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Anticonvulsant effects of sevoflurane on amygdaloid kindling and bicuculline-induced seizures in cats: comparison with isoflurane and halothane

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Abstract

Purpose. We compared the anticonvulsant effects of sevoflurane with those of isoflurane and halothane in amygdaloid kindling and bicuculline-induced seizures in cats.

Methods. In a crossover design, the effects of 70% nitrous oxide, and 0.3, 0.6, and 1.5 minimum alveolar concentration (MAC) of volatile anesthetics were studied in five cats in which the amygdala was electrically stimulated at the current used for establishing the kindled state. The effects of 0.6 and 1.5 MAC of volatile anesthetics were studied in another five cats, in which 0.2 mg·kg^{-1} of bicuculline was administered IV.

Results. In the amygdaloid kindling model, all four anesthetics decreased the duration of after-discharge (AD), the rise of multiunit activity in midbrain reticular formation (R-MUA), and the behavior scores compared with findings without anesthetics. Halothane, at 1.5 MAC, significantly decreased the number of cats showing AD ($P < 0.05$). In the bicuculline-induced seizure model, all five cats showed repetitive spikes during 1.5 MAC of sevoflurane, whereas only two and three cats, respectively, showed the repetitive spikes during 1.5 MAC of isoflurane and halothane. All three volatile anesthetics decreased the rise of R-MUA, the duration of the repetitive spikes, and the behavior scores. The suppression of the rise in R-MUA and the behavior scores with 1.5 MAC of sevoflurane was significantly less than that with 1.5 MAC of isoflurane.

Conclusion. The anticonvulsant effects of sevoflurane were less potent than those of halothane in the amygdaloid kindling model and less potent than those of isoflurane in the bicuculline-induced seizure model.

Key words Amygdaloid kindling · Bicuculline · Halothane · Isoflurane · Sevoflurane

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Introduction

General anesthetics generally have anticonvulsant effects, and a few volatile anesthetics also have proconvulsant effects [1,2]. The proconvulsant properties of enflurane are well known, and its proconvulsant effects may counteract its anticonvulsant effects. However, Oshima et al. [3] showed that enflurane significantly depressed seizure in three different feline epileptic models, and they indicated that enflurane could be used in patients with epileptic disorders. Sevoflurane is widely used, and has neurophysiological properties similar to those of enflurane. Seizure shown on electroencephalograms (EEGs) can be induced by peripheral somatic stimulation performed in cats under sevoflurane anesthesia [4]. Further, there are a few case reports of the intraoperative occurrence of seizure under sevoflurane anesthesia [5,6]. Although Oshima et al. [3] showed enflurane's anticonvulsant effects, they did not compare these effects of enflurane with the anticonvulsant effects of other volatile anesthetics. Therefore, it is not clear whether the anticonvulsant effects of enflurane or sevoflurane are less potent than those of other anesthetics.

We previously compared the anticonvulsant effects of halothane, isoflurane, and sevoflurane on lidocaine- and penicillin-induced seizures in cats [7,8]. Sevoflurane and isoflurane showed more potent anticonvulsant effects than halothane in lidocaine-induced seizure [7], whereas sevoflurane and halothane showed less potent effects than isoflurane in penicillin-induced seizure [8]. Therefore, the potency of anticonvulsant effects can be different when different epileptogenesis is involved, and anesthetics should be selected according to the type of epileptogenesis in the patient. The anticonvulsant effects of volatile anesthetics may not be mediated via an action that is common to different anesthetic agents. General anesthetics have many effects on the central nervous system (CNS), in which neuronal activities

are modulated by divergent neurotransmitters and neuromodulators. Different anesthetics would exert their anticonvulsant effects, mediating these divergent neuronal networks, in different ways.

In the present study, we compared the anticonvulsant effects of sevoflurane with those of isoflurane and halothane on another epileptic model, amygdaloid kindling, in cats; we also compared the anticonvulsant effects of these three anesthetics on bicuculline-induced seizures. Penicillin and bicuculline are antagonists of γ-aminobutyric acid $(GABA)_A$ receptors [9]. Volatile anesthetics potentiate $GABA_A$ receptor activities [10]. Therefore, the stimulatory effects of volatile anesthetics on $GABA_A$ receptors may explain the suppression of seizure induced by $GABA_A$ antagonists. The amygdaloid kindling model of epilepsy is used as a tool for investigating the mechanisms of epilepsy and the efficiency of various anticonvulsant drugs, as well as being used as a model of complex partial epilepsy [11]. We selected these models of epilepsy, because these models are similar to those used in the study of Oshima et al. [3], and we could compare the anticonvulsant effects of sevoflurane with those of enflurane, as well as those of isoflurane and halothane.

Materials and methods

After our institutional committee on animal research had approved the study, we studied ten cats, each weighing 2.5–4.0 kg.

Chronic placement of electrodes

Each animal was anesthetized by placing it in a 50-l plastic container that was filled with 5% sevoflurane in oxygen. Once the animal had lost consciousness, a catheter was inserted into the cephalic vein, and the trachea was intubated after the administration of 1 mg vecuronium IV; each cat was placed in the supine position, and anesthesia was maintained with 3.5% sevoflurane in oxygen. The lungs were mechanically ventilated using a nonrebreathing ventilator (Acoma Animal Respirator, AR-300; Acoma, Tokyo, Japan). A rectal thermistor was inserted, and the rectal temperature was maintained at 37°C–39°C, using a warm-water mattress and a heating lamp. The animal was then placed on a stereotaxic apparatus. Stainless steel screws, 2.0 mm in diameter, were inserted in the frontal bone of the skull (reference electrode) and over the parietal and occipital cortex to record the cortical electroencephalogram (EEG). Parallel stainless steel wire electrodes (0.2 mm in diameter), which were insulated with epoxylite resin, except at the tips, with a vertical separation of 0.5–1.0 mm at the tips, were inserted in the bilateral midbrain reticular formation (A2; L3; H-2, according to the atlas of Snider and Niemer [12]) to record the reticular multi-unit activity (R-MUA). Wire electrodes of the same type were inserted into the bilateral dorsal hippocampus (A2; L8; H9) and the medial amygdala (A12; L9; H-6). The electrodes were connected to a socket that was fixed to the skull with dental cement. The sevoflurane supply was turned off, and the cat was awakened. The experiment was done at least 1 week later. During the experiment, the socket was connected to the recording devices, and an electric stimulator, with a bundle of flexible cables of 1.2 m length, was used to allow for the free movement of the animals.

Administration of test anesthetics

To study the effects of 70% nitrous oxide, and 0.3 and 0.6 minimum alveolar concentration (MAC) of the volatile anesthetics, each cat was placed in a 50-l plastic container into which the study anesthetic gas in oxygen was delivered at $10 l \cdot min^{-1}$. The inspired concentration of each anesthetic in the box was measured by placing the sampling port of an infrared anesthetic gas monitor (Capnomac Ultima, Datex, Finland) near the animal's mouth. The anesthetic gas monitor was calibrated on each experimental day. To study the effects of 1.5 MAC of anesthetic agents, the cat was initially placed in the plastic container, which was filled with 2 MAC of the test agent, and a laryngeal mask airway was inserted for deepening anesthesia [13], then the trachea was intubated; no muscle relaxant was used. The lungs were mechanically ventilated, using the nonrebreathing ventilator. The inspired concentrations of the test anesthetic and the end-tidal partial pressure of carbon dioxide ($ETCO_2$) under mechanical ventilation were measured, using the anesthetic gas monitor, and the $ETCO_2$ was maintained at 30–35 mmHg. The rectal temperature was maintained at 37°C–39°C. One MAC was defined as 2.6% for sevoflurane [14], 1.6% for isoflurane, and 1.2% for halothane [15].

Recording of CNS electrical activities

The cortical and subcortical EEGs were recorded on an eight-channel polygraph (Nihon-Koden polygraph, AB621G; Nihon-Koden, Tokyo, Japan), and the reticular multiunit activity (R-MUA) was recorded on a straight writing oscillograph (Nihon-Koden; WT685G). The firing of a population of reticular neurons was measured using a MUA technique described previously [16]. In brief, the neuronal firing between the two active points of electrodes was obtained, amplified, and then sent to a high-pass filter. Because neuronal firings are high-frequency activities, the obtained signal was rectified and smoothed with an electronic circuit with a

smoothing time constant of 50 ms and was expressed by the oscillation of dc voltage: the higher the dc level, the greater the firing of a population of units. The R-MUA level was measured as the height of the lower limit of the trace from the dc level obtained by input short instead of the animal. Neuronal discharges were picked up from an area of approximately 1 mm radius around the tip of the electrode. The R-MUA level was expressed as the percentage of that observed during the nonanesthetized state in each animal.

Establishment of kindling model

Amygdaloid kindling was established, as described by Oshima et al. [3], in five cats. Electric stimulation of 60 Hz and 1-s duration was given daily through either side of the amygdaloid electrodes, using a constant current stimulator (SEN 3301; Nihon-Koden) and an isolating unit (SS-104J; Nihon-Koden). On the first day, the initial stimulating current was set at 200 μA and the current was increased by 50 μA every 15 min until local after-discharge was induced in the contralateral amygdaloid EEG (Fig. 1A). This current, which produced the local after-discharge in the contralateral amygdala, was used thereafter. From the second experimental day, the amygdala was stimulated once a day. This daily stimulation of the amygdala induced an after-discharge that initially developed in the contralateral amygdala and was propagated to the dorsal hippocampus (Fig. 1B) and then to the cerebral cortex, to form

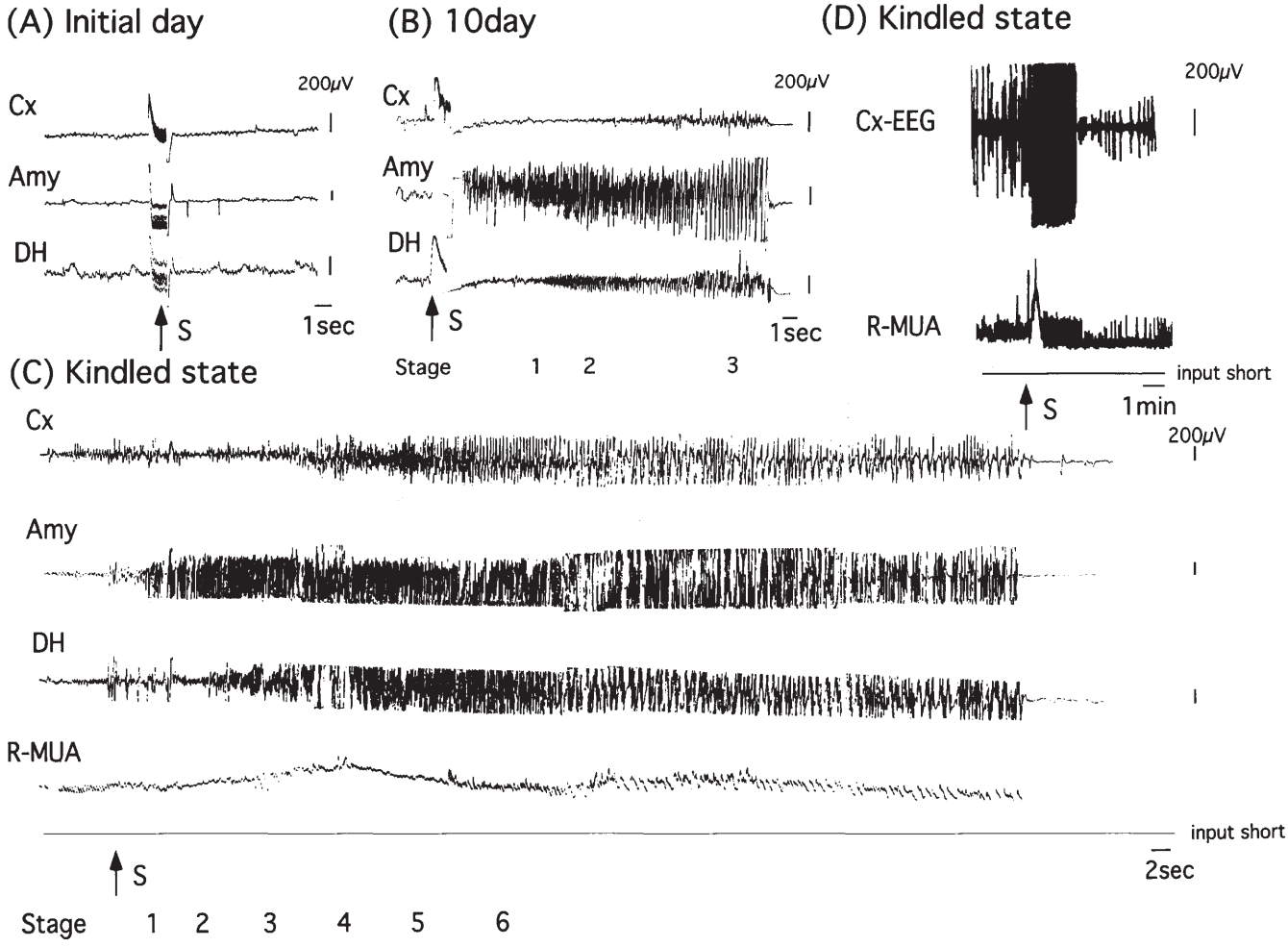

Fig. 1A–D. Amygdaloid kindling model of epilepsy in the cat. *Cx (Cx-EEG)*, Cerebral cortex electroencephalogram; *Amy*, medial amygdala; *DH*, dorsal hippocampus; *R-MUA*, multiunit activity in the midbrain reticular formation; S, electrical stimulation of the amygdala; stage, behavior scores (see text for further details). **A** On the initial day, electrical stimulation of the amygdala induced a short-period after-discharge in the contralateral amygdaloid EEG. **B** On the tenth day, electrical stimulation of the amygdala induced after-discharge that was propagated from the amygdala to the dorsal hippocampus, and the behavior score progressed to 3. **C** In the kindled state, electrical stimulation of the amygdala induced after-discharge that was propagated from the amygdala to the dorsal hippocampus and then to the cortex. With the propagation of the after-discharge, both R-MUA and the behavior score increased. **D** Compressed recordings of cortical EEG and multiunit activity in the midbrain reticular formation in the kindled state

a generalized seizure (Fig. 1C). The behavior also changed with the propagation of the after-discharge, and was scored according to seven stages of convulsion: no change (stage 0), unilateral facial twitching ipsilateral to the stimulation (stage 1), bilateral facial twitching (stage 2), head nodding (state 3), contralateral head turning with tonic extension of contralateral forepaw and circling (stage 4), generalized clonic jerking (stage 5), and, finally, generalized convulsion (stage 6). We defined that the so-called kindled state was established when this generalization of the after-discharge and convulsion were induced consistently on successive 5-day trials [3].

Amygdaloid kindling model

Five kindled cats were used for comparing the anticonvulsant effects of 70% nitrous oxide, and 0.3, 0.6, and 1.5 MAC of sevoflurane, isoflurane, and halothane. After exposure to the test anesthetic agent for 30 min, the electrical stimulus was applied to the amygdala. The control response to stimulation was obtained during inhalation of room air. Anticonvulsant effects of the anesthetics were evaluated by the number of cats showing after-discharge, the duration of the after-discharge, the rise of R-MUA after stimulation, and the behavior of the animal, scored according to the seven stages of convulsion, as outlined for the process of establishing the kindling model. Each cat was tested to study the effects of 70% nitrous oxide and the three different concentrations of each of the three test anesthetics in a randomized, cross-over design with an interval of at least 7 days between each study.

Bicuculline-induced seizure

In another five cats with chronically implanted brain electrodes, we studied the effects of 0.6 and 1.5 MAC of the three anesthetics on a bicuculline-induced seizure. Each animal was anesthetized with 3% of sevoflurane, and a catheter was inserted in the cephalic vein. Sevoflurane was discontinued. One h later, full recovery from anesthesia was confirmed by CNS electric activities and behavior. A bicuculline solution, of 0.4 mg·ml^{-1}, was made each day by dissolving this agent in normal saline; the pH of the solution was adjusted to 5.3. A control response to intravenous bicuculline, 0.2 mg·kg^{-1}, was obtained during the inhalation of room air. For the study of the volatile anesthetics, the animal was exposed to the test agent for 30 min, and bicuculline, 0.2 mg·kg^{-1}, was administered IV. The anticonvulsant effects of the anesthetics were evaluated according to the number of cats showing repetitive spikes on the EEG after the injection of bicuculline, the duration of the repetitive spikes on the EEG, the rise of R-MUA after the administration of bicuculline, and the behavior of the animal. The behavior of the animal was scored as one of four stages: no change (stage 0), twitching (stage 1), convulsion restricted to the paws (stage 2), and generalized convulsion (stage 3). Each animal was used for studying the effects of the test anesthetics at two different concentrations, in a randomized, cross-over design with an interval of at least 7 days between each study.

Correlations between EEG, R-MUA, and behavior scores

Correlations between the maximum level of R-MUA after electrical stimulation or bicuculline injection, EEG patterns, and behavior scores were investigated.

Statistical analysis [17]

The values are presented as means ± SDs, except for the behavior scores, which are presented as medians and ranges. The spontaneous levels of R-MUA, rise in R-MUA after stimulation, and duration of after-discharge or repetitive spikes were tested with repeated measures analysis of variance (ANOVA), followed by the Newman-Keuls test. The occurrence of after-discharge or repetitive spikes was tested by χ^2 contingency table analysis. Behavior scores were tested using the Kruskal-Wallis test. Correlations between the rise of R-MUA and the behavior score were studied using Spearman's rank correlation. Differences were considered statistically significant at $P < 0.05$.

Results

Amygdaloid kindled model

The kindling process was completed at 25 to 36 days (mean, 28 days). The duration of after-discharge ranged from 58 to 111 s (88 ± 21 s) and the maximum level of R-MUA during after-discharge was 391 ± 93% of that of the pre-stimulation period in the kindled state.

Before electrical stimulation, nitrous oxide did not alter the R-MUA level, whereas all volatile anesthetics depressed it (Table 1). The depression of R-MUA by sevoflurane and isoflurane was significantly greater than that by halothane at 1.5 MAC ($P < 0.05$).

All test anesthetics suppressed the seizure in the amygdaloid kindled cats (Table 1). Although electrical stimulation induced the after-discharge in all cats with 70% nitrous oxide and 0.3 MAC of the volatile anesthetics, the after-discharge was not induced in several cats with 0.6 or 1.5 MAC of the volatile anesthetics,

Table 1. Effects of anesthetics in five amygdaloid kindled cats

	Control	70% N2O	MAC	Sevoflurane	Isoflurane	Halothane
R-MUA before stimulation (%)	100	112 ± 11	0.3	79 ± 10	83 ± 8	90 ± 3
			0.6	69 ± 16*	70 ± 5*	85 ± 9
			1.5	35 ± 16*†‡	43 ± 14*†‡	62 ± 24*
Number of cats showing after-discharge	5	5	0.3	5	5	5
			0.6	3	2	3
			1.5	3	3	1*
Duration of after-discharge(s)	88 ± 21	18 ± 10*	0.3	17 ± 21*	10 ± 12*	17 ± 14*
			0.6	10 ± 17*	3 ± 4*	3 ± 3*
			1.5	12 ± 17*	5 ± 7*	1 ± 2*
R-MUA rise after stimulation (%)	291 ± 93	56 ± 39*	0.3	53 ± 33*	47 ± 20*	42 ± 34*
			0.6	25 ± 25*	14 ± 23*	23 ± 24*
			1.5	20 ± 25*	12 ± 14*	6 ± 13*
Behavior score	6 (6)	2 (1–4)*	0.3	1 (1–4)*	2 (1–4)*	2 (1–4)*
			0.6	0 (0–1)*	0 (0)*	0 (0–1)*
			1.5	0 (0)*	0 (0)*	0 (0)*

* $P < 0.05$ vs control; † $P < 0.05$ vs halothane; ‡ $P < 0.05$ vs 0.3 and 0.6 MAC (minimum alveolar concentration)
Values are means ± SD or median (range)
R-MUA, Neuronal firing in the midbrain reticular formation recorded with multiunit activity

and it was induced in only one cat with 1.5 MAC of halothane ($P < 0.05$). Both the duration of the after-discharge and the R-MUA rise after stimulation were suppressed by all the anesthetics ($P < 0.05$). However, this suppression was not dose-dependent, and no significant difference between agents was shown. The behavior scores were also suppressed, and no convulsive behavior was observed (score 0) with 0.6 MAC of isoflurane or with 1.5 MAC of any of the volatile anesthetics in any cats (Table 1). Representative changes in EEGs and R-MUA induced by 1.5 MAC of each of the volatile anesthetics in same cat are shown in Fig. 2.

Bicuculline-induced seizure model

Bicuculline induced repetitive spikes (seizures) on the EEG and a generalized convulsion (behavior score, 3) in all cats in the control state. The duration of seizure on the EEG was 283 ± 42 s, and the maximum level of R-MUA after bicuculline administration was 436 ± 155% of the pre-bicuculline level.

All the anesthetics significantly suppressed the pre-bicuculline level of R-MUA (Table 2). At 0.6 MAC, there was no significant difference in the effects between anesthetics, whereas at 1.5 MAC, sevoflurane and isoflurane depressed the R-MUA to a greater extent than halothane ($P < 0.05$).

All the anesthetics significantly suppressed the bicuculline-induced seizure (Table 2). Bicuculline failed to induce seizure in several cats under anesthesia, except for that with 1.5 MAC sevoflurane. During the inhalation of 1.5 MAC sevoflurane, repetitive spikes appeared in all cats after the injection of bicuculline. In cats in which repetitive spikes did not appear, sporadic spikes were induced. Representative traces of EEGs and R-MUA in one cat during the inhalation of different anesthetics are shown in Fig. 3. The duration of repetitive spikes was significantly shortened by all anesthetics ($P < 0.001$); there was no significant difference between the anesthetics. The R-MUA rise after bicuculline was significantly suppressed by all anesthetics, and the suppression during inhalation of 1.5 MAC sevoflurane was less than that with 1.5 MAC isoflurane ($P < 0.05$). All anesthetics suppressed the behavior scores, and isoflurane's suppression at 1.5 MAC was greater than that of sevoflurane and halothane.

Correlations between EEG, R-MUA, and behavior scores

The correlation between the maximum R-MUA and behavior scores is shown in Fig. 4. The behavior scores were correlated with maximum R-MUA both in the amygdaloid kindling ($R = 0.888$; $P < 0.001$) and in the bicuculline-induced seizure models ($R = 0.712$; $P < 0.001$). In the amygdaloid kindling model, the correlation between the propagation of after-discharge on EEGs and behavior scores was the same as that shown during the process of establishing the kindling state. In the bicuculline-induced seizure model, the following correlations between behavior score and EEG were shown: behavior score 0 was associated with no change or sporadic spikes on EEG; behavior score 1 was associated with sporadic spikes on EEG; and behavior score 2 or 3 was associated with repetitive spikes on EEG.

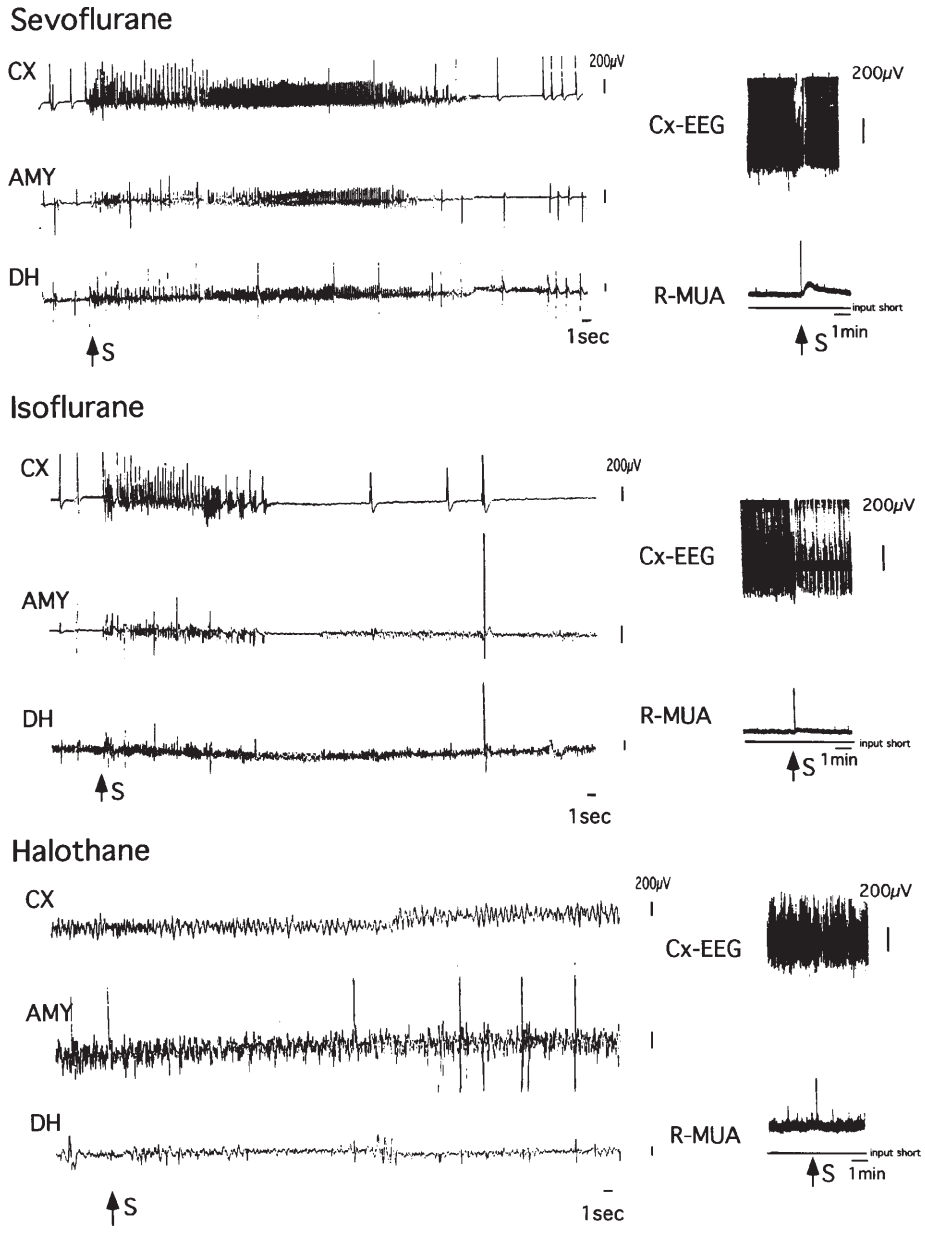

Fig. 2. Effects of anesthetics on amygdaloid kindling model in a cat. *Cx*, Cerebral cortex; *Amy*, medial amygdala; *DH*, dorsal hippocampus; *R-MUA*, multiunit activity in the midbrain reticular formation; *Cx-EEG*, cortical electroencephalogram. *Arrow(s)* indicate electrical stimulation of amygdala. Anesthetics were inhaled at 1.5 MAC in oxygen for 30 min and then the amygdala was stimulated. In the *left traces*, the propagation of after-discharge differs between anesthetics; sevoflurane shortened the duration of after-discharge compared with the control value (*see Fig. 1C*), but its effect was less than that of isoflurane; halothane suppressed the appearance of the after-discharge. Note that sporadic spontaneous spikes before the stimulation are shown at all recording sites for sevoflurane and isoflurane, but only in the amygdala for halothane. In the *right traces*, the effects of the anesthetics on the R-MUA are shown. The suppression of the R-MUA after amygdaloid stimulation in this cat was least for sevoflurane, greatest for halothane, and intermediate for isoflurane

Discussion

The present study showed that sevoflurane had anticonvulsant effects similar to those of halothane and isoflurane on both amygdaloid kindling and bicuculline-induced seizure models in cats. Differences between the volatile anesthetics in the anticonvulsant effects were shown in the occurrence of after-discharge in the kindling model, and in the suppression of the R-MUA rise after bicuculline, and in suppression of behavior scores in the bicuculline-induced seizure model. The most potent anticonvulsant effects seemed to be exerted by halothane in the amygdaloid kindling model and by isoflurane in the bicuculline-induced seizure model.

In the amygdaloid kindling model of epilepsy, 1.5 MAC halothane had the greatest effect in decreasing the number of cats showing after-discharge. Halothane suppressed the background level of R-MUA with less potency than sevoflurane and isoflurane. This indicates that the potencies of the suppressive effect on neuronal firing in spontaneous firing and firing in response to a given stimulus are not correlated. Similar potent suppressive effects of halothane on the response capability of the brain were also shown in our prior study of responses to electrical sciatic nerve stimulation in cats [18]. Although in the present study, halothane suppressed the background EEGs and R-MUA with less potency than isoflurane and sevoflurane, in the previous

Table 2. Effects of anesthetics on bicuculline-induced seizure in cats

	Control	MAC	Sevoflurane	Isoflurane	Halothane
Pre-bicuculline R-MUA (%)	100	0.6	70 ± 12*	67 ± 18*	77 ± 16*
		1.5	34 ± 13*†‡	29 ± 21*†‡	64 ± 20*
Numbers of cats showing seizure after bicuculline	5	0.6	4	3	4
		1.5	5	2	3
Duration of seizure (s)	283 ± 42	0.6	35 ± 33*	23 ± 32*	43 ± 32*
		1.5	40 ± 10*	12 ± 17*	22 ± 22*
R-MUA rise after bicuculline (%)	336 ± 155	0.6	155 ± 97*	78 ± 106*	179 ± 100*
		1.5	109 ± 38*§	18 ± 36*	58 ± 59*
Behavior score	3 (3)	0.6	3 (0–3)*	2 (0–2)*	3 (0–3)*
		1.5	2 (2)*§	1 (0–2)*	2 (1–2)*§

* $P < 0.05$ vs control; † $P < 0.05$ vs halothane; § $P < 0.05$ vs isoflurane; ‡ $P < 0.05$ vs 0.6 MAC
Values are means ± SD or median (range)
R-MUA, Neuronal firing in the midbrain reticular formation recorded with multiunit activity

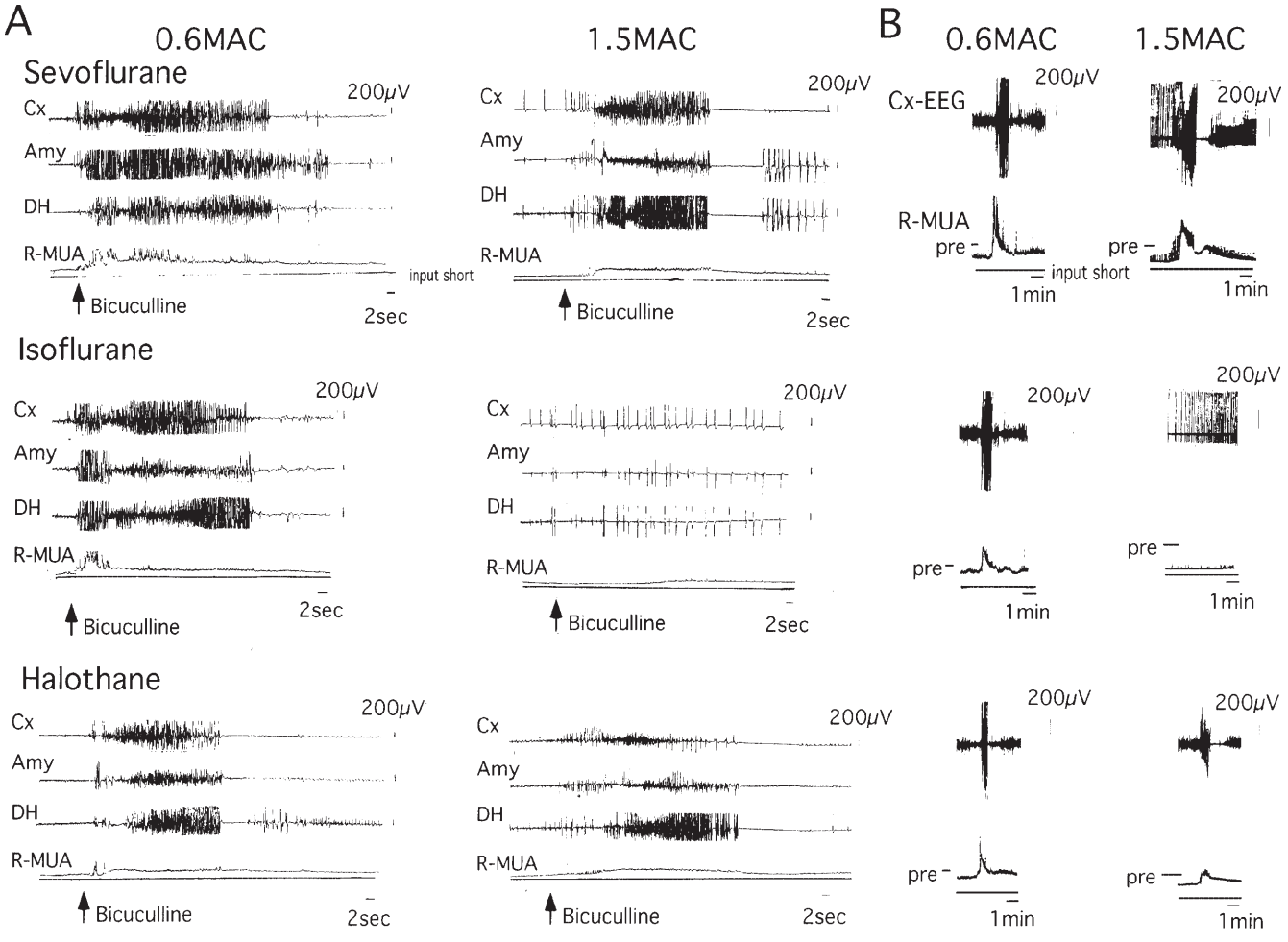

Fig. 3A,B. Effects of anesthetics on bicuculline-induced seizure in a cat. Note differences in recording paper speeds between A and B. *Cx*, Cerebral cortex; *Amy*, medial amygdala; *DH*, dorsal hippocampus; *Cx-EEG*, cortical electroencephalogram; *pre*, R-MUA level before inhalation of anesthetics. Seizure was induced by 0.2 mg·kg^{-1} bicuculline IV during the inhalation of 0.6 or 1.5 minimum alveolar concentration (*MAC*) of the anesthetics in oxygen. Anesthetics suppressed both the bicuculline-induced seizure on the EEG and the rise in R-MUA; 1.5 MAC isoflurane suppressed the development of seizure in this cat

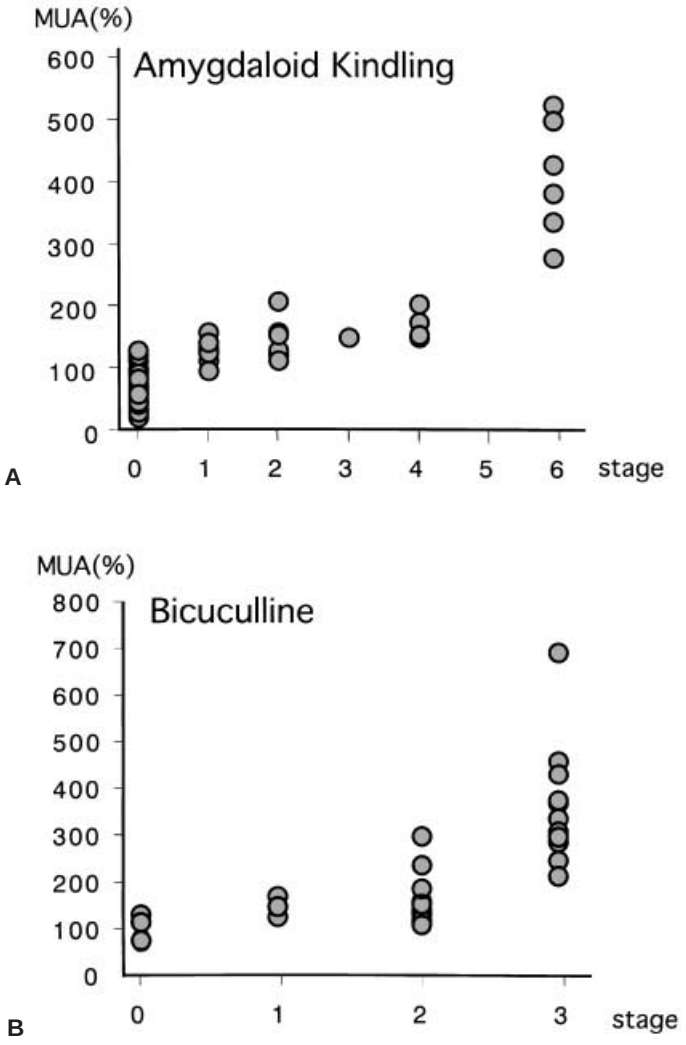

Fig. 4A,B. Correlations between maximum R-MUA and behavior scores in amygdaloid (**A**) kindling and bicuculline-induced seizure (**B**) in cats. *R-MUA*, Maximum multiunit activity in the midbrain reticular formation after amygdaloid stimulation or injection of 0.2 mg·kg^{-1} bicuculline IV; *stage*, behavioral score after stimulation or injection of bicuculline (see text for further details). There are correlations between maximum R-MUA and behavior scores in both the amygdaloid kindling model (**A**) and the bicuculline-induced seizure model (**B**)

study, halothane suppressed responses to electrical sciatic nerve stimulation in terms of both R-MUA rise and arterial blood pressure, to a greater extent than isoflurane or sevoflurane [18]. In the present study, nitrous oxide, at 70%, showed anticonvulsant effects similar to those seen with 0.3 MAC of the volatile anesthetics. The MAC value of nitrous oxide in cats is reported to be 225% [19]. This indicates that the anticonvulsant effects of nitrous oxide have a potency similar to those of volatile anesthetics in this epileptic model in cats.

In the bicuculline-induced seizure model, while increasing concentrations of isoflurane and halothane decreased the number of cats showing repetitive spikes after bicuculline injection, all cats showed repetitive spikes after the injection of bicuculline during the inhalation of 1.5 MAC sevoflurane, and the suppression of the R-MUA rise after bicuculline during the inhalation of 1.5 MAC sevoflurane was less than that seen with isoflurane. These findings indicate that sevoflurane has a proconvulsant effect, which partially masks the anticonvulsant effects. This result is consistent with our previous report of the proconvulsant effects of large concentrations of sevoflurane in cats [4]. Isoflurane also induced spontaneous sporadic spikes on EEGs in cats [18,20], but it did not induce photic stimulation-induced spikes on EEG nor did it augment the photic stimulation-evoked potential in the visual cortex [20]. Therefore, sevoflurane, but not isoflurane, has a proconvulsant effect, and its proconvulsant effect may mask the anticonvulsant effects in this model of epilepsy. The potencies of the anticonvulsant effects of the three anesthetics tested in the present study on the bicuculline-induced seizure model were consistent with those in status epilepticus in cats induced by penicillin [8], which is also an antagonist for $GABA_A$ receptors. Although volatile anesthetics enhance $GABA_A$ receptor activities [10], we could not find any report that compared the enhancing potencies of halothane, isoflurane, and sevoflurane. Therefore, we could not investigate any correlation between the potency of the anticonvulsant effects and the potency of the $GABA_A$ receptor enhancing effects of volatile anesthetics.

Oshima et al. [3] demonstrated the anticonvulsant effects of enflurane on penicillin- and bicuculline-induced seizure models and an amygdaloid kindling model in cats. Enflurane depressed seizures, even at a convulsive dose (3.5%). However, the anticonvulsant effects on the amygdaloid kindling models were biphasic, and 1.5% enflurane showed more potent anticonvulsant effects than 3.5% enflurane. The potency of the anticonvulsant effects of 1.5% enflurane in the study of Oshima et al. [3] were similar to the anticonvulsant effects of 0.6 MAC of sevoflurane in our study, whereas 3.5% enflurane in their study was less potent than 1.5 MAC of sevoflurane in ours. These results suggest that 3.5% enflurane produces a more potent proconvulsive effect (and counteracts the anticonvulsant effects) than 1.5 MAC sevoflurane. In fact, in another study, seizure was induced in all cats during deep enflurane anesthesia, but in only 2 of 11 during sevoflurane anesthesia, with the augmenting effects of sevoflurane on the somato-evoked potential being less than those of enflurane [4]. Therefore, the proconvulsant effects of 1.5 MAC sevoflurane are less than those of 3.5% enflurane, and the anticonvulsant

effects of sevoflurane are greater than those of enflurane.

In our study, the behavior scores were correlated with the maximum levels of R-MUA (the level before electrical stimulation or bicuculline plus the rise in R-MUA) in both seizure models. Anesthetics may suppress behavior not only by their effects on brain activities but also by suppressing motor systems. Although volatile anesthetics suppress activities in spinal motoneurons [21–24], the present study showed that brain activities recorded by R-MUA correlated with behavior scores. Therefore, the neuronal firing level in the brain may be a major factor in affecting the behavior scores during convulsion. However, in the present study, we could not rule out the possibility that ascending volleys caused by muscle contraction could have affected the R-MUA.

The present study was performed with animals under spontaneous respiration for the investigation of the effects of nitrous oxide and the effects of 0.3 and 0.6 MAC of volatile anesthetics, while the effects of 1.5 MAC of the volatile anesthetics were studied in animals under mechanical ventilation. Anesthetic suppression of respiration during 0.3 and 0.6 MAC may induce hypercapnia during spontaneous respiration, and hypercapnia has anticonvulsant effects [25]. Therefore, it is possible that, in our study, the anticonvulsant actions of the lower concentrations of volatile anesthetics, and these actions of nitrous oxide could have been overestimated, because hypercapnia could have been involved in these effects.

We investigated the effects of nitrous oxide in the amygdaloid kindling model, but not in the bicuculline-induced seizure model. In the amygdaloid kindling model, nitrous oxide showed an anticonvulsant potency similar to that of the volatile anesthetics. However, we can not conclude that our findings on the anticonvulsant potency of nitrous oxide would apply to other seizure models, because nitrous oxide has recently been reported to have anti *N*-methyl-D-aspartate (NMDA) receptor properties [26], which could affect its anticonvulsant effects in different seizure models. The anticonvulsant properties of nitrous oxide should be investigated in future studies.

The present study indicates that any of the three volatile anesthetics that we tested, including sevoflurane, can be used as anticonvulsants in patients with epileptic disorders. However, the proconvulsant effects of a large concentration of sevoflurane may partially counteract its anticonvulsant effects; the proconvulsant effects of sevoflurane are less potent than those of enflurane and its counteracting effects may also be less potent than those of enflurane.

In conclusion, sevoflurane has potent anticonvulsant effects in amygdaloid kindling and bicuculline-induced seizure models in cats. The anticonvulsant effects of sevoflurane were less potent than those of halothane in the amygdaloid kindling model and less potent than those of isoflurane in the bicuculline-induced seizure model.

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