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RESEARCH ARTICLE

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Increased afterdischarge threshold during kindling in epileptic rats

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Abstract The effects of daily electrical kindling stimulation of the perforant pathway were investigated in an excitotoxic rat model of epilepsy with chronic seizures in order to learn whether the preexisting epileptic condition would facilitate or retard kindling. Sprague-Dawley rats with recurrent spontaneous seizures 4–8 months after unilateral intrahippocampal kainic acid (KA) injection were implanted with recording electrodes in the hippocampus and stimulating electrodes in the perforant path. Daily stimulation for 10 s at 5 Hz was given for 15 days. The afterdischarge (AD) threshold and the AD duration of kindled KA rats were compared before and during kindling with those of a kindled control group. In the control group, as expected, mean AD thresholds decreased ($P<0.01$), while AD duration progressively increased. Although AD threshold was the same in KA and control groups at the start of kindling, in the KA group a significant *increase* in threshold occurred from the beginning to the end of kindling ($P<0.01$). Behaviorally, KA rats showed stage 4 or 5 seizures on the first stimulation, and stage 3–5 seizures during the remainder of kindling. Paired pulse testing showed facilitation of late components of the dentate gyrus field potential at the beginning of kindling, and suppression of late components at the end, in the KA rats. A significant decrease in the rate of spontaneous seizures in KA rats was noted during the period of kindling ($P=0.04$). These results suggest that electrical stimulation of the perforant path may strengthen homeostatic seizure suppressing mechanisms, and may provide insights into novel approaches to the treatment of clinical seizures in temporal lobe epilepsy.

Keywords Epilepsy · Hippocampus · Kainic acid · Stimulation · Field potentials

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Introduction

A large number of studies have shown that both positive and negative transfer effects can be seen with multiple site kindling. For instance, kindling of one amygdala decreases the number of kindling stimuli necessary to produce a stage 5 seizure at other ipsilateral or contralateral limbic sites (McIntyre and Goddard 1973), while secondary site kindling reduces epileptogenicity of the primary site (Duchowny and Burchfiel 1981; Mazarati and Wasterlain 1997), and alternate site kindling results in activation of seizure-facilitating mechanisms at one site, and seizure-suppressing mechanisms at the other (Burchfiel et al. 1982).

Furthermore, generalized seizures are followed by a transient refractory period during which subsequent seizure generation may be completely suppressed (Stripling and Russell 1989), generalized seizures in rats produced by electroconvulsive shock may severely retard kindling (Post et al. 1986), and repeated kindled seizures in fully kindled animals may cause enduring resistance to elicitation of subsequent kindled seizures (Pinel et al. 1976; Mucha and Pinel 1977). Kelly and McIntyre (1994) showed that previously kindled rats underwent less damage in piriform cortex and hippocampus after subcutaneous kainic acid injection. Such seizure-induced, seizure-suppressing mechanisms presumably reflect the natural homeostatic reactions to epilepsy necessary to maintain the interictal state (Caldecott-Hazard et al. 1982; Weiss et al. 1995). A better understanding of the fundamental neuronal basis of this protective effect may help develop approaches that can make clinical use of these mecha-

nisms to control spontaneous seizures in patients, as well as approaches to prevent failures of this protective mechanism which might precipitate spontaneous seizures.

The primary objective of our study was to compare the development of kindling in KA-treated rats with recurrent spontaneous seizures with that of naive rats. The second objective was to determine if the kindling treatment had any effect upon the frequency of spontaneous seizures in KA rats.

Materials and methods

All procedures described in this study were approved by the University of California, Los Angeles, Institutional Animal Care and Use Committee. Two groups of rats were used for kindling experiments, rats having chronic spontaneous seizures several months after a KA injection in the right posterior hippocampus (n=9), and naive age-matched rats (n=6).

Kainic acid treated rats

Adult male (250–350 g) Sprague-Dawley rats were given atropine (0.04 mg i.m.), anesthetized with chloral hydrate (400 mg/kg i.p.), and unilaterally injected with KA (0.4 µg/0.2 µl normal saline) in the right posterior hippocampus (AP=–5.6 mm, L=4.0 mm, V=7.0 mm; Bragin et al. 1999a). Beginning 3–4 months after injection, rats were observed during repeated 16- to 24-h video monitoring periods for 1–2 weeks every 1–2 months in order to detect spontaneous behavioral seizures. Nine rats that showed recurrent spontaneous seizures were chosen for experiments. They were subjected to an additional 24 h of video-monitoring for 1 week prior to electrophysiological experiments.

After completion of preliminary video monitoring, the rats were anesthetized with a mixture of ketamine (100 mg/kg), xylazine (5.2 mg/kg), and acepromazine (1.0 mg/kg). Moveable recording electrodes were implanted bilaterally at the same coordinates as in control rats, and the same electrophysiological criteria were used to identify the position of the recording electrodes. The stimulating electrode was implanted in the perforant path ipsilateral to the KA injection.

An array of four microelectrodes, fixed in relation to one another, but moveable together in the vertical plane (tungsten, 60 µm in diameter, insulated except at the tips), was used for recording from the hippocampus. The wires were arrayed in a single plane, with each wire separated horizontally from the adjacent wire by 300 µm. Vertically, the tip of each successive wire was 500 µm longer than the adjacent wire, achieving separation both horizontally and vertically. During initial implantation, the electrode tips were oriented along the long axis of the hippocampus, and lowered vertically into the brain to a point just above the dorsal hippocampus. Arrays were implanted in either or both mid dorsal hippocampus (AP=–3.0, L=2.6), and/or more posteriorly (AP=–5.6, L=4.0) (Paxinos and Watson 1997). The locations of the recording electrodes in the dentate gyrus (DG) and CA1 area of hippocampus were chosen on the basis of the shape of the evoked potentials elicited by perforant path stimulation. Electrodes with responses consisting of a positive field excitatory postsynaptic potential (fEPSP) with a superimposed negative population spike (PS) of 3–8 ms latency were considered to be located near the granule cell layer of the DG. Within 1 h before each kindling trial, paired pulse testing was used to determine the strength of paired pulse suppression over the course of kindling. Pairs of stimuli were separated by 10 s, and were given at interstimulus intervals of 5, 20, 30, 50, 70, 100, 200, 250, 300, and 500 ms (n=5 stimuli for each interval). At onset of each kindling train each day, the initial field response was measured as an indication of DG excitability at the moment the kindling train was delivered.

Perforant path kindling stimulation consisted of a 10-s train of 0.2-ms square pulses at 5 Hz. A frequency of 5 Hz was chosen because it was fast enough to produce an afterdischarge (AD), but slow enough to permit measurement of frequency facilitation. The current intensity was set initially at twice the PS threshold in the DG (50–200 µA) and was adjusted for each rat so that a 5-Hz 10-s train was effective in evoking AD with duration greater than 5 s. The stimulation trains were administered once every 24 h for 15 days. On subsequent trials the train with the same initially effective current was given. If this current intensity failed to induce an AD, stimulation was repeated again every 30–60 min with 10% increased current intensity until an AD occurred. On the last 3 days of kindling, stimulation was started at 5 times less than the initial stimulation that evoked AD on the 1st day of kindling, and repetitive trains with a 10% increase in current were delivered every 30 min to determine the threshold for AD.

In three other KA-treated rats, kindling was carried out with the same stimulus frequency, but with the current 4–5 times higher than the threshold for population spikes rather than the twice threshold current used in the others. To avoid interference of spontaneous seizures upon AD, at least 4 h elapsed after a spontaneous seizure before any kindling stimulation was applied.

Control rats

Adult male (250–350 g) Sprague-Dawley rats (n=6) were anesthetized with a mixture of ketamine (100 mg/kg), xylazine (5.2 mg/kg), and acepromazine (1.0 mg/kg). Pairs of tungsten wires (60 µm in diameter) with 0.5 mm vertical tip separation were placed in the right angular bundle to stimulate perforant path afferents to the hippocampus (AP=–7.0 mm from bregma, L=3.5 mm from midline and V=2.5 mm from the surface of neocortex). The perforant path kindling procedure for control rats was similar to that in kainate rats.

Data acquisition

Five 4-channel Mosfet input operational amplifiers, mounted in the cable connector, served to eliminate cable movement artifacts (Buzsaki et al. 1989). Physiological data were recorded wide-band (0.1 Hz to 1 kHz for field potentials or 1.0 Hz to 5.0 kHz for units and field potentials) and sampled at 10 kHz/channel (16 channels) with 12 bit precision on a Pentium PC using RC-Electronics software. Field potentials evoked by electrical stimulation were digitized and averaged (n=5) for each test. The data were stored on Jaz cartridges.

For detecting behavioral spontaneous seizures, rats were observed continuously during 24-h sessions of video monitoring and 16-channel EEG recording using a Biological Monitoring Systems Inc. (BMSI-4000; Nicolet, Madison, WI) telemetry system.

Histological procedures

At the end of the electrophysiological experiments, rats were deeply anesthetized and perfused with glutaraldehyde prior to Nissl staining to verify electrode placements.

Data analysis

Data analysis was carried out offline on a Pentium computer, using RC-Electronics software. The slope of fEPSP was estimated by the measurement of the amplitude value between the starting point of PS and 1 ms left on the fEPSP curve. The amplitude of the PS was estimated by measurement of the first deflection of the PS peak. Videotapes were reviewed and detected seizures were scored on the basis of Racine's scale (Racine 1972a). Data were analyzed with ANOVA and appropriate individual group tests of significance.

Results

Effects of kindling in control and KA treated rats

Control group data were similar to our earlier results on naive rats kindled with 200-Hz stimulation trains (Bragin et al. 1997). The afterdischarge threshold in control rats with electrodes in the perforant path varied between 80 and 150 μA. Repetitive kindling stimulation led to a decrease in the threshold current required for AD from a mean of 96 μA to a mean of 30 μA (Fig. 1A, white bars). The duration of AD increased from 22.2±5.3 s (±=SD) during the first 3 days to 48.4±9.4 s during the last 3 days of kindling ($P<0.01$). The first kindling stimulation evoking electrographic AD had no behavioral effects except occasional wetdog shakes. As kindling proceeded, the behavioral response progressed from stage 1 to stage 3 or stage 4. Stage 5 seizures were not observed in our control rats.

There were no significant differences in the threshold of AD in KA-treated rats compared to control rats at the beginning of kindling. However, with repetition of stimulation on subsequent days, the same parameters used on the 1st day became ineffective in evoking an AD in KA rats; therefore stronger stimulation was required to evoke an AD (Fig. 1A, B). At the end of the kindling procedure, the mean threshold of AD had increased to 195± 31 μA, almost twice the initial threshold ($P<0.01$). The mean duration of all ADs in KA rats was 79.3±29.6 s, which was significantly longer than the mean duration of all ADs in control rats, 31.3±9.4 s ($P<0.05$). Although there was a progressive increase in the duration of AD during kindling in the naive group, significant changes in AD duration over the course of kindling in the KA rats did not occur. The mean duration of AD during the first 3 days of kindling was 79.1±23.7 s and during the last 3 days of kindling, 76.8±45.3 s (NS).

In marked contrast to control rats, the first stimulation in KA rats was usually accompanied by a stage 4–5 seizure. With successive "kindling" stimulations, behavioral response to stimulation fluctuated between stages 2 and 4.

To insure that current strength was sufficient for effective kindling in KA rats, in a group of three KA rats, the kindling was carried out with *current 4–5 times greater* than the original threshold for a DG PS. These stimulation parameters are similar to those used in other studies showing effective kindling (Pinel et al. 1976; Burchfiel et al. 1982). The first kindling stimulation in the three KA rats receiving higher stimulating current intensity evoked ADs of significantly longer duration (171±32.3 s) than the ADs evoked in the group of epileptic rats receiving stimulation with the original threshold intensity (80.4±21.4). Figure 1C shows that in response to the second kindling stimulation with higher current intensity (day 2), there was a dramatic decrease in the AD duration in all three rats, which was maintained for the remaining 15 days of kindling. In spite of the decreased AD duration, the behavioral component in these animals did not change significantly, fluctuating between stages 3 and 5 (Fig. 1C).

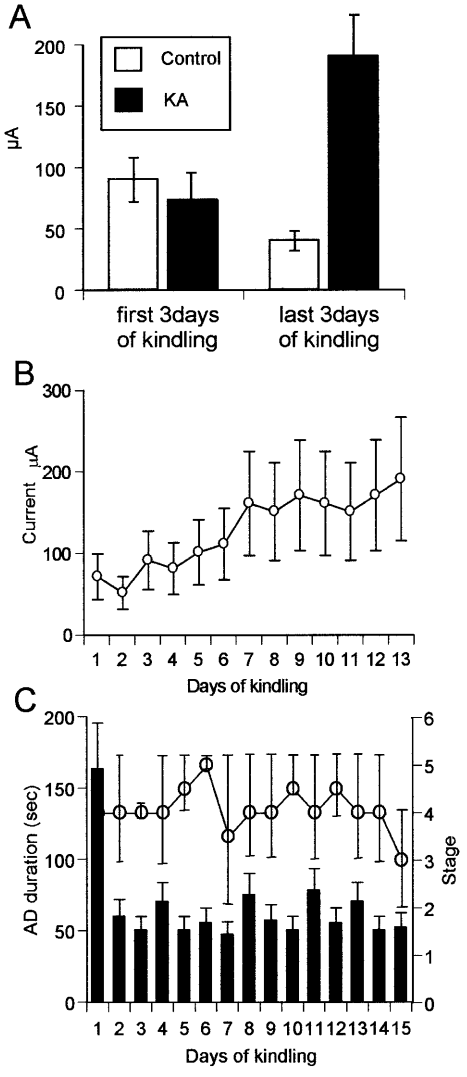

Fig. 1 **A** Kindling in control and KA epileptic rats. *The white bars* show the mean threshold in microamperes for afterdischarge (AD) in control and *black bars* in kainic acid treated groups during the first 3 days of kindling (*left*) and last 3 days of kindling (*right*). *Error bars* indicate SD. **B** Change in afterdischarge threshold during consecutive tetani in KA-treated rats. *Lines with open circles* show the mean change for six rats. *Error bars* indicate SD. **C** Changes in AD duration (*bars*) and behavioral kindling stage (*lines*) on the basis of Racine's scale in the KA-treated rat group (n=3) with suprathreshold kindling. *Error bars* = SD of AD duration and range for behavioral stage

Other electrophysiological correlates of repetitive kindling stimulation

In order to explore possible mechanisms underlying the resistance of KA animals to kindling and to evaluate the electrophysiological correlates of KA rat kindling, we studied changes in excitability, frequency potentiation and the strength of perforant path inhibition using the paired pulse paradigm.

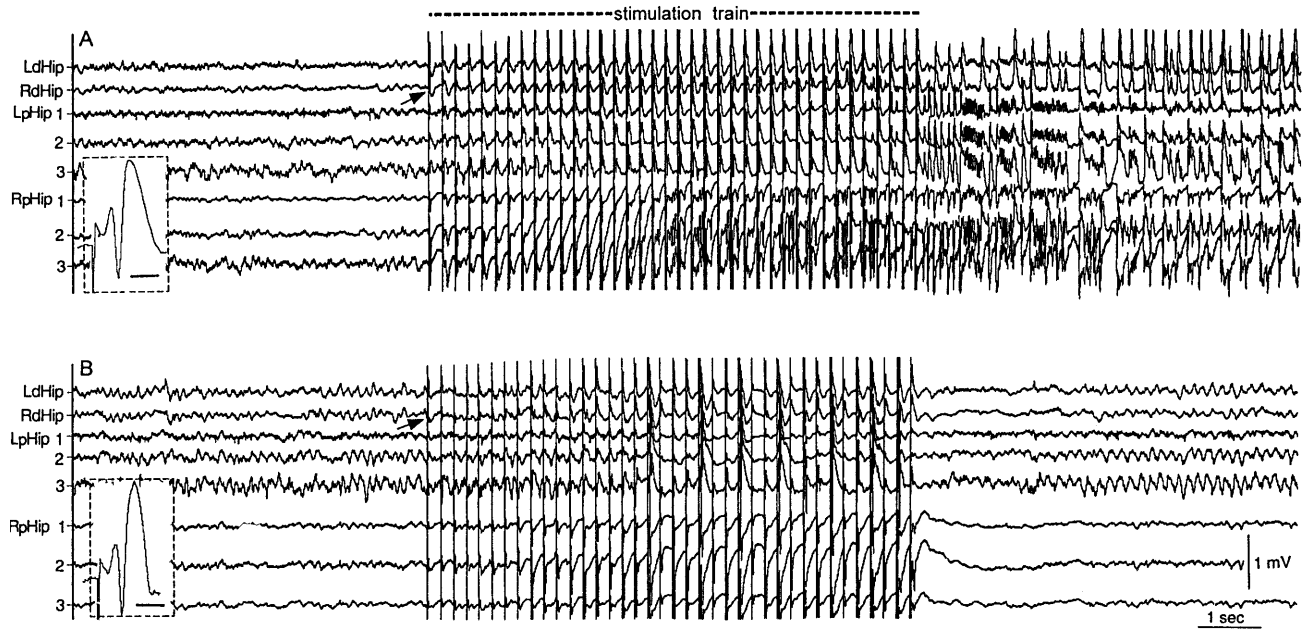

Fig. 2 The effect of identical tetanic stimulation delivered to the perforant path of a KA rat on hippocampal responses on the first **(A)** and the last **(B)** days of the kindling procedure. Expanded examples of the evoked potentials in response to the first pulses are shown as an *inset on the left side of each electrograph*. Excitability as measured by the evoked potential amplitude in response to perforant path stimulation (see *inset*) is similar on the first and the last days of the kindling procedure; however, AD is evoked on the first, but not the last, day. Time scale in the *inset in* **A** is 5 ms and applies also to the *inset in* **B** (*LdHip and RpHip* left and right dorsal and posterior hippocampus, *RpHip* right posterior hippocampus)

In control rats, during the kindling procedure we found an increase in the slope of the fEPSP without a significant change of PS amplitude in two rats, and an increase in both the fEPSP and PS in one rat. Similar electrophysiological correlates have been described in several previous papers (see Douglas and Goddard 1975; Racine et al. 1986; Maru and Goddard 1987a; Grace et al. 1990). In KA rats, the amplitude of the fEPSP and PS in the EC-DG pathway during single pulse stimulation was measured prior to kindling, to the initial response at the onset of the kindling train on the first 3 days of kindling, and to the initial response to the kindling train on the last 3 days of kindling. No significant changes in the fEPSP or PS amplitudes recorded under these three conditions were found. An example is shown in Fig. 2, illustrating kindling trains delivered to a KA rat on the first and last day of kindling. Although the stimulating train successfully evoked an AD on the 1st day, no AD occurred in response to stimulation with the same parameters on the last day, suggesting excitability was greater at the beginning of kindling than at the end of kindling. However, comparison of the first fEPSP and PS of each train shows that their amplitude remained unchanged.

In control rats, over the course of kindling, prominent frequency facilitation developed in response to the 5-Hz kindling stimulation. The same facilitation occurred in KA-treated rats during the first 1–3 days of kindling (Figs. 2A, 3A). In KA rats, however, frequency facilitation diminished on subsequent days of the kindling stimulation. Facilitation was considerably impaired on the last 3 days of kindling compared to the first 3 days of kindling in all six KA rats kindled with regular parameters (Figs. 2B, 3B).

One specific feature of DG evoked potentials to perforant path stimulation in KA rats with spontaneous seizures was the existence of secondary components in response to a single electrical pulse. Whereas the amplitude of the early components of potentials evoked in response to perforant path stimulation did not change significantly, the late components of potentials evoked in response by single or paired pulses decreased progressively following consecutive kindling stimulation. This is illustrated in Fig. 4, where paired pulses were delivered with intervals ranging from 5 to 500 ms. Figure 4A shows that on the 1st day pairs with 5- or 20-ms intervals did not affect the late components of the response to the conditioning pulse. With a 50-ms paired pulse interval, the late component of the response to the test (second) pulse partially recovered, and became significantly augmented at 100- to 500-ms intervals (Fig. 4A, inset). On the last day of kindling (Fig. 4B) the duration of the response was shorter. The late responses to the test pulse partially recovered at 20- to 50-ms intervals, but were still depressed at the 100- to 500-ms intervals (Fig. 4B, inset).

Kindling in KA rats and spontaneous seizures

As previously described (Bragin et al. 1999b) for this model of chronic epilepsy, spontaneous behavioral seizures occurred sporadically, with frequencies ranging

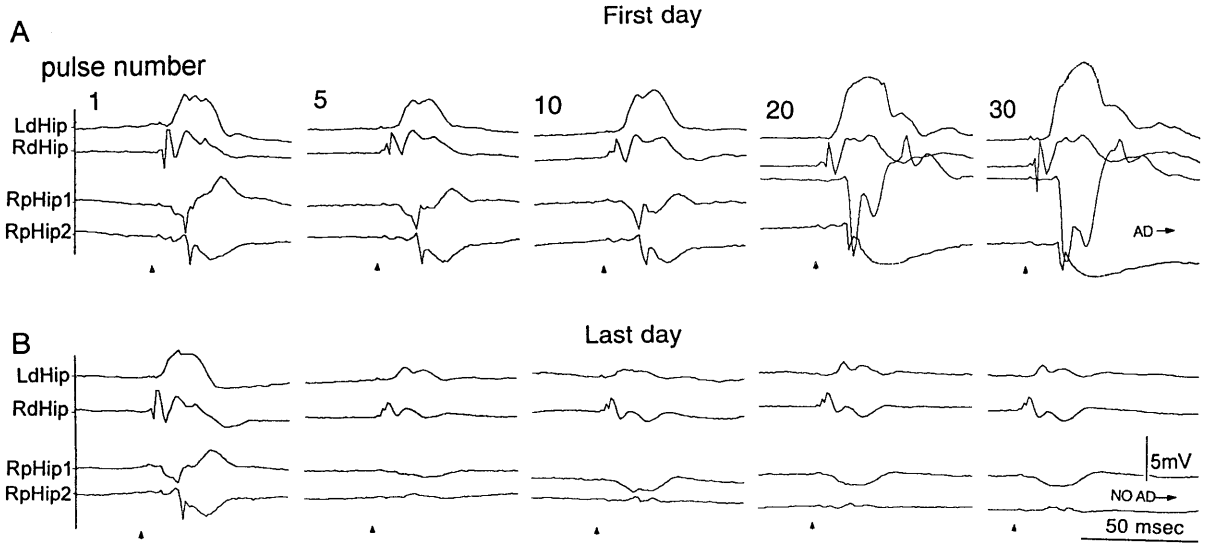

Fig. 3 The pattern of frequency response during a 5-Hz train of stimulation in an epileptic KA-rat applied to the right perforant path on the first **(A)** and last **(B)** days of kindling. There is prominent frequency facilitation in all recording points both ipsilateral and contralateral to stimulation of each successive single pulse stimulation on the 1st day of kindling, while after kindling there is a progressive decrease in response amplitude. Note the similarity of the amplitude of the evoked potentials to the first pulse in the first and last days of kindling (*LdHip and RpHip* left and right dorsal and posterior hippocampus, *RpHip1 and RpHip2* two recording points in the right posterior hippocampus)

Fig. 4 Examples of responses to paired pulse perforant path stimulation recorded in the ipsilateral dorsal dentate gyrus in epileptic KA rat on the first **(A)** and the last **(B)** days of kindling (averages of seven responses). The same current (100 μA) was used on the first and last day. Note that the earlier response component both S1 and S2 (*arrow with filled circle*) is larger on the last day of kindling, while the late component (*arrow with open circle*) is greatly reduced on the last day of kindling and does not show facilitation to the S2 pulse as on the first day. *Insets* show the change in the EPSP slope in the paired pulse paradigm (ratio between the conditioning and the test pulses) for the earlier response (*open circles*) and late response (*filled circles*). Note the extended polysynaptic components of the response to the first pulse at the beginning are reduced at the end of kindling

from several seizures per day to no seizures over periods of several weeks. This limited number of seizures made it difficult to obtain a large enough number of seizures to determine the influence of the kindling procedure on the frequency of spontaneous seizures. In spite of this limitation in the number of seizures, we found a significant (P=0.04) difference in the frequency of spontaneous seizures observed before and during the kindling procedure. These results are presented in Table 1, which shows the rate of seizures for a 1-week period before kindling and during the 2nd week of the kindling period (threshold stimulation for KA01-06 and suprathreshold KA07-09). The rate of seizures during the kindling period was 3.5 times lower than during the prekindling period.

Although no significant difference in seizure rate was found before and during the 2nd week of kindling in the three rats with suprathreshold stimulation, it is difficult to draw any conclusions from these data due to the small number of animals. The long-term consequences of kindling KA-treated rats were not studied in the present experiments.

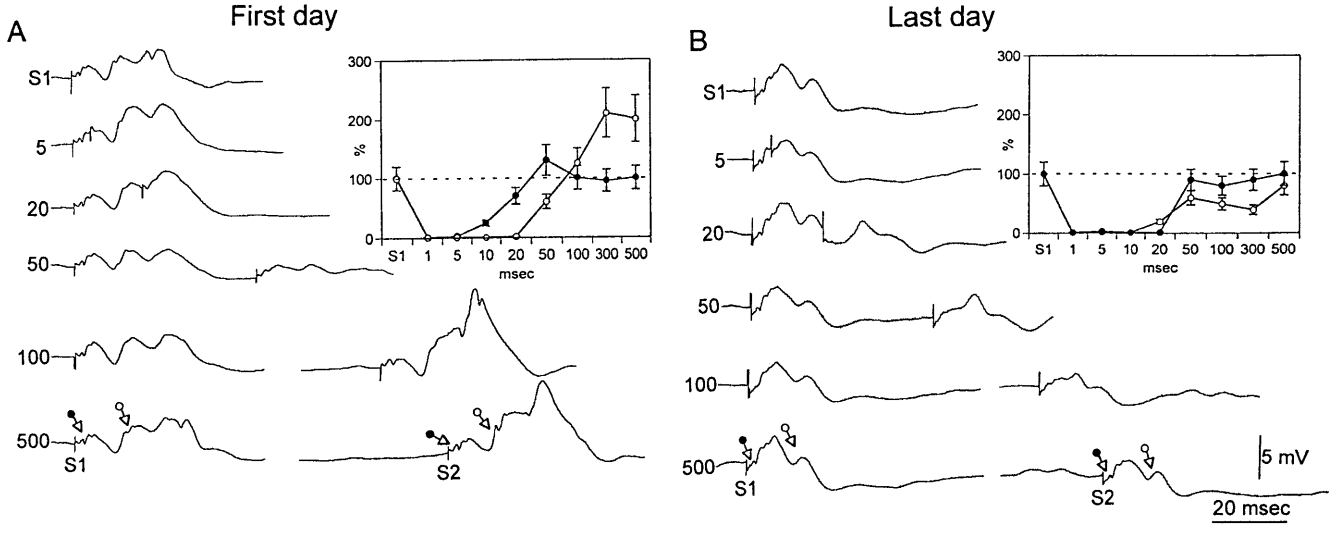

Table 1 Number of spontaneous behavioral seizures before and during kindling in KA treated rats

Rat no.	Before kindling	During kindling
KA01	4	1
KA02	4	0
KA03	2	1
KA04	0	1
KA05	2	1
KA06	2	0
Total	14	4
KA07	1	1
KA08	1	0
KA09	1	2
Total	3	3

P=0.04 for KA01–KA06, Mann-Whitney test; differences are not significant for KA07–KA09

Discussion

This paper investigates the consequences of ipsilateral perforant path kindling in hippocampal KA-treated rats with recurrent spontaneous seizures. The primary findings were that: (a) subsequent stimulation resulted in an increase in the AD threshold and (b) continuation of kindling suppressed recurrence of spontaneous seizures. These interesting observations presumably represent an interaction between kindling stimuli and the structurally and functionally altered limbic neuronal substrate which has been induced by intrahippocampal KA injection. Neuronal loss following KA injection is known to initiate a complex series of changes including granule cell deafferentation, dispersion, and neurogenesis, as well as mossy fiber and interneuron sprouting (Ben-Ari et al. 1979; Cronin and Dudek 1988; Davenport et al. 1990; Mathern et al. 1993). These network changes are accompanied by changes in the properties of NMDA and GABA receptors (Tasker and Dudek 1991; McNamara 1994; Mody 1999) and monoaminoergic and peptidergic neuromodulators (Simmons and Murphy 1995; Simonato and Romualdi 1996).

Seizure prompting in KA-treated rats

We have described previously (Bragin et al. 1999b) that two types of spontaneous seizures occur in this model of epilepsy. Type one seizures have hypersynchronous onset and hippocampal origin. Our data in naive rats are in accordance with several previous studies in which low frequency stimulation was shown to be effective in kindling limbic seizures (Goddard et al. 1969; Cain and Corcoran 1981; Racine and Burnham 1984). The fact that KA-treated rats initially responded to this kindling stimulation with stage 4 or 5 seizures suggests an overlap between the population of neurons responsible for spontaneous seizures in the KA model and the population recruited during kindling (Burchfiel et al. 1982). Because of this full-blown response to the first stimulation, "kindling" in the classical sense is meaningless in KA rats. The severe behavioral component accompanying the AD after the first stimulation shows that KA-treated rats are "seizure prompt" as described by Feldblum and Ackermann (1987), who observed that a decreased number of kindling stimulations were required to reach a stage 5 seizure 2–6 weeks after KA treatment.

Afterdischarge threshold during kindling in KA-treated rats

Paradoxically, we found a progressively increasing AD threshold on subsequent stimulations. This increase in AD threshold indicates that the long-term consequences of kindling stimulation in rats with spontaneous seizures differ from those in naive rats (Goddard et al. 1969; Racine 1972b). One possible reason for this difference might be that the epileptic brain develops natural protective mechanisms to suppress seizure activity. Such mechanisms may increase the AD threshold and decrease AD duration in the hippocampal formation following high intensity stimulation. However, once an AD occurs it propagates to produce a stage 3–5 behavioral seizure. After spontaneous seizures or electrically evoked AD, there is a suppression of excitability in the brain which prevents frequent recurrence of seizures (Stripling and Russell 1989). This effect is more prominent in fully kindled animals and may last for several days (Goddard et al. 1969; Mucha and Pinel 1977). Likewise, in our model of epilepsy, the brain appears to have properties similar to those observed in fully kindled rats. It is unlikely that the occurrence of spontaneous seizures between kindling stimulations in the KA rats contributed to the increase in AD thresholds because a total of only four spontaneous seizures occurred in all KA rats during the 2nd week of the kindling period (see Table 1).

Our 5-Hz stimulation parameters were similar to those used to produce the phenomenon of "quenching" described by Weiss et al. (1998), in which low frequency stimulation applied after kindling stimulation led to an increase in AD threshold and to inhibition of kindled seizures (Weiss et al. 1995). These investigators found that the stimulating frequencies which cause "quenching" of kindling are between 2 and 10 Hz, indicating that both kindling and KA excitotoxically induced epileptic manifestations can be modulated by low frequency trains of stimulation. The fact that 5 Hz stimulation causes kindling in naive rats and has a seizure suppressing effect in KA-treated rats probably reflects features of the altered circuitry induced by KA injection that act to homeostatically prevent continuous seizure generation. In order to pursue the electrophysiological characteristics of this effect we observed the following electrophysiological correlates of increase in AD threshold: lack of frequency facilitation during rhythmic electrical stimulation, increase in paired pulse inhibition at short interstimulus intervals,

and depression of the late components of the perforant path evoked potential.

High frequency tetanus leads to long-term potentiation of synaptic efficacy in the hippocampal formation (see reviews: Bliss and Collingridge 1993; Malenka and Nicoll 1999), and, similarly, facilitation of synaptic transmission has been shown during kindling (Racine 1972b; Maru and Goddard 1987a). On the basis of these results, frequency facilitation followed by a decrease in the AD threshold was expected in KA rats during repetitive electrical stimulation. In normal rats, kindling leads to potentiation of both excitatory and inhibitory pathways (Maru and Goddard 1987a, 1987b); however, facilitation of excitatory synapses dominates and leads to progressive kindling. In our experiments, using perforant path single pulse stimulation (intervals of 10 s or greater) there was no significant change in the amplitude of DG population EPSP or population spike responses over the course of kindling, suggesting that excitability remained unchanged. In contrast, there was an initial facilitation of DG population spikes and fEPSPs during 5 Hz stimulation (Fig. 3), which was prominent on the 1st day of kindling but no longer occurred as 5 Hz stimulation was repeated over the period of kindling.

An increase in paired pulse depression of excitatory responses which may last for weeks commonly occurs during kindling (Tuff et al. 1983; Maru and Goddard 1987b) and it is present in KA-treated rats (Buckmaster and Dudek 1997), as well as in patients with temporal lobe epilepsy (Wilson et al. 1998). However, it does not prevent the development of kindling in naive rats or the occurrence of spontaneous seizures in chronically epileptic rats or patients. Paradoxically, the late polysynaptic component of the evoked potentials during paired-pulse testing at the 100-, 500-ms intervals was facilitated on the 1st day of kindling and became suppressed over the course of kindling (Fig. 4). This loss of facilitation may be due to restoration of the inhibitory DG gate function within entorhinal-hippocampal circuitry, which has previously been shown to deteriorate in the epileptic brain (Stringer and Lothman 1989; Gloveli et al. 1997).

Long-term depression (LTD) in stimulated pathways might be another mechanism that acts to suppress the kindling process. We did not find a decrease in excitability of the perforant path as measured by the amplitude of early evoked potentials during consecutive kindling stimulations. Therefore, LTD as it is described in in vitro experiments (Cummings et al. 1996; Oliet et al. 1996) is probably not responsible for an increase in AD threshold.

Suppression of spontaneous seizures

It would appear that the seizure-induced, seizure-suppressing mechanisms which are responsible for an increase in AD threshold during kindling in KA-treated rats also act to reduce the generation of spontaneous seizures. It is possible that daily stimulation, or daily AD induction, activates those endogenous mechanisms in KA rats which maintain the interictal state, thereby suppressing spontaneous seizures.

These data are in agreement with recent observations that recurrent electrical hippocampal stimulation in patients with mesial temporal lobe epilepsy can suppress generation of spontaneous temporal lobe seizures (Velasco et al. 2000). Further studies of this experimental model to elucidate the mechanism underlying stimulation-induced seizure suppression will be important in determining how and when to apply hippocampal stimulation as an alternative clinical treatment.

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