia that could explain late-onset seizures and epileptogenesis. In particular, this review emphasizes the change in GABAergic and glutamatergic signaling leading to hyperexcitability and seizure genesis. NEUROSCIENTIST 14(1):78–90, 2008. DOI: 10.1177/1073858407301681

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Patients surviving ischemic stroke often express delayed epileptic seizures (Cocito and others 1982; Kilpatrick and others 1992; McNamara 1979; Varelas and Mirski 2001). A distinction is often made between early- and late-onset seizures (Olafsson and others 2000; Reith and others 1997; Sung and Chu 1989). Early-onset seizures occur within the first or second week after stroke, but most of them occur within the first 24 hours (Gupta and others 1988; Lancman and others 1993; Jensen and others 1991). Late poststroke seizures occur after a latency period lasting from several months to years after the insult. These seizures might result from ischemia-induced neuronal death and associated morphological and physiological changes that are only partly elucidated. In half of cases, seizures after stroke have partial onset, including simple partial and secondary generalized seizures. After stroke, the estimated incidence of epilepsy has been reported to vary from 2% to 20% depending on the study design (Bladin and others 2000; Burn and others 1997; Cheung and others 2003; Kotila and Waltimo 1992; So and others 1996). However, in a recent multicenter prospective study, 28% of the patients who experienced at least one poststroke seizure developed epilepsy (i.e., recurrent epileptic seizures). As in posttraumatic epilepsy, a late occurrence of the first seizure represents a higher risk for epilepsy (Willmore 1990). Only a few studies have focused on characterizing and understanding the mechanisms of late-onset epileptogenesis in animal models of ischemia (Kelly and others 2001; Kharlamov and others 2003; Karhunen and others 2003; Luhmann and others 1995; Epsztein and others 2006; Congar and others 2000). A better understanding of these mechanisms is, however, crucial for the treatment of poststroke epilepsy and the design of new therapeutic strategies. In this review, we summarize the long-term morphofunctional alterations observed in animal models of both focal and global ischemia that could explain late-onset seizures and epileptogenesis.

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Animal Models of Cerebral Ischemia

Human ischemic stroke is diverse in its epidemiology, pathophysiology, and clinical manifestations (Molinari 1983). The use and development of relevant animal models are crucial to the study of ischemic brain injury and associated epileptogenic mechanisms. Rodent species are highly suitable for the investigation of cerebral ischemia. Rodent ischemic models can be divided into two major categories: 1) models of transient global ischemia (as occurs in cardiac arrest, coronary artery occlusion, or heart bypass surgery in humans) affecting widespread brain areas but typically giving rise to selective neuronal alterations within vulnerable brain regions (e.g., CA1 in the hippocampus) and 2) models of focal ischemia producing local brain infarction, which represents a model for human acute ischemic stroke (Brierley and others 1971). This section briefly summarizes rodent models of greatest current utility (see also excellent reviews by Ginsberg and Busto 1989; Karhunen and others 2005).

Rat Models of Global Cerebral Ischemia

There are currently two types of the global cerebral ischemic model: the two-vessel occlusion (TVO) model

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and the four-vessel occlusion (FVO) model. In the TVO model, reversible high-grade forebrain ischemia is produced by bilateral common carotid artery occlusions (CCAO) under anesthesia combined with systemic hypotension sufficient to reduce forebrain blood flow markedly. First proposed more than 15 years ago and used to characterize energy state following high-grade incomplete ischemia (Eklof and Siesjö 1972; Nordstrom and others 1978; Nordstrom and Siesjö 1978), this model induces neuronal injury to the hippocampal CA1 and CA4 pyramidal cells and subiculum after as little as 2 minutes of ischemia. The neocortex is then involved within 4 minutes, and injury to the caudoputamen is observed after 8 to 10 minutes of ischemia. Side-to-side differences in the extent of hippocampal lesions can be observed with short periods of ischemia (Smith and others 1984). The two-vessel occlusion model has the advantage of only requiring a one-step surgical procedure with a high survival rate suitable for chronic long-term studies. This model is thus associated with a lower experimental failure rate (from either acute death or an inadequate extent of ischemia) than the four-vessel occlusion model (see below). The principal drawback of the two-vessel occlusion model is the need for anesthetic drugs that might complicate the interpretation of the pathologic outcomes. The variability in the level of induced hypotension also generates interanimal inconsistency with respect to the pathologic outcomes. Finally, this model cannot be used in awake animals, so behavioral alterations immediately after occlusion cannot be assessed.

The FVO model is widely employed to produce high-grade forebrain ischemia in awake, freely moving rats with a high reproducibility in the resulting neuropathology. This model is produced in two stages (Pulsinelli and Buchan 1988). In the first stage, under anesthesia, an atraumatic arterial clamp (Brown and Brierley 1968) is placed around each common carotid artery (CCA) and exteriorized through a ventral midline neck incision. Through a dorsal incision, the alar foramina of the first cervical vertebrae are identified; beneath these foramina pass the two vertebral arteries. A small monopolar electrocautery needle is then inserted through each foramen to electrocoagulate the vertebral arteries. Successful electrocoagulation of the vertebral arteries is essential to the success of this model. Forebrain ischemia is produced the next days, when the animals have recovered from the procedure, by clamping the carotid arteries in the unanesthetized rat. Among rats with previous successful vertebral artery cauterization, the additional bilateral CCA occlusion induces a prompt unconsciousness, attested by the loss of the righting reflex associated with an isoelectric electroencephalogram (EEG) within two to three minutes that persists until recirculation. The lack of critical ischemia in the brainstem is attested by the persistence of spontaneous respiration and preservation of the corneal reflex. Radiotracer studies during ischemia have shown a marked reduction of the local cerebral blood flow (CBF) in the striatum and neocortex (<3% of control) and in the hippocampus (3%–7% of control) (Pulsinelli and others 1982b). Diencephalic and cerebellar CBFs are less severely affected (10%–15% of control), and CBF in the brainstem is maintained at approximately 25% to 30% of control; the latter is probably obligatory if one wants to avert acute death from brainstem ischemia (Pulsinelli and others 1982b). Histopathology has been extensively assessed in this model (Brown and Brierley 1968; Pulsinelli and Brierley 1979; Pulsinelli and Buchan 1988; Yoshida and others 1985; Blomqvist and others 1984; Globus and others 1988; Pulsinelli and others 1982a). Ten to 20 minutes of ischemia are required to observe ischemic cell change in the vulnerable parts of the hippocampus after approximately three days of survival. Consistent neuronal injury to the striatum requires at least 20 to 30 minutes of ischemia, and these changes are maximally expressed by 24 hours. Behavioral studies in the four-vessel occlusion model have shown a permanent impairment of "working" memory and a transient alteration of "reference" memory following 30 minutes of ischemia—deficits probably reflecting hippocampal injury (Volpe and others 1984), as observed in patients after global ischemia (Zola-Morgan and others 1986). To summarize, the four-vessel occlusion model has been solidly validated and is widely employed to produce reversible high-grade incomplete forebrain ischemia. The advantages of this model include applicability to either awake or anesthetized rats.

Rat Models of Focal Cerebral Ischemia

Rat models of middle cerebral artery occlusion (MCAO) have demonstrated their relevance to the human clinical settings of cerebral infarction (Laurent and others 1976; Garcia 1984). The subtemporal approach (Bederson and others 1986; Osborne and others 1987; Shigeno and others 1985) has emerged as a standard method of proximal MCAO and typically produces large cortical and subcortical infarcts ipsilateral to the lesion. In rodents, the hippocampus is likely to be also affected because it is perfused by the posterior cerebral artery, which can be blocked by MCAO. In this model, the lenticulostriate arteries and the small cortical branches must be isolated from both proximal and distal sources of collateral blood supply for consistent infarcts to result. In general, a very close topographic correspondence has been observed between the zone of reduced regional CBF and the area of histological abnormalities (Shigeno and others 1985). Despite its widespread use, MCAO is an invasive and technically demanding procedure, and the size and distribution of the lesions are often variable. Also, animal morbidity and mortality can be high (Kelly 2002). An alternative approach to reversible MCAO makes use of a variant of the photochemical method, in which thrombosis is induced in a middle cerebral artery (MCA) segment by laser illumination following systemic administration of rose Bengal (Shigeno and others 1985; Prado and others 1988; Futrell and others 1988; Watson and others 1985; Nakayama and others 1988). The occlusive thrombus is composed of aggregated platelets and erythrocytes (Nakayama and others 1988; Watson

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and others 1985). When instituted after one hour, the neocortex is largely preserved from infarction, although a mixed pattern of infarction and ischemic cell change is evident in the striatum (Nakayama and others 1988). This method is well characterized, is relatively noninvasive, and produces cortical infarcts within highly reproducible areas in terms of depth and location (Kelly 2002). The MCAO in rats can also be performed by inserting a filament into the lumen of the internal carotid artery that is then advanced beyond the bifurcation of the MCA (Longa and others 1989).

Possible Mechanisms of Late-Onset Epilepsy after Ischemia

Acute Neuronal Death of Principal Cells

The brain is critically dependent on its blood flow for a continuous supply of oxygen and glucose (Siesjö 1978). Only a few minutes of severe global ischemia can induce selective damage to particularly sensitive brain structures. After a stroke insult, a specific pattern of neuronal degeneration is observed in cerebral structures, both in patients and animal models (Crepel and others 1989; Arabadzisz and Freund 1999; Pulsinelli and Brierley 1979; Pulsinelli 1985; Schmidt-Kastner and Freund 1991). For example, in the hippocampus, an ischemic episode induces a delayed selective damage of CA1 pyramidal neurons (two to four days after ischemic episode), whereas CA3 and dentate gyrus neurons are largely resistant (Petito and others 1987; Pulsinelli 1985; Zola-Morgan and others 1992; Fig. 1*A*,*B*). Most of the studies on ischemia have focused on short-term effects (up to one week) (Schmidt-Kastner and Freund 1991). Neuronal cell death is thought to result from acute increased extracellular glutamate concentration, as well as N-methyl-D-aspartate (NMDA) receptor- and calcium-dependent mechanisms (Krantic and others 2005). Among those acute processes, many studies have reported a long-term enhancement of the glutamatergic excitatory drive following energy deprivation that has been primarily called anoxic long-term potentiation (Crepel and others 1993a, 1993b; Crepel and Ben Ari 1996; Hammond and others 1994; Tekkok and Krnjevic 1995; Hsu and Huang 1997, 1998; Crepel and others 2003; Tsintsadze and others 1996; Klishin and others 1995; Urban and others 1990). This pathological long-term potentiation is associated with a marked increase of AMPA and NMDA receptor subunits, including the GluR1, GluR2/3, NR1, and NR2B subunits (Quintana and others 2006; Picconi and others 2006). In addition, it was shown that the anoxia-induced LTP is correlated to a structural remodeling, including the growth of filopodia, enlargements of existing spines, and formation of new spines (Jourdain and others 2002). It is largely suggested that the anoxic LTP that leads to an increase of excitatory drive can play a role in delayed neuronal cell death of principal cells through the activation of calcium-dependent pathways (Andiné and others 1988, 1992; Kirino 2000; Meldrum 2002; Calabresi and others 2003; Papas and others 1993).

Acute Neuronal Death of Different Types of Interneurons

Beside the loss of excitatory principal cells, many studies have shown that GABAergic inhibitory interneurons are strongly affected by an ischemic insult (Mody and others 1995; Neumann-Haefelin and others 1998; Hsu and Buzsaki 1993; Arabadzisz and Freund 1999). Global ischemia induces in the resistant CA3 area of the hippocampus an important reduction in the number of GABAergic interneurons together with a loss of GABAergic terminals within the stratum pyramidale. Unlike pyramidal cells, GABAergic cells form a very heterogeneous population of neurons that can be divided into two main classes according to their axonal projection: somatic and dendritic targeting interneurons. These two classes are known to play different functional roles in the hippocampus (Miles and others 1996). The specific expression of various neuropeptides (such as cholecystokinin or somatostatin) or calcium-binding proteins (such as parvalbumin or calretinin) is also a useful tool to distinguish between different types of interneurons.

An important reduction in the number of cholecystokinin (CCK)–positive interneurons and CCK-positive terminals within the stratum pyramidale from postischemic rats is observed months after ischemia (Epsztein and others 2006; Fig. 1*C*,*D*). This suggests a permanent loss for CCK-positive basket cells that contact the soma of principal cells. In contrast, parvalbumin (PV)–positive basket cells are well preserved (Luhmann and others 1995; Arabadzisz and Freund 1999; Freund and others 1990; Mudrick and Baimbridge 1989; Epsztein and others 2006). Therefore, an important part of the perisomatic inhibition is impaired in the resistant CA3 area months after ischemia. Basket cells play a pivotal role in the generation of fast network oscillations in the hippocampus (Cobb and others 1995; Fisahn and others 1998; Csicsvari and others 2003; Hajos and others 2004). Recently, it has been proposed that this process mainly involves PV basket cells (Freund 2003), whereas CCK basket cells are involved in long-lasting inhibition of principal neurons (Hefft and Jonas 2005) that might contribute to the sparse coding of CA1 pyramidal place cells (Klausberger and others 2005). Thus, the preferential loss of CCK but not PV basket cells after ischemia may result in more selective coding impairments with a good preservation of high-frequency oscillations.

The peridendritic inhibition is also strongly affected by a stroke episode. A dramatic decrease of somatostatin-positive interneurons—which are known to preferentially innervate pyramidal cell dendrites (Freund and Buzsaki 1996; Somogyi and Klausberger 2005)—is also observed in the CA3 area (Epsztein and others 2006; Fig. 1*E*,*F*) and the dentate gyrus after an ischemic episode (Arabadzisz and Freund 1999; Represa and others 1991). The densities of calretinin-positive cell bodies are also significantly reduced after ischemia (Freund and Magloczky 1993; Epsztein and others 2006; Hsu and others 1994). Because most of these neurons innervate other interneurons that are also affected by ischemia

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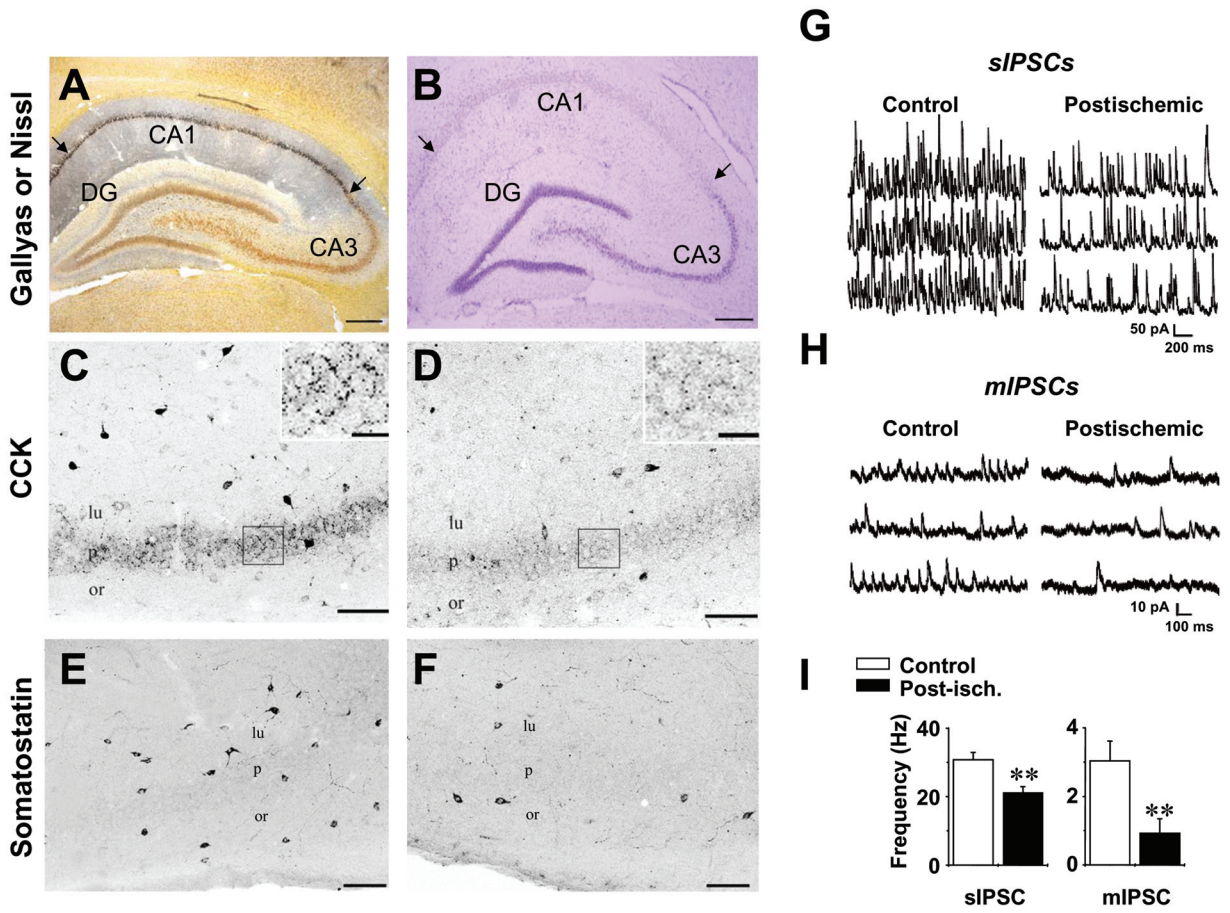

Fig. 1. Loss of interneurons and reduction of the inhibitory drive in the CA3 area of the hippocampus from ischemic rats. Gallyas or Nissl staining of hippocampal sections from rats sacrificed 4 days (Gallyas, *A*) or 10 days (Nissl, *B*) after the ischemic insult. Note that CA3 pyramidal cells do not degenerate after an ischemic episode, in contrast to CA1 pyramidal neurons that are highly sensitive to an ischemic episode, as revealed by the Gallyas and Nissl staining (degeneration of CA1 pyramidal cells shown between arrows). Representative sections from control (*C*, *E*) and ischemic (*D*, *F*) animals stained with cholecystokinin (CCK; *C*, *D*) or somatostatin (*E*, *F*) antibodies performed in the CA3 area of the hippocampus. Immunostaining with CCK antibodies reveals a significant loss of immunopositive cell bodies in CA3 associated with a decrease of immunoreactive puncta around pyramidal cell bodies in the CA3 field of ischemic (*D*) as compared to control (*C*) animals. Somatostatin immunopositive neurons are significantly lost in the CA3 field of ischemic (*F*) as compared to control (*E*) animals. lu, stratum lucidum; p, stratum pyramidale; or, stratum oriens. Scale bars: (*A*, *B*) 600 μm; (*C*–*F*) 100 μm; inset, 25 μm. Spontaneous (*G*) and miniature (*H*) inhibitory postsynaptic currents (IPSCs) respectively recorded in the absence and the presence of tetrodotoxin (TTX; 1 μM) in a control and postischemic CA3 pyramidal cell. Vh: +10 mV. (*I*) Bar graph of the averaged frequency value of spontaneous (*top*) and miniature IPSCs (*bottom*) in control (open bar; n = 12 and 10 for spontaneous and miniature events, respectively) and postischemic (closed bar; n = 13 and 10 for spontaneous and miniature events, respectively) CA3 pyramidal cells (**$P < 0.01$). Note that the frequency of spontaneous and miniature IPSCs is decreased in postischemic cells.

(i.e., other calretinin, somatostatin, and CCK interneurons; Gulyas and others 1996), the functional implications of their loss are not easy to predict. Interestingly, a loss of specific populations of interneurons is also observed in temporal lobe epilepsy, both in patients and in animal models (Buckmaster and Jongen-Relo 1999; Cossart and others 2001; Dinocourt and others 2003; Kobayashi and Buckmaster 2003; Obenaus and others 1993), in different hippocampal fields.

Quantitative and Qualitative Permanent Changes in GABAergic-Mediated Synaptic Transmission in Resistant Areas Several Months after Ischemia

One major issue raised by brain lesions is the fate of surviving neurons and synapses. Several days after the brain injury, an impairment of the GABAergic transmission is observed in the area adjacent to cortical lesions, as well as in remote brain regions (Buchkremer-Ratzmann and others

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1996; Mittmann and others 1994; Neumann-Haefelin and others 1995). These early alterations are likely to be permanent because the amplitude of evoked GABAergic events is permanently reduced in the visual cortex several months after the insult (Luhmann and others 1995). A permanent reduction in the frequency of spontaneous and miniature synaptic GABAergic events has also been reported in resistant CA3 pyramidal cells of the postischemic hippocampus (Epsztein and others 2006; Fig. 1*G–I*). All together, these data indicate that a stroke episode can induce a drastic reduction of the inhibitory drive in the resistant area of the brain. Interestingly, in the postischemic CA3 area of the hippocampus, the remaining inhibitory synaptic events display an important enhancement of their decay time constant (without any significant modification of their rise times and amplitudes) (Epsztein and others 2006).

In control pyramidal cells, many observations have reported the existence of two main kinetic classes of $GABA_A$ synaptic responses in the rat hippocampus (Pearce 1993; Banks and others 1998) that correspond to two anatomically segregated GABAergic synapses (Pearce 1993; Banks and others 1998; Miles and others 1996). $GABA_{A,\ fast}$ synaptic events—which are rapidly activated, rapidly decaying events—are mediated by somatic and proximal dendritic synapses, arising from basket and chandelier cells (Freund and Buzsaki 1996). $GABA_{A,\ slow}$ synaptic events—which are slowly rising and decaying events—are mediated by dendritic synapses (Banks and others 2000). Therefore, in the postischemic pyramidal cells, it can be hypothesized that the change in decay time constant of GABAergic events could be due to a preferential loss of a specific population of interneurons. However, PV-positive interneurons that are mostly preserved by ischemia are known to generate fast decaying events in CA3 pyramidal cells (Bartos and others 2002). Alternatively, the kinetics changes observed could correspond to changes in the subunit composition of $GABA_A$ receptors. $GABA_A$ receptors are composed of five different subunits commonly including tow α, tow β, and either a γ or a δ subunit.

The distribution of specific subtypes is highly brain region and cell type specific and varies during development and in certain disease states. The presence of a specific subunit subtype confers different pharmacological and physiological properties to receptor isoforms. Notably, the decay time constant of GABAergic events is tightly regulated by the subunit composition of $GABA_A$ receptors. In most brain regions, the postsynaptic $GABA_A$ receptors in very young animals incorporate α2, α3, and/or α5 subunits (Poulter and others 1999; Laurie and others 1992; Taketo and Yoshioka 2000; Fritschy and others 1994). These receptors mediate relatively long-lasting inhibitory postsynaptic currents (IPSCs) (Bosman and others 2002; Brussaard and others 1997; Okada and others 2000; Dunning and others 1999). During development, the relative abundance of α2, α3, and α5 subunits diminishes (Fritschy and others 1994; Heinen and others 2004; Laurie and others 1992). Simultaneously, the α1 subunit, which is initially rare, is strongly up-regulated and forms the dominant α subunit in most brain regions during adulthood (Fritschy and others 1994; Heinen and others 2004; Laurie and others 1992; Pirker and others 2000).

Synapses that have mostly α1 subunit–containing $GABA_A$ receptors mediate relatively short-lasting IPSCs (Bosman and others 2002; Goldstein and others 2002; Koksma and others 2003; Vicini and others 2001). Moreover, in adult hippocampal neurons, synapses on axon-initial segments of pyramidal cells—which are presumably formed by CCK-positive cells—still express the α2 subunit (Nusser and others 1996; Nyiri and others 2001). In contrast, synapses formed by PV-positive basket cells on the somata of pyramidal neurons express mainly α1 subunit–containing receptors (Klausberger and others 2002). As a result, altered expression and distribution of certain $GABA_A$ receptor isoforms have the potential to profoundly affect inhibitory neurotransmission. Perturbed expression of $GABA_A$ receptors has been particularly well studied in epilepsy. Several studies in animal models have consistently found an up-regulation of α4-subtype protein expression in animals with experimental epilepsy (Schwarzer and others 1997; Sperk and others 1998; Brooks-Kayal and others 1998). A down-regulation of the α5 subunit has also been reported in the pilocarpine model of temporal lobe of epilepsy (Houser and Esclapez 2003). In the postischemic tissue, it has been shown that the α1 subunit shows an important decrease in the visual cortex one week after the injury (Zepeda and others 2004; but see Liu and others 2002), as well as in the photochemically induced cortical infarct (Neumann-Haefelin and others 1998). In contrast, a significant up-regulation of the α3 subunit has been observed in the contralateral cortex one month after the ischemic insult (Redecker and others 2002). Interestingly, this dysregulation is modulated by the NMDA receptor because application of MK-801 completely blocked this alteration (Redecker and others 2002). Prolongations of miniature synaptic GABAergic events have also been reported in interneurons and pyramidal cells from mice lacking the α1 subunit (Goldstein and others 2002). Therefore, it can be suggested that the change in the decay kinetics of inhibitory events recorded months after ischemia in the resistant CA3 area could result from a change in the subunit composition of $GABA_A$ receptors in surviving pyramidal neurons. This question is of particular interest because GABAergic receptors have different pharmacological properties depending on their subunit composition. For example, studies in genetically engineered mice have shown that the sedative, amnesic, and, in part, anticonvulsive actions of benzodiazepines appear to be mediated by α1 subunit–containing receptors, whereas anxiolytic actions seem to be mediated by receptors that contain α2, α3, or α5 subunits (Sieghart 2000). Therefore, depending on the subunit composition of the GABAergic receptors, different pharmacological agents may be used to support the inhibitory drive in postischemic and epileptic tissue. Finally, because the decay kinetics for GABAergic events in postischemic CA3 pyramidal cells is close to the value reported for neonates'

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pyramidal cells (ca. 30 ms; Taketo and Yoshioka 2000), it can be suggested that this change could reflect a "recapitulation" of an early developmental mechanism, as reported in epileptic tissue for the chloride-extruding K^+-Cl^- cotransporter KCC2 and the GABAergic signaling (Rivera and others 2005; Cohen and others 2002), which could also have a positive effect on recovery after ischemia (Cramer and Chopp 2000).

What could be the functional consequences of a change in the kinetics of GABAergic synaptic events? One can suggest that the prolongation of these events can partly oppose the reduction of the inhibitory drive due to the loss of the subset of GABAergic cells. Moreover, interneurons are involved in the synchronization of synaptic networks (Freund 2003; Traub and others 1996). In vivo, the CA3 region of the hippocampus can generate oscillatory activities in the γ frequency band (Csicsvari and others 2003), and rhythmic inhibitory postsynaptic potentials may be pivotal in phase-locking the oscillatory activity of principal neurons (Cobb and others 1995). Studies performed in vitro and in vivo have shown that prolonging the decay time constant of GABAergic events decreases the oscillation frequency of the CA3 network (Fisahn and others 1998; Traub and others 2000; Khazipov and Holmes 2003). Therefore, the preferential loss of fast GABAergic events in postischemic CA3 neurons would tune the oscillatory behavior of the network toward slowest bands. Because slow oscillations may be more efficient in recruiting synaptic pathways, such a pattern could also encourage the development of synchronized bursts in a large population of neurons (Poulter and others 1999). Accordingly, GABAergic synaptic events decay more slowly in seizure-prone rats overexpressing the "embryonic" α2, α3, and α5 subunits than synaptic events recorded in the control rat expressing mostly the adult α1 subunit (McIntyre and others 2002; Poulter and others 1999).

Axonal Sprouting and Increased Excitation Several Months after Ischemia

Functional map reorganization is often observed in cerebral tissues several months after an insult and lead to a partial recovery of function, as recently exemplified in the visual cortex (Zepeda and others 2004). This phenomenon is thought to involve delayed morphological changes in the neurons that survive the insult, as suggested by the complex changes observed in the level of expression of proteins involved in dendritic and axonal plasticity, such as MAP-2 and GAP-43 (Zepeda and others 2004; Stroemer and others 1993; Miyake and others 2002; Li and others 1998). In the FVO model of global ischemia, an up-regulation of the vesicular glutamate transporter 1 (vGLUT1) is observed in the CA3 region of the hippocampus several months after the insult, both in the stratum oriens and in the stratum lucidum of CA3 (Epsztein and others 2006; Fig. 2*A*,*B*). This is compatible with the sprouting of glutamatergic fibers, as observed in animal models of temporal lobe epilepsy (TLE) (Buckmaster and Dudek 1999; Epsztein and others 2005; Esclapez and others 1999; Okazaki and others 1995; Represa and others 1987; Sutula and others 1988).

The stratum lucidum of the CA3 area is the projection zone of the dentate gyrus granule cells via the mossy fibers. There, mossy fibers contact CA3 pyramidal cells through the large mossy fiber boutons and stratum lucidum interneurons through small en passant boutons or filopodia extensions (Acsady and others 1998). It has been previously shown that the number of mossy fiber filopodia extensions is decreased several months after ischemia (Arabadzisz and Freund 1999). Thus, the increased vGLUT1 staining in the stratum lucidum might correspond to mossy terminals contacting CA3 pyramidal cells. A similar sprouting of mossy fiber synapses has also been observed in the inner molecular layer of the dentate gyrus in some animal models of focal ischemia (Karhunen and others 2005) and in a perinatal model of hypoxia-ischemia (Williams and others 2004). In keeping with a sprouting of mossy fibers, the frequency of miniature synaptic glutamatergic events mediated both by AMPA and kainate receptors—which are specifically generated at mossy fiber synapses (Castillo and others 1997; Cossart and others 2002)—was significantly and permanently increased months after the insult in CA3 pyramidal cells (Epsztein and others 2006; Fig. 2*C*–*E*).

These results are also compatible with the postlesional plasticity of kainate receptor (KAR)–operated synapses following mossy fiber sprouting in animal models of epilepsy (Epsztein and others 2005). The increased vGLUT1 staining in the stratum oriens after global ischemia does not result from the aberrant sprouting of the mossy fibers in the CA3 infrapyramidal band because no change is observed in the trajectory of the mossy fiber pathway in postischemic animals (Arabadzisz and Freund 1999; Epsztein and others 2006). Therefore, this enhancement might be due to the sprouting of other fibers such as associational/commissural glutamatergic fibers that massively project in this area (Sik and others 1993). However, a sprouting of mossy fiber in the infrapyramidal band of the CA3 area has been reported in a neonatal model of hypoxia-ischemia (Williams and others 2004). Similarly, an enhancement of NMDA receptor–mediated synaptic transmission has been observed in neocortical slices 10 to 17 months after global ischemia (Luhmann and others 1995).

All together, these observations show that glutamatergic fibers sprout after global ischemia, resulting in a permanent enhancement of excitatory synaptic transmission mediated by the three main types of ionotropic glutamatergic receptors—namely, AMPA, kainate, and NMDA. These, in addition to the reduced inhibition, result in an imbalance between excitation and inhibition (Luhmann and others 1995; Epsztein and others 2006; Fig. 3*C*) that could have important consequences both for poststroke recovery and epileptogenesis.

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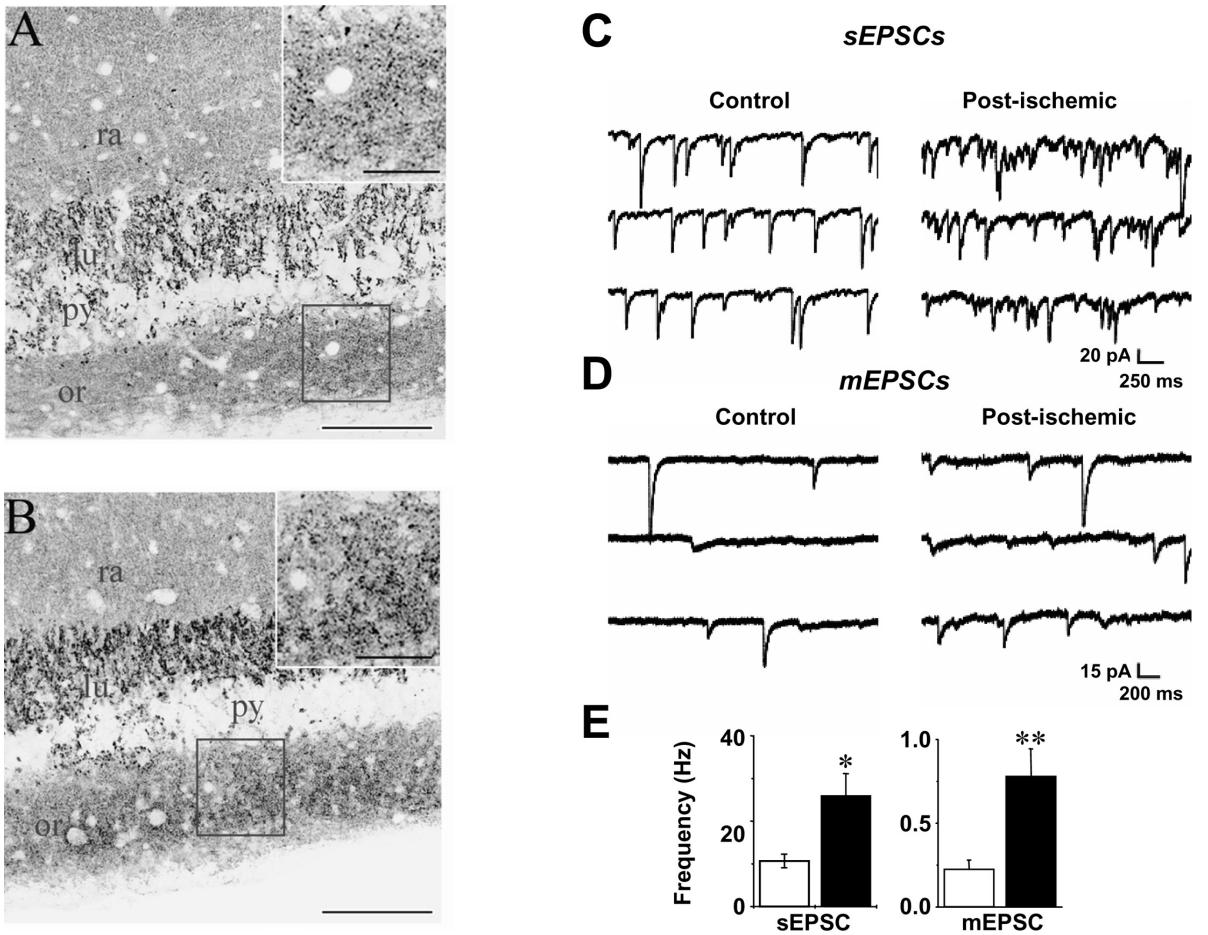

Fig. 2. Enhancement of glutamatergic terminals and excitatory drive in the CA3 area of the hippocampus from ischemic rats. Representative sections from (*A*) control and (*B*) ischemic animals stained with vesicular glutamate transporter 1 (vGLUT1) antibodies performed in the CA3 area of the hippocampus. Note that vGLUT1 immunoreactivity is significantly increased in the CA3 strata lucidum and oriens of postischemic rats. lu, stratum lucidum; or, stratum oriens; py, stratum pyramidale; ra, stratum radiatum. Scale bars: (*A*, *B*) 100 μm; inset, 25 μm. Spontaneous (*C*) and miniature (*D*) excitatory postsynaptic currents (EPSCs) respectively recorded in the absence and presence of tetrodotoxin (TTX; 1 μM), D-APV (50 μM), and bicuculline (10 μM) in a control and postischemic CA3 pyramidal cell. Vh: –70 mV. (*E*) Bar graphs of the averaged frequency values of spontaneous (*left*) and miniature (*right*) EPSCs in control (open bar; n = 11 and 13 for spontaneous and miniature events, respectively) and postischemic (closed bar; n = 11 and 13 for spontaneous and miniature events, respectively) CA3 pyramidal cells. Note that the frequency of both spontaneous and miniature events is significantly increased several months after global ischemia.

Changes in Intrinsic Membrane Properties after Ischemia

The study of the fate of intrinsic membrane properties of pyramidal cells in resistant cortical structures months after global ischemia reveals that there was a clear shift of the resting membrane potential of CA3 pyramidal neurons toward positive values (Congar and others 2000). In contrast, those cells did not display significant changes of the input resistance, spike threshold, spike amplitude, accommodation, fast and slow afterhyperpolarization, and slow afterdepolarization in comparison with control cells (Congar and others 2000). The resting membrane potential of pyramidal neurons is a key factor in the regulation of the cell excitability and mainly depends on the Na^+/K^+-ATPase (Haglund and others 1985; Haglund and Schwartzkroin 1990) and potassium channel activities (for review, see Storm 1990). Down-regulation of the Na^+/K^+-ATPase and potassium channel activities has been described in different models of lesions. For example, a decrease of the Na^+/K^+-ATPase has been reported in the CA1 area several weeks after the lesion of the CA3 pyramidal cells (Anderson and others 1994), and a down-regulation of potassium currents has been observed in motoneurons after axotomy (Gustafsson 1979; Laiwand and others 1988). Therefore, similar phenomena may take place in the partially deefferented postischemic CA3 pyramidal neurons and underlie the depolarization of the resting membrane potential. Further studies are needed to assess whether

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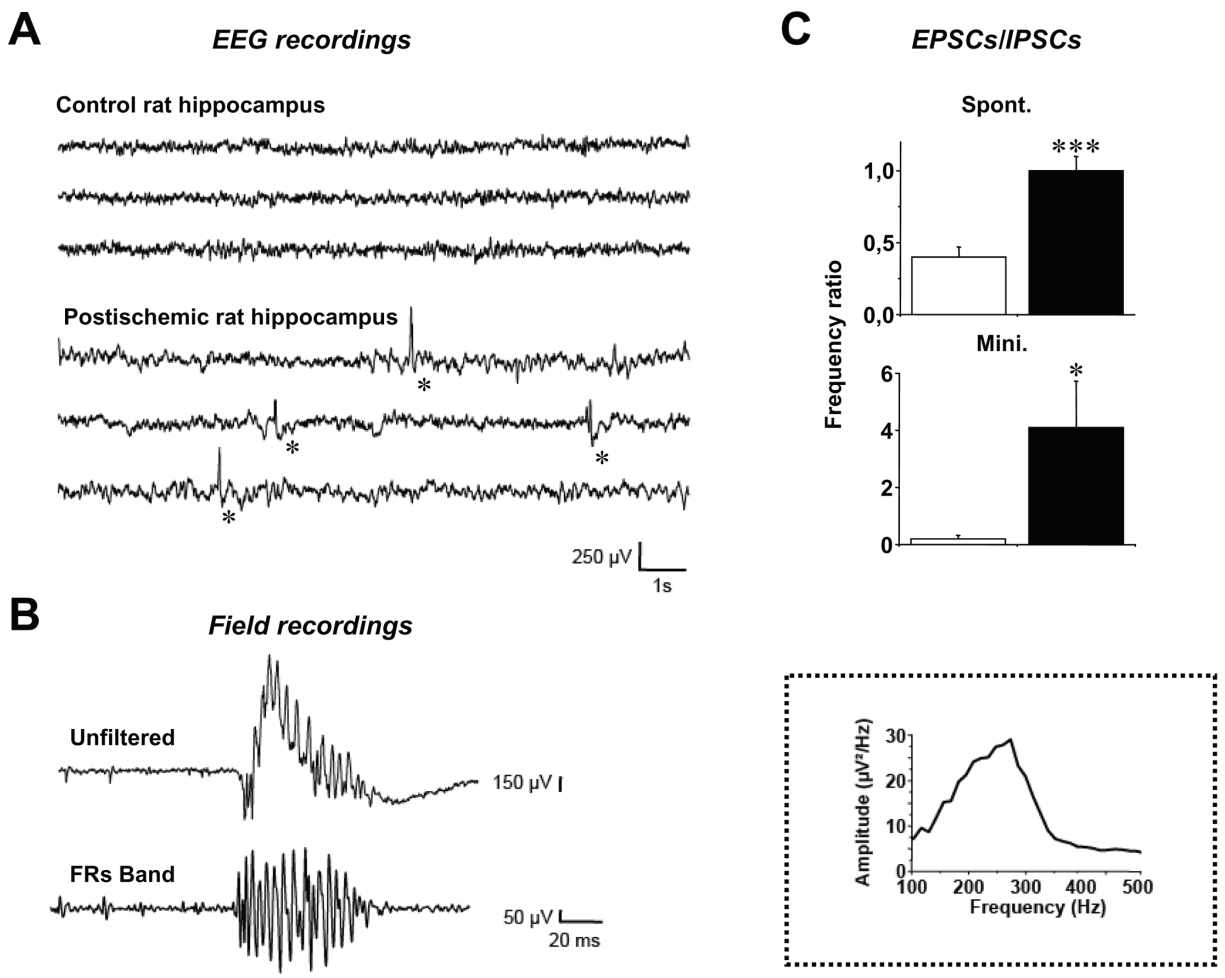

Fig. 3. In vivo and in vitro epileptiform discharges in the hippocampus from the control and postischemic rat. (*A*) Depth electroencephalographic (EEG) recordings of the hippocampus from a control and a postischemic rat (four months after the insult) in awake state. Recordings from the postischemic rat showed recurrent interictal epileptiform discharges (IEDs; indicated by stars, *lower panel*). No IED was recorded in the hippocampus from the age-matched control rat (*upper panel*). (*B*) Example of a population burst extracellularly recorded in the CA3 pyramidal cell layer performed in vitro in slices before (unfiltered; *top*) and after filtering in the fast ripple (FR) band (high-pass 200 Hz; *bottom*). (*Right inset*) Power spectrum of in vitro recorded IED-like activities (n = 791 in seven slices) reveals high-frequency oscillations in the fast ripple band (between 150 and 350 Hz, mean 240 Hz). (*C*) Bar graphs of the averaged values of the frequency ratio of excitatory postsynaptic currents (EPSCs) versus IPSCs for both spontaneous (*top*) and miniature (*bottom*) synaptic activity in control (open bar; n = 8 and 7 for spontaneous and miniature events, respectively) and postischemic (closed bar; n = 8 and 7 for spontaneous and miniature events, respectively) CA3 pyramidal cells (Mann-Whitney, $^{*}P < 0.05$; $^{***}P < 0.001$).

other acquired channelopathies are observed in animal models of both focal and global ischemia as observed in temporal lobe and generalized epilepsy (Remy and Beck 2006; Bernard and others 2004; Poolos 2006).

Ischemic Episode Induces a Permanent Hyperexcitability and Spontaneous Epileptiform Discharges in the Resistant Area in Cortical Structures

Epileptic seizures, recorded using a video-EEG system, have been reported in the rat with focal infarct in the sensory motor cortex (photothrombosis model) (Kelly and others 2001; Kharlamov and others 2003). These consisted of focal epileptic discharges ipsilateral to the cortical infarct, characterized by rhythmic spike-wave discharges (7–9 Hz) lasting from two seconds to more than one minute, associated with motor arrest and occasional mild orofacial twitches. Intermittently, passive wakefulness was characterized by periods (up to a minute or longer) of repetitive generalized solitary spike discharges of moderate to high amplitude with interspike intervals of one to two seconds. During these periods, animals usually demonstrated mild multifocal body jerks but would appear to be undisturbed (Karhunen and others 2005).

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A long-term hyperexcitability has also been reported in animal models of global ischemia ex vivo in neocortical (Luhmann and others 1995) and hippocampal slices (Congar and others 2000), as attested by long-lasting epileptiform responses. More recently, EEG recordings revealed spontaneous interictal epileptiform discharges (IEDs) in the FVO model (Epsztein and others 2006; Fig. 3*A*). Filtering of these IEDs recorded in hippocampal slices revealed the presence of high-frequency oscillations (200–300 Hz), termed *fast ripples* (FRs), whereas only ripple events were observed in slices from control rats (Epsztein and others 2006; Fig. 3*B*). Similar FR activities were specifically observed in the hippocampus from patients with TLE and from chronic epileptic rats (Bragin and others 1999) but not from control rats, in which only ripples were observed (Buzsaki and others 1992). In animal models of TLE, FRs are specifically generated in regions capable of generating seizures (Bragin and others 1999), and their observation is correlated to the subsequent development of epilepsy (Bragin and others 2004). Altogether, these studies suggest that FRs may reflect neuronal mechanisms responsible for epilepsy in TLE. The CA3 recurrent network forms a powerful excitatory circuit that can easily generate IEDs and FRs in conditions of reduced inhibition (Miles and Wong 1987) or enhanced excitation (Dzhala and Staley 2003, 2004). Recently, it was shown that they are generated through an increase in the firing rate of CA3 pyramidal cells, a buildup of excitatory events, and synchronization through the recurrent CA3 excitatory network (Dzhala and Staley 2004; Menendez de la Prida and others 2006). Thus, both the enhanced excitation and decreased inhibition, together with changes in intrinsic membrane properties observed in the CA3 area of the hippocampus several months after global ischemia, could explain the generation of spontaneous IEDs. Because IED- and FR-generating areas are associated with an enhanced susceptibility for seizure generation, these modifications could have profound implications for the occurrence of poststroke epilepsy (Staley and others 2005).

In conclusion, ischemia, in addition to inducing neuronal degeneration that leads to various impairments of brain functions, can also lead to profound network rewiring. Altogether, these mechanisms induce long-term and presumably permanent quantitative and qualitative changes of both glutamatergic and GABAergic synaptic transmission. In-depth understanding of these mechanisms is essential to prevent both poststroke late-onset seizures and epileptogenesis. This could also have important consequences in terms of functional recovery after ischemia because ictal epileptiform discharges as well as the nonperiodic long-term "stable" interictal epileptiform bursts have an important cognitive impact (Aldenkamp and Arends 2004; Karhunen and others 2005).

References

Acsady L, Kamondi A, Sik A, Freund T, Buzsaki G. 1998. GABAergic cells are the major postsynaptic targets of mossy fibers in the rat hippocampus. J Neurosci 18:3386–403.

Aldenkamp AP, Arends J. 2004. Effects of epileptiform EEG discharges on cognitive function: is the concept of "transient cognitive impairment" still valid? Epilepsy Behav 5(suppl 1):S25–34.

Anderson WR, Franck JE, Stahl WL, Maki AA. 1994. Na,K-ATPase is decreased in hippocampus of kainate-lesioned rats. Epilepsy Res 17:221–31.

Andiné P, Jacobson I, Hagberg H. 1988. Calcium uptake evoked by electrical stimulation is enhanced postischemically and precedes delayed neuronaldeath in CA1 of the rat hippocampus: involvement of the N-methyl-D-aspartate receptors. J Cerebr Blood Flow Metab 8:799–807.

Andiné P, Jacobson I, Hagberg H. 1992. Enhanced calcium uptake by CA1 pyramidal cell dendrites in the postischemic phase despite subnormal evoked field potentials: excitatory amino acid receptor dependency and relationship to neuronal damage. J Cereb Blood Flow 12:773–83.

Arabadzisz D, Freund TF. 1999. Changes in excitatory and inhibitory circuits of the rat hippocampus 12-14 months after complete forebrain ischemia. Neuroscience 92:27–45.

Banks MI, Li TB, Pearce RA. 1998. The synaptic basis of GABAA,slow. J Neurosci 18:1305–17.

Banks MI, White JA, Pearce RA. 2000. Interactions between distinct GABA(A) circuits in hippocampus. Neuron 25:449–57.

Bartos M, Vida I, Frotscher M, Meyer A, Monyer H, Geiger JR, Jonas P. 2002. Fast synaptic inhibition promotes synchronized gamma oscillations in hippocampal interneuron networks. Proc Natl Acad Sci USA 99:13222–7.

Bederson JB, Pitts LH, Tsuji M, Nishimura MC, Davis RL, Bartkowski H. 1986. Rat middle cerebral artery occlusion: evaluation of the model and development of a neurologic examination. Stroke 17:472–6.

Bernard C, Anderson A, Becker A, Poolos NP, Beck H, Johnston D. 2004. Acquired dendritic channelopathy in temporal lobe epilepsy. Science 305:532–5.

Bladin CF, Alexandrov AV, Bellavance A, Bornstein N, Chambers B, Cote R, and others. 2000. Seizures after stroke: a prospective multicenter study. Arch Neurol 57:1617–22.

Blomqvist P, Mabe H, Ingvar M, Siesjö BK. 1984. Models for studying long-term recovery following forebrain ischemia in the rat: 1. Circulatory and functional effects of 4-vessel occlusion. Acta Neurol Scand 69:376–84.

Bosman LW, Rosahl TW, Brussaard AB. 2002. Neonatal development of the rat visual cortex: synaptic function of GABAA receptor alpha subunits. J Physiol 545:169–81.

Bragin A, Engel J Jr, Wilson CL, Fried I, Mathern GW. 1999. Hippocampal and entorhinal cortex high-frequency oscillations (100–500 Hz) in human epileptic brain and in kainic acid–treated rats with chronic seizures. Epilepsia 40:127–37.

Bragin A, Wilson CL, Almajano J, Mody I, Engel J Jr. 2004. High-frequency oscillations after status epilepticus: epileptogenesis and seizure genesis. Epilepsia 45:1017–23.

Brierley JB, Graham DI, Adams JH, Simpsom JA. 1971. Neocortical death after cardiac arrest: a clinical, neurophysiological, and neuropathological report of two cases. Lancet 2:560–5.

Brooks-Kayal AR, Shumate MD, Jin H, Rikhter TY, Coulter DA. 1998. Selective changes in single cell GABA(A) receptor subunit expression and function in temporal lobe epilepsy. Nat Med 4:1166–72.

Brown AW, Brierley JB. 1968. The nature, distribution and earliest stages of anoxic-ischaemic nerve cell damage in the rat brain as defined by the optical microscope. Br J Exp Pathol 49:87–106.

Brussaard AB, Kits KS, Baker RE, Willems WP, Leyting-Vermeulen JW, Voorn P, and others. 1997. Plasticity in fast synaptic inhibition of adult oxytocin neurons caused by switch in GABA(A) receptor subunit expression. Neuron 19:1103–14.

Buchkremer-Ratzmann I, August M, Hagemann G, Witte OW. 1996. Electrophysiological transcortical diaschisis after cortical photothrombosis in rat brain. Stroke 27:1105–9.

Buckmaster PS, Dudek FE. 1999. In vivo intracellular analysis of granule cell axon reorganization in epileptic rats. J Neurophysiol 81:712–21.

Buckmaster PS, Jongen-Relo AL. 1999. Highly specific neuron loss preserves lateral inhibitory circuits in the dentate gyrus of kainate-induced epileptic rats. J Neurosci 19:9519–29.

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Burn J, Dennis M, Bamford J, Sandercock P, Wade D, Warlow C. 1997. Epileptic seizures after a first stroke: the Oxfordshire Community Stroke Project. BMJ 315:1582–7.
Buzsaki G, Horvath Z, Urioste R, Hetke J, Wise K. 1992. High-frequency network oscillation in the hippocampus. Science 256:1025–7.
Calabresi P, Centonze D, Pisani A, Cupini L, Bernardi G. 2003. Synaptic plasticity in the ischaemic brain. Lancet Neurol 2:622–9.
Castillo PE, Malenka RC, Nicoll RA. 1997. Kainate receptors mediate a slow postsynaptic current in hippocampal CA3 neurons. Nature 388:182–6.
Cheung CM, Tsoi TH, Au-Yeung M, Tang AS. 2003. Epileptic seizure after stroke in Chinese patients. J Neurol 250:839–43.
Cobb SR, Buhl EH, Halasy K, Paulsen O, Somogyi P. 1995. Synchronization of neuronal activity in hippocampus by individual GABAergic interneurons. Nature 378:75–8.
Cocito L, Favale E, Reni L. 1982. Epileptic seizures in cerebral arterial occlusive disease. Stroke 13:189–95.
Cohen I, Navarro V, Clemenceau S, Baulac M, Miles R. 2002. On the origin of interictal activity in human temporal lobe epilepsy in vitro. Science 298:1418–21.
Congar P, Gaiarsa JL, Popovici T, Ben Ari Y, Crepel V. 2000. Permanent reduction of seizure threshold in post-ischemic CA3 pyramidal neurons. J Neurophysiol 83:2040–6.
Cossart R, Dinocourt C, Hirsch JC, Merchan-Perez A, De Felipe J, Esclapez M, and others. 2001. Dendritic but not somatic GABAergic inhibition is decreased in experimental epilepsy. Nat Neurosci 4:52–62.
Cossart R, Epsztein J, Tyzio R, Becq H, Hirsch J, Ben Ari Y, and others. 2002. Quantal release of glutamate generates pure kainate and mixed AMPA/kainate EPSCs in hippocampal neurons. Neuron 35:147–59.
Cramer SC, Chopp M. 2000. Recovery recapitulates ontogeny. Trends Neurosci 23:265–71.
Crepel V, Ben Ari Y. 1996. Intracellular injection of a Ca2+ chelator prevents generation of anoxic LTP. J Neurophysiol 75:770–9.
Crepel V, Epsztein J, Ben Ari Y. 2003. Ischemia induces short- and long-term remodeling of synaptic activity in the hippocampus. J Cell Mol Med 7:401–7.
Crepel V, Hammond C, Chinestra P, Diabira D, Ben-Ari Y. 1993a. A selective LTP of NMDA receptor-mediated currents induced by anoxia in CA1 hippocampal neurons. J Neurophysiol 70:2045–55.
Crepel V, Hammond C, Krnjevic K, Chinestra P, Ben-Ari Y. 1993b. Anoxia-induced LTP of isolated NMDA receptor-mediated synaptic responses. J Neurophysiol 69:1774–8.
Crepel V, Represa A, Beaudoin M, Ben-Ari Y. 1989. Hippocampal damage induced by ischemia and intra-amygdaloid kainate injection: effects on N-methyl-D-aspartate, (N-(1-[2-thienyl]cyclohexyl)piperidine and glycine binding sites. Neuroscience 31:605–12.
Csicsvari J, Jamieson B, Wise KD, Buzsaki G. 2003. Mechanisms of gamma oscillations in the hippocampus of the behaving rat. Neuron 37:311–22.
Dinocourt C, Petanjek Z, Freund TF, Ben Ari Y, Esclapez M. 2003. Loss of interneurons innervating pyramidal cell dendrites and axon initial segments in the CA1 region of the hippocampus following pilocarpine-induced seizures. J Comp Neurol 459:407–25.
Dunning DD, Hoover CL, Soltesz I, Smith MA, O'Dowd DK. 1999. GABA(A) receptor-mediated miniature postsynaptic currents and alpha-subunit expression in developing cortical neurons. J Neurophysiol 82:3286–97.
Dzhala VI, Staley KJ. 2003. Transition from interictal to ictal activity in limbic networks in vitro. J Neurosci 23:7873–80.
Dzhala VI, Staley KJ. 2004. Mechanisms of fast ripples in the hippocampus. J Neurosci 24:8896–906.
Eklof B, Siesjö BK. 1972. The effect of bilateral carotid artery ligation upon the blood flow and the energy state of the rat brain. Acta Physiol Scand 86:155–65.
Epsztein J, Milh M, Bihi RI, Jorquera I, Ben-Ari Y, Represa A, and others. 2006. Ongoing epileptiform activity in the post-ischemic hippocampus is associated with a permanent shift of the excitatory-inhibitory synaptic balance in CA3 pyramidal neurons. J Neurosci 26:7082–92.
Epsztein J, Represa A, Jorquera I, Ben Ari Y, Crepel V. 2005. Recurrent mossy fibers establish aberrant kainate receptor-operated synapses on granule cells from epileptic rats. J Neurosci 25:8229–39.
Esclapez M, Hirsch JC, Ben Ari Y, Bernard C. 1999. Newly formed excitatory pathways provide a substrate for hyperexcitability in experimental temporal lobe epilepsy. J Comp Neurol 408:449–60.
Fisahn A, Pike FG, Buhl EH, Paulsen O. 1998. Cholinergic induction of network oscillations at 40 Hz in the hippocampus in vitro. Nature 394:186–9.
Freund TF. 2003. Interneuron Diversity series: Rhythm and mood in perisomatic inhibition. Trends Neurosci 26:489–95.
Freund TF, Buzsaki G. 1996. Interneurons of the hippocampus. Hippocampus 6:347–470.
Freund TF, Buszaki G, Leon A, Baimbridge KG, Somogyi P. 1990. Relationship of neuronal vulnerability and calcium-binding protein immunoreactivity in ischemia. Exp Brain Res 83:55–66.
Freund TF, Magloczky Z. 1993. Early degeneration of calretinin-containing neurons in the rat hippocampus after ischemia. Neuroscience 56:581–96.
Fritschy JM, Paysan J, Enna A, Mohler H. 1994. Switch in the expression of rat GABAA-receptor subtypes during postnatal development: an immunohistochemical study. J Neurosci 14:5302–24.
Futrell N, Watson BD, Dietrich WD, Prado R, Millikan C, Ginsberg MD. 1988. A new model of embolic stroke produced by photochemical injury to the carotid artery in the rat. Ann Neurol 23:251–7.
Garcia JH. 1984. Experimental ischemic stroke: a review. Stroke 15:5–14.
Ginsberg MD, Busto R. 1989. Rodent models of cerebral ischemia. Stroke 20:1627–42.
Globus MY, Busto R, Dietrich WD, Martinez E, Valdes I, Ginsberg MD. 1988. Effect of ischemia on the in vivo release of striatal dopamine, glutamate, and gamma-aminobutyric acid studied by intracerebral microdialysis. J Neurochem 51:1455–64.
Goldstein PA, Elsen FP, Ying SW, Ferguson C, Homanics GE, Harrison NL. 2002. Prolongation of hippocampal miniature inhibitory postsynaptic currents in mice lacking the GABA(A) receptor alpha1 subunit. J Neurophysiol 88:3208–17.
Gulyas AI, Hajos N, Freund TF. 1996. Interneurons containing calretinin are specialized to control other interneurons in the rat hippocampus. J Neurosci 16:3397–411.
Gupta SR, Naheedy MH, Elias D, Rubino FA. 1988. Postinfarction seizures: a clinical study. Stroke 19:1477–81.
Gustafsson B. 1979. Changes in motoneurone electrical properties following axotomy. J Physiol 293:197–215.
Haglund MM, Schwartzkroin PA. 1990. Role of Na-K pump potassium regulation and IPSPs in seizures and spreading depression in immature rabbit hippocampal slices. J Neurophysiol 63:225–39.
Haglund MM, Stahl WL, Kunkel DD, Schwartzkroin PA. 1985. Developmental and regional differences in the localization of Na,K- ATPase activity in the rabbit hippocampus. Brain Res 343:198–203.
Hajos N, Palhalmi J, Mann EO, Nemeth B, Paulsen O, Freund TF. 2004. Spike timing of distinct types of GABAergic interneuron during hippocampal gamma oscillations in vitro. J Neurosci 24:9127–37.
Hammond C, Crepel V, Gozlan H, Ben Ari Y. 1994. Anoxic LTP sheds light on the multiple facets of NMDA receptors. Trends Neurosci 17:497–503.
Hefft S, Jonas P. 2005. Asynchronous GABA release generates long-lasting inhibition at a hippocampal interneuron-principal neuron synapse. Nat Neurosci 8:1319–28.
Heinen K, Bosman LW, Spijker S, van PJ, Smit AB, Voorn P, and others. 2004. GABAA receptor maturation in relation to eye opening in the rat visual cortex. Neuroscience 124:161–71.
Houser CR, Esclapez M. 2003. Downregulation of the alpha5 subunit of the GABA(A) receptor in the pilocarpine model of temporal lobe epilepsy. Hippocampus 13:633–45.
Hsu KS, Huang CC. 1997. Characterization of the anoxia-induced long-term synaptic potentiation in area CA1 of the rat hippocampus. Br J Pharmacol 122:671–81.
Hsu KS, Huang CC. 1998. Protein kinase C inhibitors block generation of anoxia-induced long-term potentiation. NeuroReport 9: 3525–9.

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Hsu M, Buzsaki G. 1993. Vulnerability of mossy fiber targets in the rat hippocampus to forebrain ischemia. J Neurosci 13:3964–79.
Hsu M, Sik A, Gallyas F, Horvath Z, Buzsaki G. 1994. Short-term and long-term changes in the postischemic hippocampus. Ann N Y Acad Sci 743:121–39.
Jensen FE, Applegate CD, Holtzman D, Belin TR, Burchfiel JL. 1991. Epileptogenic effect of hypoxia in the immature rodent brain. Ann Neurol 29:629–37.
Jourdain P, Nikonenko I, Alberi S, Muller D. 2002. Remodeling of hippocampal synaptic networks by a brief anoxia-hypoglycemia. J Neurosci 22:3108–16.
Karhunen H, Jolkkonen J, Sivenius J, Pitkanen A. 2005. Epileptogenesis after experimental focal cerebral ischemia. Neurochem Res 30:1529–42.
Karhunen H, Pitkanen A, Virtanen T, Gureviciene I, Pussinen R, Ylinen A, and others. 2003. Long-term functional consequences of transient occlusion of the middle cerebral artery in rats: a 1-year follow-up of the development of epileptogenesis and memory impairment in relation to sensorimotor deficits. Epilepsy Res 54:1–10.
Kelly KM. 2002. Poststroke seizures and epilepsy: clinical studies and animal models. Epilepsy Curr 2:173–7.
Kelly KM, Kharlamov A, Hentosz TM, Kharlamova EA, Williamson JM, Bertram EH III, and others. 2001. Photothrombotic brain infarction results in seizure activity in aging Fischer 344 and Sprague Dawley rats. Epilepsy Res 47:189–203.
Kharlamov EA, Jukkola PI, Schmitt KL, Kelly KM. 2003. Electrobehavioral characteristics of epileptic rats following photothrombotic brain infarction. Epilepsy Res 56:185–203.
Khazipov R, Holmes GL. 2003. Synchronization of kainate-induced epileptic activity via GABAergic inhibition in the superfused rat hippocampus in vivo. J Neurosci 23:5337–41.
Kilpatrick CJ, Davis SM, Hopper JL, Rossiter SC. 1992. Early seizures after acute stroke: risk of late seizures. Arch Neurol 49:509–11.
Kirino T. 2000. Delayed neuronal death. Neuropathology 20(suppl): S95–7.
Klausberger T, Marton LF, O'Neill J, Huck JH, Dalezios Y, Fuentealba P, and others. 2005. Complementary roles of cholecystokinin- and parvalbumin-expressing GABAergic neurons in hippocampal network oscillations. J Neurosci 25:9782–93.
Klausberger T, Roberts JD, Somogyi P. 2002. Cell type- and input-specific differences in the number and subtypes of synaptic GABA(A) receptors in the hippocampus. J Neurosci 22:2513–21.
Klishin A, Tsintsadze T, Lozovaya N, Krishtal O. 1995. Latent N-methyl-D-aspartate receptors in the recurrent excitatory pathway between hippocampal CA1 pyramidal neurons: Ca(2+)-dependent activation by blocking A1 adenosine receptors. Proc Natl Acad Sci USA 92:12431–5.
Kobayashi M, Buckmaster PS. 2003. Reduced inhibition of dentate granule cells in a model of temporal lobe epilepsy. J Neurosci 23:2440–52.
Koksma JJ, van Kesteren RE, Rosahl TW, Zwart R, Smit AB, Luddens H, and others. Oxytocin regulates neurosteroid modulation of GABA(A) receptors in supraoptic nucleus around parturition. J Neurosci 23: 788–97.
Kotila M, Waltimo O. 1992. Epilepsy after stroke. Epilepsia 33:495–8.
Krantic S, Mechawar N, Reix S, Quirion R. 2005. Molecular basis of programmed cell death involved in neurodegeneration. Trends Neurosci 28:670–6.
Laiwand R, Werman R, Yarom Y. 1988. Electrophysiology of degenerating neurones in the vagal motor nucleus of the guinea-pig following axotomy. J Physiol 404:749–66.
Lancman ME, Golimstok A, Norscini J, Granillo R. 1993. Risk factors for developing seizures after a stroke. Epilepsia 34:141–3.
Laurent JP, Molinari GF, Oakley JC. 1976. Primate model of cerebral hematoma. J Neuropathol Exp Neurol 35:560–68.
Laurie DJ, Wisden W, Seeburg PH. 1992. The distribution of thirteen GABAA receptor subunit mRNAs in the rat brain: III. Embryonic and postnatal development. J Neurosci 12:4151–72.
Li Y, Jiang N, Powers C, Chopp M. 1998. Neuronal damage and plasticity identified by microtubule-associated protein 2, growth-associated protein 43, and cyclin D1 immunoreactivity after focal cerebral ischemia in rats. Stroke 29:1972–80.
Liu J, Schmitt KL, Kharlamov EA, Stolarski CJ, Grayson DR, Kelly KM. 2002. Quantitative reverse transcription-polymerase chain reaction of GABA(A) alpha1, beta1 and gamma2S subunits in epileptic rats following photothrombotic infarction of neocortex. Epilepsy Res 52:85–95.
Longa EZ, Weinstein PR, Carlson S, Cummins R. 1989. Reversible middle cerebral artery occlusion without craniectomy in rats. Stroke 20:84–91.
Luhmann H, Mudrick-Donnon LA, Mittmann T, Heinemann U. 1995. Ischaemia-induced long-term hyperexcitability in rat neocortex. Eur J Neurosci 7:180–91.
McIntyre DC, Hutcheon B, Schwabe K, Poulter MO. 2002. Divergent GABA(A) receptor-mediated synaptic transmission in genetically seizure-prone and seizure-resistant rats. J Neurosci 22:9922–31.
McNamara JO. 1979. Human hypoxia and seizures: effects and interactions. Adv Neurol 26:137–43.
Meldrum BS. 2002. Concept of activity-induced cell death in epilepsy: historical and contemporary perspectives. Prog Brain Res 135:3–11.
Menendez de la Prida L, Huberfeld G, Cohen I, Miles R. 2006. Threshold behavior in the initiation of hippocampal population bursts. Neuron 49:131–42.
Miles R, Toth K, Gulyas AI, Hajos N, Freund TF. 1996. Differences between somatic and dendritic inhibition in the hippocampus. Neuron 16:815–23.
Miles R, Wong RKS. 1987. Inhibitory control of local excitatory circuits in the guinea-pig hippocampus. J Physiol (London) 388: 611–29.
Mittmann T, Luhmann HJ, Schmidt-Kastner R, Eysel UT, Weigel H, Heinemann U. 1994. Lesion-induced transient suppression of inhibitory function in rat neocortex in vitro. Neuroscience 60: 891–906.
Miyake K, Yamamoto W, Tadokoro M, Takagi N, Sasakawa K, Nitta A, and others. 2002. Alterations in hippocampal GAP-43, BDNF, and L1 following sustained cerebral ischemia. Brain Res 935:24–31.
Mody I, Otis TS, Bragin A, Hsu M, Buzsaki G. 1995. GABAergic inhibition of granule cells and hilar neuronal synchrony following ischemia-induced hilar neuronal loss. Neuroscience 69:139–50.
Molinari GF. 1983. Neurologists are like cops. Neurology 33:1102–3.
Mudrick LA, Baimbridge KG. 1989. Long-term structural changes in the rat hippocampal formation following cerebral ischemia. Brain Res 493:179–84.
Nakayama H, Ginsberg MD, Dietrich WD. 1988. (S)-emopamil, a novel calcium channel blocker and serotonin S2 antagonist, markedly reduces infarct size following middle cerebral artery occlusion in the rat. Neurology 38:1667–73.
Neumann-Haefelin T, Hagemann G, Witte OW. 1995. Cellular correlates of neuronal hyperexcitability in the vicinity of photochemically induced cortical infarcts in rats in vitro. Neurosci Lett 193: 101–4.
Neumann-Haefelin T, Staiger JF, Redecker C, Zilles K, Fritschy JM, Mohler H, and others. 1998. Immunohistochemical evidence for dysregulation of the GABAergic system ipsilateral to photochemically induced cortical infarcts in rats. Neuroscience 87:871–9.
Nordstrom CH, Rehncrona S, Siesjö BK. 1978. Effects of phenobarbital in cerebral ischemia: Part II. Restitution of cerebral energy state, as well as of glycolytic metabolites, citric acid cycle intermediates and associated amino acids after pronounced incomplete ischemia. Stroke 9:335–43.
Nordstrom CH, Siesjö BK. 1978. Effects of phenobarbital in cerebral ischemia: Part I. cerebral energy metabolism during pronounced incomplete ischemia. Stroke 9:327–35.
Nusser Z, Sieghart W, Benke D, Fritschy JM, Somogyi P. 1996. Differential synaptic localization of two major gamma-aminobutyric acid type A receptor alpha subunits on hippocampal pyramidal cells. Proc Natl Acad Sci USA 93:11939–44.
Nyiri G, Freund TF, Somogyi P. 2001. Input-dependent synaptic targeting of alpha(2)-subunit-containing GABA(A) receptors in synapses of hippocampal pyramidal cells of the rat. Eur J Neurosci 13:428–42.
Obenaus A, Esclapez M, Houser CR. 1993. Loss of glutamate-decarboxylase messenger-RNA containing neurons in the rat dentate gyrus following pilocarpine-induced seizures. J Neurosci 13:4470–85.

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Okada M, Onodera K, Van RC, Sieghart W, Takahashi T. 2000. Functional correlation of GABA(A) receptor alpha subunits expression with the properties of IPSCs in the developing thalamus. J Neurosci 20:2202–8.
Okazaki MM, Evenson DA, Nadler JV. 1995. Hippocampal mossy fiber sprouting and synapse formation after status epilepticus in rats: visualization after retrograde transport of biocytin. J Comp Neurol 352:515–34.
Olafsson E, Gudmundsson G, Hauser WA. 2000. Risk of epilepsy in long-term survivors of surgery for aneurysmal subarachnoid hemorrhage: a population-based study in Iceland. Epilepsia 41:1201–5.
Osborne KA, Shigeno T, Balarsky AM, Ford I, McCulloch J, Teasdale GM, and others. 1987. Quantitative assessment of early brain damage in a rat model of focal cerebral ischaemia. J Neurol Neurosurg Psychiatry 50:402–10.
Papas S, Crepel V, Ben-Ari Y. 1993. The NMDA receptor contributes to anoxic aglycemic induced irreversible inhibition of synaptic transmission. Brain Res 607:54–60.
Pearce RA. 1993. Physiological evidence for two distinct GABAA responses in rat hippocampus. Neuron 10:189–200.
Petito CK, Feldmann E, Pulsinelli WA, Plum F. 1987. Delayed hippocampal damage in humans following cardiorespiratory arrest. Neurology 37:1281–6.
Picconi B, Tortiglione A, Barone I, Centonze D, Gardoni F, Gubellini P, and others. 2006. NR2B subunit exerts a critical role in postischemic synaptic plasticity. Stroke 37:1895–901.
Pirker S, Schwarzer C, Wieselthaler A, Sieghart W, Sperk G. 2000. GABA(A) receptors: immunocytochemical distribution of 13 subunits in the adult rat brain. Neuroscience 101:815–50.
Poolos NP. 2006. H-channel dysfunction in generalized epilepsy: it takes two. Epilepsy Curr 6:88–90.
Poulter MO, Brown LA, Tynan S, Willick G, William R, McIntyre DC. 1999. Differential expression of alpha1, alpha2, alpha3, and alpha5 GABAA receptor subunits in seizure-prone and seizure-resistant rat models of temporal lobe epilepsy. J Neurosci 19:4654–61.
Prado R, Ginsberg MD, Dietrich WD, Watson BD, Busto R. 1988. Hyperglycemia increases infarct size in collaterally perfused but not end-arterial vascular territories. J Cereb Blood Flow Metab 8:186–92.
Pulsinelli WA. 1985. Selective neuronal vulnerability: morphological and molecular characteristics. Prog Brain Res 63:29–37.
Pulsinelli WA, Brierley JB. 1979. A new bilateral hemispheric ischemia in the unanesthetised rat. Stroke 10:267–72.
Pulsinelli WA, Brierley JB, Plum F. 1982a. Temporal profile of neuronal damage in a model of transient forebrain ischemia. Ann Neurol 11:491–8.
Pulsinelli WA, Buchan AM. 1988. The four-vessel occlusion rat model: method for complete occlusion of vertebral arteries and control of collateral circulation. Stroke 19:913–4.
Pulsinelli WA, Levy DE, Duffy TE. 1982b. Regional cerebral blood flow and glucose metabolism following transient forebrain ischemia. Ann Neurol 11:499–502.
Quintana P, Alberi S, Hakkoum D, Muller D. 2006. Glutamate receptor changes associated with transient anoxia/hypoglycaemia in hippocampal slice cultures. Eur J Neurosci 23:975–83.
Redecker C, Wang W, Fritschy JM, Witte OW. 2002. Widespread and long-lasting alterations in GABA(A)-receptor subtypes after focal cortical infarcts in rats: mediation by NMDA-dependent processes. J Cereb Blood Flow Metab 22:1463–75.
Reith J, Jorgensen HS, Nakayama H, Raaschou HO, Olsen TS. 1997. Seizures in acute stroke: predictors and prognostic significance. The Copenhagen Stroke Study. Stroke 28:1585–9.
Remy S, Beck H. 2006. Molecular and cellular mechanisms of pharmacoresistance in epilepsy. Brain 129:18–35.
Represa A, Crepel V, Ben Ari Y. 1991. Transient cerebral ischemia induces changes in SRIF mRNA in the fascia dentata. Brain Res Mol Brain Res 10:337–42.
Represa A, Tremblay E, Ben-Ari Y. 1987. Kainate binding sites in the hippocampal mossy fibers: localization and plasticity. Neuroscience 20:739–48.
Rivera C, Voipio J, Kaila K. 2005. Two developmental switches in GABAergic signalling: the K+-Cl-cotransporter KCC2 and carbonic anhydrase CAVII. J Physiol 562:27–36.
Schmidt-Kastner R, Freund TF. 1991. Selective vulnerability of the hippocampus in brain ischemia. Neuroscience 40:599–636.
Schwarzer C, Tsunashima K, Wanzenbock C, Fuchs K, Sieghart W, Sperk G. 1997. GABA(A) receptor subunits in the rat hippocampus II: altered distribution in kainic acid–induced temporal lobe epilepsy. Neuroscience 80:1001–17.
Shigeno T, Teasdale GM, McCulloch J, Graham DI. 1985. Recirculation model following MCA occlusion in rats: cerebral blood flow, cerebrovascular permeability, and brain edema. J Neurosurg 63:272–7.
Sieghart W. 2000. Unraveling the function of GABA(A) receptor subtypes. Trends Pharmacol Sci 21:411–3.
Siesjö BK. 1978. Brain energy metabolism. New York: John Wiley.
Sik A, Tamamaki N, Freund TF. 1993. Complete axon arborization of a single CA3 pyramidal cell in the rat hippocampus, and its relationship with postsynaptic parvalbumin-containing interneurons. Eur J Neurosci 5:1719–28.
Smith ML, Auer RN, Siesjö BK. 1984. The density and distribution of ischemic brain injury in the rat following 2-10 min of forebrain ischemia. Acta Neuropathol (Berl) 64:319–32.
So EL, Annegers JF, Hauser WA, O'Brien PC, Whisnant JP. 1996. Population-based study of seizure disorders after cerebral infarction. Neurology 46:350–5.
Somogyi P, Klausberger T. 2005. Defined types of cortical interneurone structure space and spike timing in the hippocampus. J Physiol 562:9–26.
Sperk G, Schwarzer C, Tsunashima K, Kandlhofer S. 1998. Expression of GABA(A) receptor subunits in the hippocampus of the rat after kainic acid-induced seizures. Epilepsy Res 32:129–39.
Staley K, Hellier JL, Dudek FE. 2005. Do interictal spikes drive epileptogenesis? Neuroscientist 11:272–6.
Storm JF. 1990. Potassium currents in hippocampal pyramidal cells. Prog Brain Res 83:161–87.
Stroemer RP, Kent TA, Hulsebosch CE. 1993. Acute increase in expression of growth associated protein GAP-43 following cortical ischemia in rat. Neurosci Lett 162:51–4.
Sung CY, Chu NS. 1989. Epileptic seizures in intracerebral haemorrhage. J Neurol Neurosurg Psychiatry 52:1273–6.
Sutula T, He XX, Cavazos J, Scott G. 1988. Synaptic reorganization in the hippocampus induced by abnormal functional-activity. Science 239:1147–50.
Taketo M, Yoshioka T. 2000. Developmental change of GABA(A) receptor-mediated current in rat hippocampus. Neuroscience 96:507–14.
Tekkok S, Krnjevic K. 1995. Long-term potentiation in hippocampal slices induced by temporary suppression of glycolysis. J Neurophysiol 74:2763–6.
Traub RD, Bibbig A, Fisahn A, Lebeau FEN, Whittington MA, Buhl EH. 2000. A model of gamma-frequency network oscillations induced in the rat CA3 region by carbachol in vitro. Eur J Neurosci 12:4093–106.
Traub RD, Whittington MA, Colling SB, Buzsaki G, Jefferys JG. 1996. Analysis of gamma rhythms in the rat hippocampus in vitro and in vivo. J Physiol 493(pt 2):471–84.
Tsintsadze T, Lozovaya N, Klishin A, Krishtal O. 1996. NMDA receptor-mediated synapses between CA1 neurons: activation by ischaemia. NeuroReport 7:2679–82.
Urban L, Neill KH, Crain BJ, Nadler JV, Somjen GG. 1990. Postischemic synaptic excitation and N-methyl-D-aspartate receptor activation in gerbils. Stroke 21(suppl III):III-23.
Varelas PN, Mirski MA. 2001. Seizures in the adult intensive care unit. J Neurosurg Anesthesiol 13:163–75.
Vicini S, Ferguson C, Prybylowski K, Kralic J, Morrow AL, Homanics GE. 2001. GABA(A) receptor alpha1 subunit deletion prevents developmental changes of inhibitory synaptic currents in cerebellar neurons. J Neurosci 21:3009–16.
Volpe BT, Pulsinelli WA, Tribuna J, Davis HP. 1984. Behavioral performance of rats following transient forebrain ischemia. Stroke 15:558–62.
Watson BD, Dietrich WD, Busto R, Wachtel MS, Ginsberg MD. 1985. Induction of reproducible brain infarction by photochemically initiated thrombosis. Ann Neurol 17:497–504.
Williams PA, Dou P, Dudek FE. 2004. Epilepsy and synaptic reorganization in a perinatal rat model of hypoxia-ischemia. Epilepsia 45:1210–8.

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Willmore LJ. 1990. Post-traumatic epilepsy: cellular mechanisms and implications for treatment. Epilepsia 31(suppl 3):S67–73.
Yoshida S, Busto R, Watson BD, Santiso M, Ginsberg MD. 1985. Postischemic cerebral lipid peroxidation in vitro: modification by dietary vitamin E. J Neurochem 44:1593–601.
Zepeda A, Sengpiel F, Guagnelli MA, Vaca L, Arias C. 2004. Functional reorganization of visual cortex maps after ischemic lesions is accompanied by changes in expression of cytoskeletal proteins and NMDA and GABA(A) receptor subunits. J Neurosci 24:1812–21.
Zola-Morgan S, Squire LR, Amaral DG. 1986. Human amnesia and the medial temporal region: enduring memory impairment following a bilateral lesion limited to field CA1 of the hippocampus. J Neurosci 6:2950–67.
Zola-Morgan S, Squire LR, Rempel NL, Clower RP, Amaral DG. 1992. Enduring memory impairment in monkeys after ischemic damage to the hippocampus. J Neurosci 12:2582–96.

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