PD in the three genetically and geographically isolated foci remain to be determined. Preliminary analyses of environmental samples indicate that each of these regions have soils and local water supplies that are rich in aluminum but are virtually devoid of calcium and magnesium (*5–7*).

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19 April 1982; revised 22 July 1982

General Anesthetics Hyperpolarize Neurons in the Vertebrate Central Nervous System

Abstract. *The effect of general anesthetics on frog motoneurons and rat hippocampal pyramidal cells was examined with sucrose gap and intracellular recording, respectively. A number of volatile and intravenous anesthetics directly hyperpolarized the motoneurons. The potency of these agents in hyperpolarizing motoneurons was strongly correlated with their anesthetic potency. While the responses to barbiturates and α-chloralose were blocked by γ-aminobutyric acid antagonists and were dependent on the chloride gradient, the responses to all the other anesthetics tested were generated by a separate mechanism. Intracellular recording from hippocampal pyramidal cells suggested that an increase in potassium conductance accounts for these responses. Such a nonsynaptic action would contribute to the decreased neuronal responsiveness observed for these compounds and thus to their anesthetic action.*

General anesthetics have been reported to have numerous synaptic effects in the central nervous system (CNS). In general, it has been found that synaptic excitation is depressed while synaptic inhibition is preserved or augmented (*1–3*). While physiological studies have focused primarily on synaptic transmission, there is some evidence that anesthetics can have nonsynaptic effects on the postsynaptic membrane. Barbiturates act directly on vertebrate neurons in a manner similar to that of the inhibitory transmitter γ-aminobutyric acid (GABA) (*2–5*). Evidence from studies of invertebrates (*6*), myelinated nerves (*7*), and artificial membranes (*8*) suggests that general anesthetics may increase the permeability of membranes to potassium. However, except for a recent report on ethanol (*9*), such an effect has not been demonstrated for vertebrate CNS neurons. We report here that a number of general anesthetics hyperpolarize central neurons and that this response appears to be due to an increase in potassium permeability.

Thirty-three experiments were performed on isolated frog spinal cords (*10*). Frogs (*Rana pipiens*) were chilled on ice and their spinal cords were removed, hemisected, and placed in a sucrose gap chamber. The activity of motoneurons or primary afferents was recorded by placing the ventral or dorsal root across the sucrose gap, and the potential difference across the gap was monitored with two calomel electrodes. Intracellular recording experiments were also done on CA1 pyramidal cells from rat hippocampal slices (*11*).

Ether caused a slow, dose-dependent hyperpolarization at concentrations similar to that required to induce anesthesia (Fig 1A). The maximum hyperpolarization was modest, rarely exceeding 2 mV. In the same preparations GABA caused large hyperpolarizations (up to 8 mV) and the maximum responses for pentobarbital and α-chloralose approached the size of the GABA response (*4*). Several anesthetics were tested on the frog motoneurons; all were found to cause a hyperpolarization. In Fig. 1B the minimum effective concentration of these anesthetics is graphed against the anesthetic concentration. There is a strong correlation (r = .90) between the hyperpolarizing action and the clinical effect of these agents; furthermore, the concentrations producing these actions are similar to the anesthetic concentrations (*12*). In four preparations the local anesthetic procaine, in concentrations up to 5 m*M*, did not elicit a hyperpolarization. Xylocaine (5 m*M*), another local anesthetic, caused a small hyperpolarization (0.7 mV) in two of three preparations.

It has been reported that both barbiturates and α-chloralose have a GABA-like effect on frog motoneurons (*4*). However, neither picrotoxin nor bicuculline, which block the action of GABA, affected the response to ether (Fig. 2A). The responses to the other anesthetics were also insensitive to GABA antagonists. Changing the chloride gradient across the motoneuron either with ammonium chloride, which blocks chloride extrusion (Fig. 2A) (*13*), or by reducing extracellular chloride (Fig. 2A) did not reduce the response to ether but did reduce or abolish the hyperpolarizing response to GABA. These results suggest that ether and the other anesthetics tested do not hyperpolarize motoneurons by increasing chloride conductance. This conclusion is supported by sucrose gap recordings from frog primary afferent fibers. At this site, GABA had a depolarizing action, presumably because of a reversed (depolarizing) chloride gradient (*14*). While pentobarbital (*15*) and GABA depolarized primary afferents, ether had a hyperpolarizing action (Fig. 2B). All the other anesthetics included in Fig. 1B also hyperpolarized primary afferents except for α-chloralose and phenobarbital, which produced depolarizations.

We studied the effects of ether and

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halothane on rat hippocampal pyramidal cells in order to examine the ionic mechanism in more detail and extend our observations to mammalian cortical neurons. Ether (20 to 40 m*M*) was applied 17 times to ten neurons and halothane (1 to 4 m*M*) was applied 11 times to eight neurons. Both anesthetics caused a moderate hyperpolarization (3.7 ± 2.7 mV for ether; 4.7 ± 2.6 mV for halothane) (Fig. 2C). The hyperpolarization was associated with a decrease in membrane input resistance, as measured by the response to hyperpolarizing constant current pulses (Fig. 2C). This conductance change was not a result of a voltage-dependent change in resistance because, when the membrane was passively hyperpolarized to the same extent with current through the microelectrode, no change in conductance occurred. Since these responses were recorded in the presence of tetrodotoxin to minimize indirect synaptic effects and with a chloride-filled electrode to rule out chloride-dependent hyperpolarizations, the most likely explanation for this response is that the anesthetics increase the permeability of cell membranes to potassium ions.

Some years ago, Krnjevic (*16*) proposed that general anesthetics might activate a calcium-dependent potassium conductance by releasing calcium from intracellular sites. In support of this hypothesis, Carlen *et al.* (*9*) found that ethanol augments the calcium-activated potassium response elicited by depolarizing current pulses. We did not find this effect for halothane or ether, and with high concentrations the response was depressed. However, this negative evidence certainly does not exclude the possibility that such a mechanism is responsible for the hyperpolarizations seen in the present study. Indeed, it was recently shown that high intracellular calcium can depress calcium-activated potassium currents (*17*).

Although barbiturates and α-chloralose differ from the other anesthetics tested in this study in that they have a GABA-like action, it is likely that they also increase potassium conductance. On frog motoneurons these compounds often elicit small hyperpolarizations in the presence of GABA antagonists. However, we have not entirely dismissed the possibility that such responses result from incomplete blockade of GABA receptors.

In summary, we have found that a variety of general anesthetics hyperpolarize vertebrate CNS neurons by increasing potassium conductance. The potency of these drugs for this action is strongly correlated with their anesthetic potency. Such a nonsynaptic action, combined with the well-documented changes in excitatory and inhibitory synaptic transmission, would result in decreased neuronal excitability and thus to a decreased responsiveness of the CNS.

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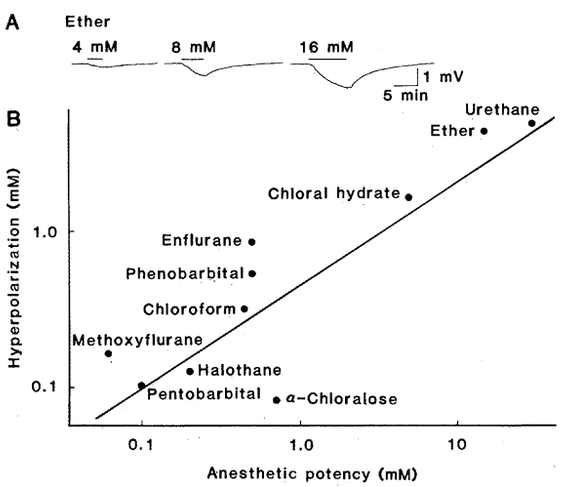

Fig. 1. Hyperpolarization of spinal motoneurons by general anesthetics. (A) Pen recording of responses from a ventral root (sucrose gap recording). A downward deflection indicates that the bath electrode is positive to the root electrode, signifying a hyperpolarization of the motoneurons. (B) Minimum effective hyperpolarizing concentration of the anesthetic plotted against the anesthetic concentration of the drug (*18*). Each point represents the average of three to six experiments. The line was constructed by linear regression.

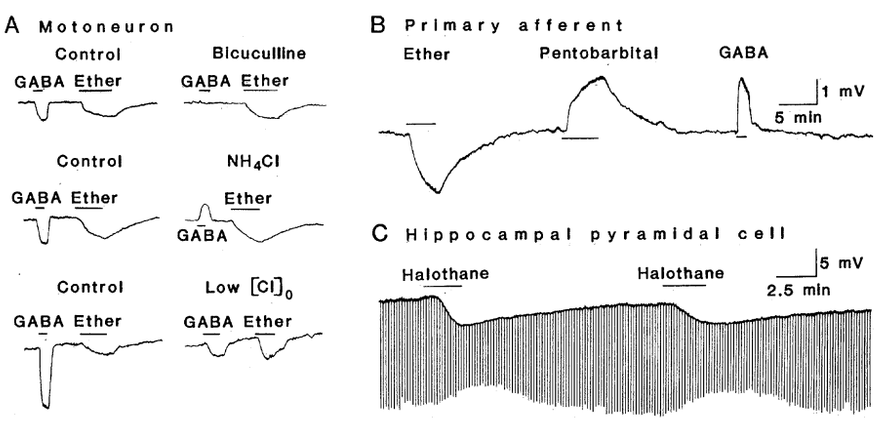

Fig. 2. Properties of hyperpolarizing responses. (A) Sucrose gap records from the ventral root of frog spinal cords, showing that bicuculline methiodide (0.1 m*M*), ammonium chloride (2 m*M*), and low extracellular chloride (10 m*M*) do not reduce the hyperpolarizing response to ether (20 m*M*). Bicuculline was applied for 15 minutes, ammonium chloride for 20 minutes, and the low-chloride Ringer for 8 minutes (GABA concentrations were 20, 60, and 200 μ*M* respectively). (B) Sucrose gap record from the dorsal root of a frog spinal cord, showing that while GABA (0.1 m*M*) and pentobarbital (0.1 m*M*) have a depolarizing action, ether (40 m*M*) is hyperpolarizing. (C) Responses of a rat hippocampal pyramidal cell to halothane (2 m*M*). The recording was obtained with a KCl (3*M*) microelectrode and the preparation was bathed in tetrodotoxin (1 μ*M*). The calibration bar in (B) also applies to the responses shown in (A).

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more than 1 minute before their application. A plexiglass cover was placed on the sucrose gap chamber to completely enclose the compartment containing the spinal cord. Even with these precautions, some loss of anesthetic may well have occurred. Hence the minimum effective concentration would, if anything, be lower than the values reported.
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10 March 1982; revised 24 May 1982

Lizards Infected with Malaria: Physiological and Behavioral Consequences

Abstract. *In northern California, western fence lizards,* Sceloporus occidentalis, *are frequently parasitized by* Plasmodium mexicanum, *which causes malaria. Animals with this naturally occurring malarial infection are anemic; immature erythrocytes in peripheral blood become abundant (1 to 30 percent), and blood hemoglobin concentration decreases 25 percent. Maximal oxygen consumption decreases 15 percent and aerobic scope drops 29 percent in infected lizards; both correlate with blood hemoglobin concentration. Running stamina, but not burst running speed, is reduced in malarious lizards. There is a hierarchical relation between infection with malaria and effects on hematology, physiological function, and behavioral capacity. The results suggest that malarial infection may have significant effects on the ecology of lizard hosts.*

Over the past several decades, lizard models have played a central role in development of modern concepts in population, community, physiological, and behavioral ecology. However, lizard ecologists almost never consider the impact of parasites on individual lizards or lizard populations. This is curious since lizards are hosts of a wide range of parasite taxa (*1*) and parasites frequently have considerable effects on the biology of hosts (*2*). One common group of lizard parasites consists of the malarial organisms (genus *Plasmodium*); indeed, half of the 120 (more or less) described *Plasmodium* species are lizard parasites (*3*). The diversity of lizard malaria parasite-host associations make them ideal systems in which to examine the impact of parasitic infection on physiology and ecology of host organisms.

Although lizard malaria is considered a relatively benign parasitic infection (*1*), several lizard malaria species produce anemia, tissue damage, and even mortality in their hosts (*3–5*). Here we report physiological and behavioral effects of a lizard malaria, produced by *P. mexicanum*, on the host, the western fence lizard, *Sceloporus occidentalis*. Our data demonstrate that hematological alterations resulting from malarial infection are correlated with ecologically important effects on activity metabolism and behavior of the host.

Since 1977 a wild population of *Sceloporus* infected with malaria has been under study at the University of California Hopland Field Station, a tract of foothill oak woodland in southern Mendocino County (*5*). Fence lizards are abundant there and approximately 25 percent of wild adult lizards are infected with malaria at any given time.

As *Plasmodium* infects and reproduces in vertebrate erythrocytes, hematological effects of infection could be an important source of pathology to lizard hosts. Hematological and parasitological variables of field-caught lizards were measured by standard techniques. Blood was drawn from a toe clip and a smear was made for Giemsa staining and examination for parasites (*5, 6*). Parasitemia, expressed as parasites per 10,000 red blood cells (RBC), and the percentage of immature red blood cells (iRBC) were determined by scoring 2000 to 3000 RBC (*7*). Also measured were erythrocyte abundance (RBC per cubic millimeter of blood), hematocrit, and hemoglobin concentration in postorbital sinus blood (*8*).

Lizards infected with malaria respond by rapid production of iRBC (*6, 9, 10*). This response serves to replace cells destroyed by the parasite and possibly to reduce parasite population growth (*5, 11*). Infected *Sceloporus occidentalis* show a marked increase in circulating iRBC (Table 1). Abundance of iRBC ranged from 0 to 2 percent for noninfected and 1 to 30 percent for infected fence lizards. Abundance of iRBC for lizards with very low parasitemia (< 25 per 10,000 RBC) ranged from < 1 to 30 percent. For lizards in which the number of parasites was substantially greater (400 to 2800 per 10,000 RBC), the proportion of iRBC was about 5 to 30 percent. This weak relation between parasitemia levels and percentage of iRBC may be a result of time lags between changes in parasitemia levels and the hemopoietic response. Therefore, the percentage of iRBC appears more likely than parasitemia to be correlated with other physiological effects.

Hemoglobin concentration in blood of parasitized lizards is lower (~ 25 percent less) than in noninfected lizards, but hematocrit and RBC counts do not differ between groups (Table 1). The percentage of iRBC and hemoglobin concentration are negatively correlated ($r = -.51$, $P < .01$, $N = 49$). Thus, hemoglobin deficiency in infected lizards seems to be a result of reduced hemoglobin in iRBC rather than of a decrease in RBC number per volume of blood (*12*).

A 25 percent deficit in blood hemoglobin in malarious lizards should result in a reduction in the ability of the blood to deliver oxygen to tissues. Resting and maximal oxygen consumption were measured (*13*) in adult male *Sceloporus* at 35°C, the preferred body temperature for this species. Blood hemoglobin concentrations were measured within several hours after metabolic measurements. Infected and noninfected lizards do not differ in resting oxygen consumption. However, maximal oxygen consumption and aerobic scope, the increment between resting and maximal oxygen consumption (*14*), differ significantly between groups (Table 1). There is a strong positive relationship between hemoglobin concentration and both maximal oxygen consumption (Fig. 1) and aerobic scope. The fact that data for both infected and noninfected lizards fall on the same regression line shown in Fig. 1

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