

Cerebral Protection by Isoflurane during Hypoxemia or Ischemia

L. A. Newberg, M.D.,* and J. D. Michenfelder, M.D.†

The cerebral metabolic effects of isoflurane suggest that it may provide a degree of cerebral protection similar to that demonstrated for barbiturates. Accordingly, the possible cerebral protection afforded by isoflurane against hypoxemia and ischemia was studied in mice and dogs, respectively. In mice breathing 5% oxygen survival time was increased significantly over control in groups exposed to 1.0% and 1.4% isoflurane. At higher concentrations (2.0% and 3.0%) it is presumed that cardiorespiratory depression contributed to shorter survival times. In six dogs the effects of 3% isoflurane on the rates of cerebral ATP and phosphocreatine depletion and lactate accumulation during incomplete global ischemia were compared with six control dogs exposed to N₂O. Incomplete global ischemia was produced by acute hemorrhagic hypotension to 30 mmHg for 9 minutes, a situation that does not abolish cortical electrical activity (active EEG). In the dogs exposed to isoflurane, the cerebral energy stores of ATP and PCr and the cerebral energy charge were sustained at significantly higher levels than in dogs exposed to N₂O, and the cerebral lactate accumulation was significantly less in the initial 7 minutes of hypotension. It is concluded that in the circumstances of oxygen deprivation insufficient to abolish cortical electrical activity, isoflurane, like the barbiturates, can provide some cerebral protection presumably by depressing cortical electrical activity and cerebral metabolism. (Key words: Anesthetics, volatile: isoflurane. Blood pressure: hypotension. Brain: metabolism; protection. Hemorrhage. Hypoxia. Shock.)

IT HAS BEEN SUGGESTED that the brain might be protected in situations of impaired oxygen delivery if the cerebral oxygen demand could be decreased so as to match the decrease in oxygen supply.¹ The energy requirements of the brain might be viewed as normally subserving two components: that needed for cortical electrical activity, reflected by an active EEG, and that required for the preservation of cellular integrity (*e.g.*, maintenance of ion gradients and biosynthesis). It is postulated that certain anesthetics might provide a degree of cerebral protection, in part, because of their ability to depress or abolish cortical electrical activity, thereby decreasing that component of cerebral oxygen consumption (CMR_{O₂}).¹ However, any such protection would operate only in those circumstances in which cortical activity was still present, such as in incomplete regional or global ischemia. It has been demonstrated that

barbiturates,^{1,2} as well as other anesthetics,³⁻⁵ can provide limited protection in animal models of incomplete ischemia.

Isoflurane has been shown, in dogs, to have an effect on CMR_{O₂} and the cerebral energy state similar to the effect of barbiturates.⁶ It produces a dose-related reduction in CMR_{O₂} that appears to be secondary to a decrease in cortical electrical activity because once an isoelectric EEG has been achieved with 3% isoflurane, there is no further reduction in CMR_{O₂} by increasing the concentration of isoflurane to 6%. Furthermore, at these concentrations of isoflurane, the cerebral energy state remains normal. The purpose of the present study was to investigate possible cerebral protection afforded by isoflurane in two animal models in which it has been demonstrated previously that thiopental provides some protection.^{1,2}

Methods and Materials

MOUSE HYPOXEMIA STUDY

One hundred male ARS HA/ICR albino mice (Sprague Dawley, Madison, Wisconsin) weighing 28-36 g, with free access to food pellets and tap water, were studied by using a hypoxic mouse model and methods previously described.^{2,7,8} One animal was placed in each of five air-tight 2-l flow-through chambers mounted in parallel and placed in a versa-range test chamber (Blue-M Engineering Co.). The latter maintained the ambient temperature at 33-35°C, which in this model is known to maintain the temperature of anesthetized mice at 37 ± 0.2°C.⁹

Control animals breathed room air supplied at 4 l/min for 30 min. Test groups were exposed to one of four concentrations of isoflurane (1%, 1.4%, 2%, and 3%) in room air supplied at 4 l/min for the 30-min equilibration period. Inspired isoflurane concentrations were measured by an infrared analyzer (Beckman Medical Gas Analyzer LB-2) and inspired oxygen concentrations by a paramagnetic oxygen analyzer (Beckman Oxygen Analyzer Model E2). At the onset of each test period, the supply of room air was stopped and the chambers were flushed with nitrogen at 8 l/min and the hypoxic gas mixture at 15 l/min. After 1 minute the nitrogen was turned off, and after another minute the flow of the hypoxic gas mixture was reduced to 3 l/min. A previous study had shown that with the above

* Research Fellow in Anesthesiology.

† Professor of Anesthesiology.

Received from the Department of Anesthesiology, the Mayo Clinic, and the Mayo Medical School, Rochester, Minnesota 55905. Accepted for publication January 4, 1983. Supported in part by Research Grant NS-7507 from the National Institutes of Health, Public Health Service, and a grant from Ohio Medical Anesthetics.

Address reprint requests to Dr. Newberg: Department of Anesthesiology, Mayo Clinic, Rochester, Minnesota 55905.

flows, the oxygen concentration in all chambers reached 5% after the first minute and was maintained at 5% throughout the study.²

The hypoxic gas mixture was blended with the concentration of isoflurane used in the 30-min equilibration period in a Tissot 120 liter spirometer, which then supplied gas to the chambers during the test period. Uniformity of gas mixing was assured by continuous analysis of isoflurane and oxygen concentrations in the outflow tubing of the spirometer. The oxygen concentration measured in the individual chambers ranged from 4.85–5.05%. Results from any chamber where the oxygen concentration exceeded this range were discarded.

Survival time, defined as the interval between the initiation of the hypoxic gas flow and the cessation of respiration, was recorded for each animal. Nineteen to 23 animals were studied in each group. The mean survival times of the groups exposed to each isoflurane concentration were compared by analysis of variance. Any differences found were tested by the Bonferroni *t* test for unpaired data.

CANINE ISCHEMIA STUDY

Twelve unmedicated fasting mongrel dogs weighing 10.5–19.0 kg were studied according to a protocol described by Michenfelder and Theye.¹ Anesthesia was induced and maintained with 1.4% end-expired isoflurane in 30% oxygen plus nitrogen for the surgical preparation. Succinylcholine (40 mg) was injected intravenously to facilitate endotracheal intubation and thereafter infused at a rate of 150 ml/h to maintain muscle paralysis. Ventilation was controlled with a Harvard® pump adjusted to maintain normocarbia. A peripheral intravenous catheter was placed for the administration of drugs and maintenance fluid, isotonic saline, infused at a rate of 75 ml/h. A cannula was placed in the left femoral artery for pressure measurements and blood sampling. A large-bore cannula was placed in the right femoral artery. This was then cross-clamped, and connected to the bottom of a liter reservoir containing 5,000 units heparin. The top of the reservoir was attached to an anaeroid manometer, which permitted control of the pressure within the reservoir. Esophageal and parietal epidural thermistor probes were placed to monitor temperature, which was maintained at 37°C with the use of a heating pad and lamps. Biparietal electroencephalogram (EEG) was monitored continuously from electrodes cemented to the skull. Inspired and end-expired isoflurane concentrations were measured with an infrared analyzer (Beckman Medical Gas Analyzer LB-2).

A bilateral frontal-parietal craniectomy was performed, and the dura was excised to expose the dorsal aspect of the cerebral hemispheres in preparation for

taking brain biopsies. During the surgical preparation, serial blood-gas analyses were made and the following conditions established (mean \pm SEM): PaO₂—148 \pm 11 mmHg; PaCO₂—40 \pm 1 mmHg; pH—7.38 \pm 0.01 units; and buffer base (BB+)—45 \pm 1 mEq/l adjusted by the administration of sodium bicarbonate.

In the control group of six dogs, the isoflurane was discontinued, and the animals were maintained on nitrous oxide, 70%, and oxygen. Skin and muscle edges of the craniectomy were infiltrated with 1–2 ml of 1% lidocaine. A period of 30 min was allowed for the animals to eliminate isoflurane before control measurements were obtained. The test group of six dogs was exposed to 3% isoflurane, end-expired, in oxygen and nitrogen for the 30-min before control measurements and throughout the hypotensive period.

During a 20-min control period, blood samples were analyzed for arterial blood gases (IL electrodes), hemoglobin concentration (IL CO-oximeter 282), and arterial lactate and pyruvate concentrations (enzymatic technique).¹⁰

Thereafter, in each dog the femoral artery cannula was opened to the reservoir and the mean arterial pressure (MAP) was decreased within 30 s to 30–32 mmHg. Arterial blood pressure and EEG were recorded continuously throughout the hypotensive period. Thirty seconds after the desired MAP had been achieved, a biopsy was taken from the exposed cerebral cortex according to the method of Kramer *et al.*¹¹ Subsequent biopsies were taken from alternating cerebral hemispheres at 1.5, 3, 5, 7, and 9 min. Each sample was analyzed for adenosine triphosphate (ATP), adenosine diphosphate (ADP), adenosine monophosphate (AMP), phosphocreatine (PCr), glucose, lactate, and pyruvate, as described by Lowry and Passoneau.¹⁰ The energy state of the tissues was expressed as the energy charge (EC) of the adenine nucleotide pool (EC = [ATP] + 0.5 [ADP]/[ATP] + [ADP] + [AMP]).¹²

The cerebral metabolic data obtained at each period after the onset of hypotension were compared between the untreated control group and the group exposed to isoflurane. Significant differences were determined by Student's *t* test for unpaired data.

Results

MOUSE HYPOXEMIA STUDY

The mean survival times with standard error of the mean are presented in figure 1. The mean survival time of the control mice was 4.97 \pm 0.25 min. This did not differ significantly from the mean control value reported in a previous study.² Mean survival times of the groups exposed to 1.0% and 1.4% isoflurane were 9.61 \pm 0.52 and 7.20 \pm 0.33 min, respectively. These were

prolonged significantly over the survival time of the control group. The mean survival time of the group exposed to 2.0% isoflurane did not differ from that of control, while the mean survival time of the group exposed to 3.0% isoflurane was 3.04 ± 0.25 min, significantly less than that of the control group.

All mice in the control group convulsed terminally. None of the mice exposed to the various isoflurane concentrations had seizures.

CANINE ISCHEMIA STUDY

During the 9-min period of hemorrhagic hypotension, the mean arterial pressures (calculated from measurements made at 1-min intervals) in both groups were 31 ± 2 mmHg. Arterial blood-gas values for the two groups before and after 9 minutes of hypotension are presented in table 1. While P_{CO_2} decreased slightly in both groups, a mild metabolic acidosis, exemplified by a significantly decreased pH and buffer base, occurred only in the untreated group after 9 min of hypotension. Arterial lactate concentrations and L/P ratios increased in all dogs, but the increase was significant only in the untreated group. The hemoglobin concentration in the group exposed to isoflurane was significantly less than that of the untreated group during the control period and remained so throughout the period of hypotension.

In the untreated dogs, the EEG remained active throughout the period of hypotension. However, within 30–45 s of the onset of hypotension, the EEG changed from a pattern of low-amplitude high-frequency to a pattern of higher amplitude slow waves with intermittent bursts of low-amplitude waves. Three per cent isoflurane produced an EEG pattern of electrical silence with rare spikes or bursts of high-amplitude slow waves superimposed. Biopsy sampling had no obvious effect on the EEG.

Cerebral energy stores of ATP and phosphocreatine and the calculated energy charge decreased in all dogs during the period of hypotension (fig. 2–4), while the

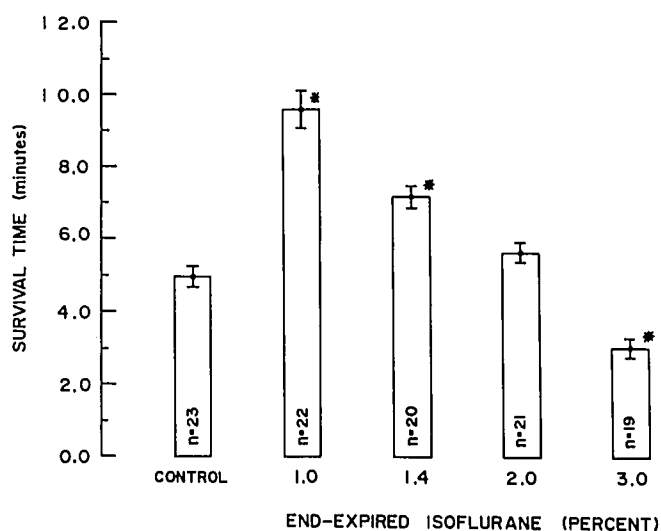


FIG. 1. Mean survival time of control mice exposed to 5% oxygen in nitrogen, compared with test mice exposed to various concentrations of isoflurane, 5% oxygen, in nitrogen. Test mice were exposed to the given concentration of isoflurane in room air for 30 min before the onset of the hypoxic gas mixture. Vertical bars represent \pm SEM. *Difference from control statistically significant $P < 0.05$.

cerebral lactate concentrations increased (fig. 5). In the dogs exposed to isoflurane, the energy stores were maintained at levels greater than those observed in the control group. This difference reached statistical significance at 7 min for the ATP concentration, and at 1.5, 5, and 7 min for the phosphocreatine concentration. The energy charge was significantly greater in the isoflurane group throughout the hypotensive period. Lactate accumulation in the isoflurane-treated group was significantly less during the hypotensive period, starting at the 1.5-min interval.

Discussion

Under normothermic conditions and in the absence of toxicity, maximal CMR_{O_2} suppression is produced by

TABLE 1. Arterial Blood Values in the Untreated and Isoflurane Groups before and after 9 Minutes of Ischemia

	Control		After 9 Minutes of Hypotension	
	Untreated	3% Isoflurane	Untreated	3% Isoflurane
P_{O_2} mmHg	147 ± 7	151 ± 13	132 ± 6	144 ± 9
P_{CO_2} mmHg	39 ± 1.5	41 ± 1	32 ± 4	$35 \pm 0.9^*$
pH	7.37 ± 0.02	7.39 ± 0.01	$7.32 \pm 0.02^*$	7.40 ± 0.01
BB^+ mEq/l	44 ± 1	46 ± 0.4	$37 \pm 1.2^*$	$44 \pm 0.7^{\dagger}$
Hb g/dl	18 ± 0.7	$11 \pm 0.6^{\dagger}$	$16 \pm 0.4^*$	$10 \pm 0.6^{\dagger}$
Lactate μ mol/ml	2.08 ± 0.89	1.63 ± 0.32	$5.00 \pm 0.72^*$	$2.29 \pm 0.48^{\dagger}$
Pyruvate μ mol/ml	0.18 ± 0.03	0.16 ± 0.02	$0.15 \pm 0.01^*$	0.16 ± 0.03
L/P	12 ± 0.3	10 ± 1.0	$36 \pm 5^*$	$14 \pm 2^{\dagger}$

Mean \pm SEM for six dogs.

L/P: ratio of lactate to pyruvate concentration.

* Significantly different ($P < 0.05$) from control value.

\dagger Significantly different ($P < 0.05$) from the value in the untreated group.

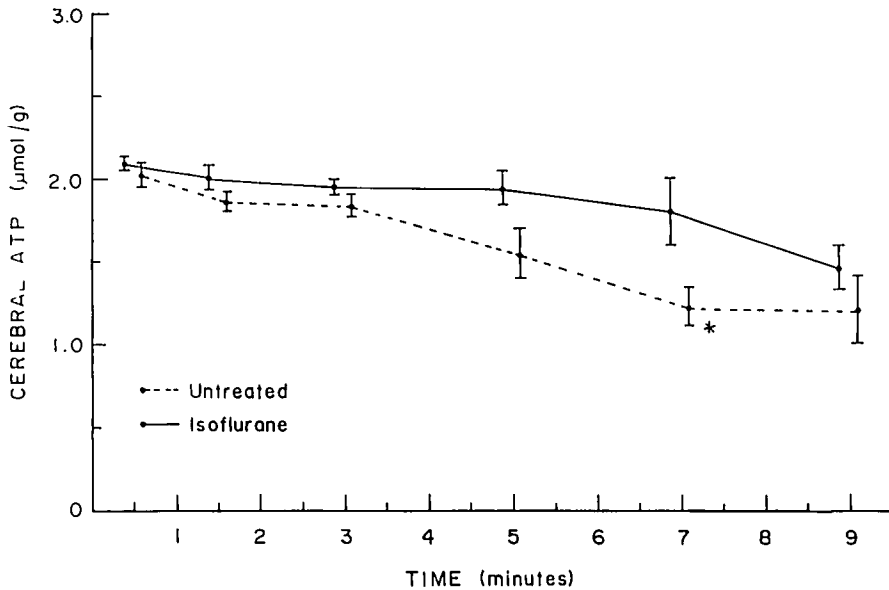


FIG. 2. Effect of 3% isoflurane on cerebral ATP concentrations during cerebral ischemia produced by hemorrhagic hypotension. Mean \pm SEM for six dogs. *Difference statistically significant $P < 0.05$.

barbiturates when given in doses sufficient to suppress cortical electrical activity. Isoflurane is unique among the volatile anesthetics because it can produce an isoelectric EEG in man at a clinically relevant concentration (2.4%). At this concentration it produces moderate peripheral vasodilation with little myocardial depression such that cardiac output is maintained¹³; this differs from the hemodynamic depression produced by barbiturates when given in doses sufficient to abolish cortical electrical activity. In dogs, 3% isoflurane readily and safely suppresses cortical electrical activity and produces apparent maximal, normothermic, nontoxic

suppression of CMR_{O_2} .⁶ If this phenomenon can be extrapolated to humans, maximal suppression of CMR_{O_2} should occur at 2.4% isoflurane. Assuming that metabolic suppression is the primary mechanism responsible for the brain protection reported for the barbiturates and other anesthetics, it follows that isoflurane could provide maximal protection among anesthetics with the least hemodynamic consequences.

The purpose of the present studies was to investigate the possible protective effect of isoflurane using two animal models in which barbiturates were previously shown to provide some protection.

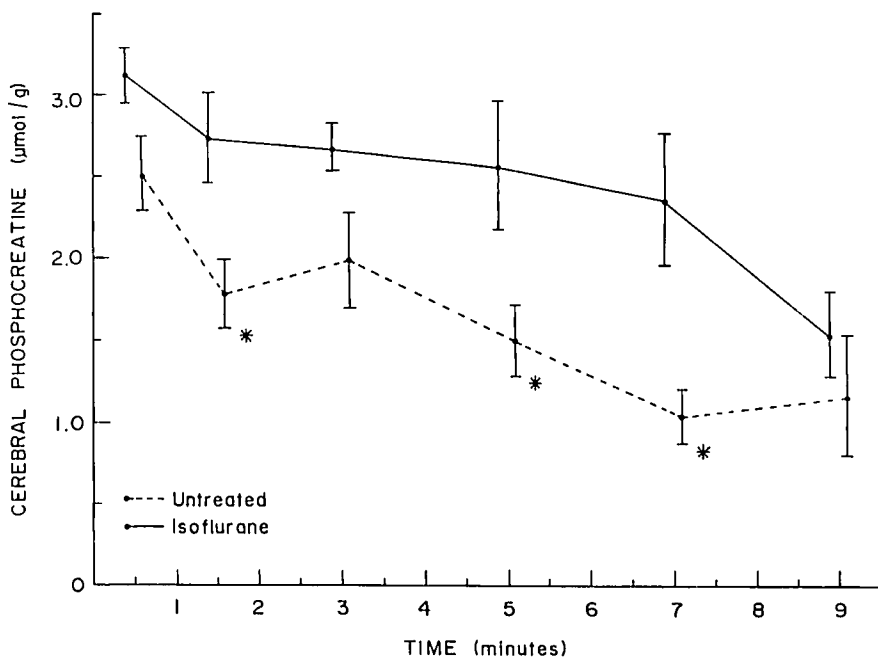
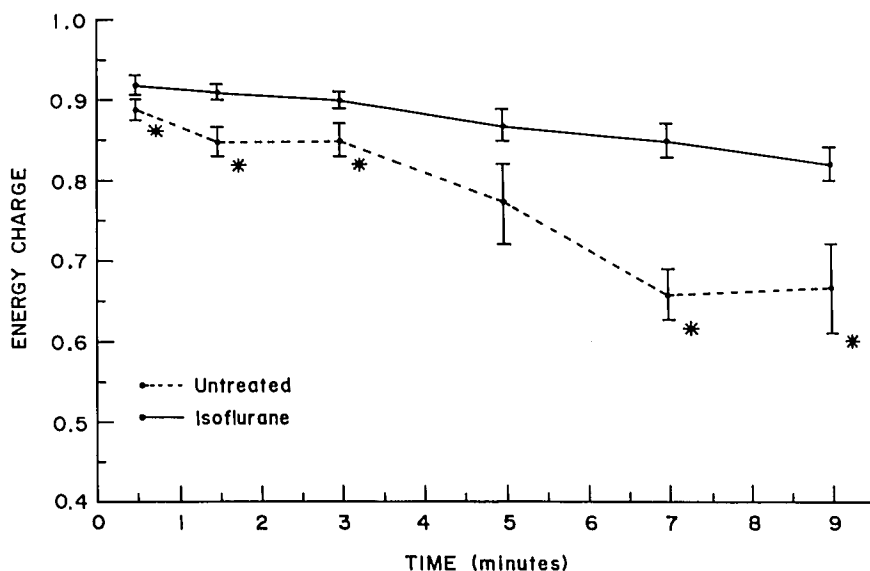


FIG. 3. Effect of 3% isoflurane on cerebral phosphocreatine concentrations during cerebral ischemia produced by hemorrhagic hypotension. Mean \pm SEM for six dogs. *Difference statistically significant $P < 0.05$.

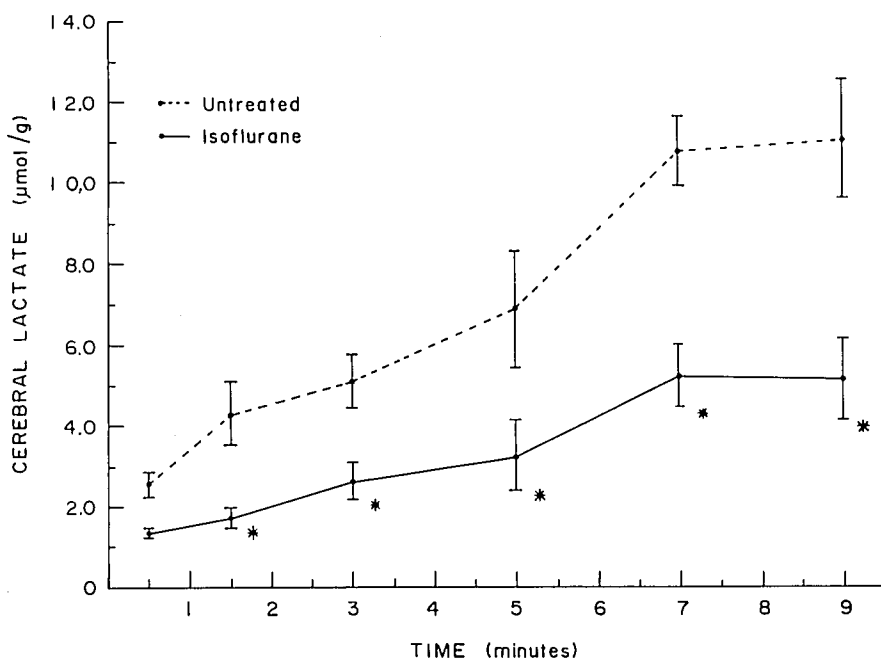
FIG. 4. Effect of 3% isoflurane on the calculated energy charge during cerebral ischemia produced by hemorrhagic hypotension. Mean \pm SEM for six dogs. *Difference statistically significant $P < 0.05$.



Proposed mechanisms for protection in the hypoxic mouse model include, in addition to cerebral metabolic depression produced by the anesthetic,¹ suppression of convulsions,^{2,14} increased cerebral blood flow,⁷ and reduction of potassium flux.¹⁵ Pretreatment with thiopental increased mean survival time by 154%,³ pentobarbital increased it by 303%,¹⁶ and the anesthetically active isomer (-) of mephobarbital increased it by 197%.² Prolonged survival by the barbiturates is believed to result primarily from cerebral metabolic depression,^{1,2,17} because the protective action of the barbiturates cannot be separated from their anesthetic action.² Some protection also is attributed to suppression

of hypoxic convulsions.² Halothane prolongs survival by 84% in this model. This is believed to be secondary to some cerebral metabolic depression and increased cerebral blood flow.⁷ Diazepam prolongs survival by 47–53%, which is believed to result solely from suppression of terminal hypoxic convulsions, because it has little effect on CMR_{O_2} .² However, another benzodiazepine, midazolam maleate, provides greater protection than diazepam by prolonging survival 183%, which is believed to be secondary to its ability to reduce CMR_{O_2} .¹⁸ Phenytoin has no effect on CMR_{O_2} , decreases cerebral blood flow, and is less effective than diazepam in suppressing convulsions in this model. It prolongs

FIG. 5. Effect of 3% isoflurane on cerebral lactate accumulation during cerebral ischemia produced by hemorrhagic hypotension. Mean \pm SEM for six dogs. *Difference statistically significant $P < 0.05$.



survival by 127%, an effect that is believed to be secondary to a reduction in potassium flux.¹⁵ The present study demonstrated a 93% prolongation of the mean survival time by exposure to 1% isoflurane. Possible mechanisms of protection by 1% isoflurane include its ability to decrease CMR_{O_2} ,⁶ increase cerebral blood flow,¹⁹ and suppress hypoxic convulsions.

Although a previous study⁶ demonstrated that 3% isoflurane produced maximal depression of CMR_{O_2} in dogs, exposure of mice to this concentration significantly shortened their survival time. This was probably because of cardiorespiratory depression that was not treated by appropriate support measures.

In the ischemic study, as a means of restricting oxygen delivery to the brain without abolishing cortical electrical activity, dogs were exposed to acute hemorrhagic hypotension. In the isoflurane group, the dogs were treated with 3% end-expired isoflurane before and throughout the hypotensive period. This reduced the cortical electrical activity as reflected by burst suppression or an isoelectric EEG such that CMR_{O_2} was presumed close to being maximally suppressed and at or near that level necessary for the maintenance of cellular integrity only,⁶ both before and during the period of reduced oxygen delivery. During the period of hypotension, isoflurane did provide some cerebral metabolic protection similar in magnitude to that reported for thiopental in this model.¹

In the absence of isoflurane the EEG was altered and the cerebral energy stores of ATP and phosphocreatine decreased progressively to 57% and 24% of normal²⁰ within 7 min. The energy charge decreased to 72% of normal, while the lactate concentration increased 10-fold. This indicated that oxygen and substrate delivery to the brain in the absence of isoflurane was insufficient to maintain either normal cortical electrical activity or cellular integrity. In the presence of isoflurane, the decrease in cerebral oxygen delivery during the period of hypotension was less deleterious, as reflected by better maintenance of cerebral energy stores and less accumulation of lactate. After 7 min of hypotension cerebral concentrations of ATP and phosphocreatine were 84% and 78% of normal, respectively. The energy charge was 94% of normal, while lactate accumulation was half that of the untreated animals. After 9 minutes of hypotension, the energy stores were decreased further to the same levels, as reported previously in the presence of thiopental in this model.¹ This demonstrates the temporal limits to the protection provided during such continued severe oxygen deprivation.

We propose that the primary mechanism of cerebral protection provided by isoflurane in this model of ischemia is that of a reduction in CMR_{O_2} correlated to the observed suppression of cortical electrical activity. This

also is the mechanism usually proposed for barbiturate protection.^{1,21,22} Other proposed mechanisms for barbiturate protection include scavenging of free radicals²³ (a property of some but not all barbiturates), attenuation of free fatty acid liberation,²⁴ decrease in edema formation,²⁵ a reverse steal effect (in focal ischemia),²⁶ and a decrease in intracranial hypertension.²⁷ None of these properties have been shown as yet for isoflurane. In our model of incomplete global ischemia, the ability of 3% isoflurane to maintain or increase cerebral blood flow^{6,19} may have contributed to oxygen delivery and therefore provided some protection. This is in contrast with the decreased cerebral blood flow produced by thiopental,²⁸ which actually may be deleterious in this model.

The isoflurane group differed from the untreated group in two respects other than cerebral protection. Despite random selection of animals, the isoflurane group had a significantly decreased hemoglobin concentration (Hb) throughout the study period (table 1). This decreased Hb provided a reduced oxygen-carrying capacity. Whether this was compensated by an increased cerebral blood flow resulting from the decreased blood viscosity is unknown. Secondly, it is likely that the volume of blood loss required to achieve hypotension in the isoflurane group was less than that in the untreated group because of peripheral vasodilation. There is no evidence, however, that circulating blood volume *per se* has any direct influence on cerebral blood flow. The latter is presumed to be determined by cerebral perfusion pressure alone, at pressure levels that exceed the limits of autoregulation. It seems unlikely that either of these differences impacted meaningfully on the observed cerebral metabolic differences.

We conclude that isoflurane via its anesthetic effect on cortical electrical activity and consequent reduction of CMR_{O_2} can provide some cerebral protection against ischemia in situations of decreased oxygen delivery that are insufficient to abolish electrical activity. Because isoflurane can provide normothermic cerebral protection at concentrations that produce relatively minor cardiorespiratory effects, and because its effect can be rapidly reversed, it may have clinical application for cerebral protection.

References

1. Michenfelder JD, Theye RA: Cerebral protection by thiopental during hypoxia. *ANESTHESIOLOGY* 39:510-517, 1973
2. Steen PA, Michenfelder JD: Cerebral protection with barbiturates: relation to anesthetic effect. *Stroke* 9:140-142, 1978
3. Wilhjelm BJ, Arnfred I: Protective action of some anesthetics against anoxia. *Acta Pharmacol Toxicol* 22:95-98, 1965
4. Keykhah MM, Welsh FA, Harp JA: Cerebral energy levels during trimethaphan induced hypotension in the rat. *ANESTHESIOLOGY* 50:36-39, 1979
5. Nugent M, Artru AA, Michenfelder JD: Cerebral metabolic, vas-

- cular and protective effects of midazolam maleate. *ANESTHESIOLOGY* 56:172-176, 1982
6. Newberg LA, Milde JH, Michenfelder JD: The cerebral metabolic effects of isoflurane at and above concentrations that suppress the electroencephalogram. *ANESTHESIOLOGY* 59:32-37, 1983
 7. Secher O, Wilhelm B: The protective action of anaesthetics against hypoxia. *Can Anaesth Soc J* 15:423-440, 1968
 8. Arnfred I, Secher O: Anoxia and barbiturates. Tolerance to anoxia in mice influenced by barbiturates. *Arch Int Pharmacodyn Ther* 139:67-74, 1962
 9. Artru AA, Michenfelder JD: A reexamination of physostigmine-induced cerebral protection in the hypoxic mouse. A critical assessment of the model. *Stroke* 11:197-199, 1980
 10. Lowry OH, Passoneau JV: *A Flexible System of Enzymatic Analysis*. New York, Academic Press, 1972, pp 144-145
 11. Kramer RS, Sander AP, Lesage AM, Woodhall B, Sealy WC: The effect of profound hypothermia on preservation of cerebral ATP content during circulatory arrest. *J Thoracic Cardiovasc Surg* 56:699-709, 1968
 12. Atkinson DE: The energy charge of the adenylate pool as a regulatory parameter. Interaction with feedback modifiers. *Biochemistry* 7:4030-4034, 1968
 13. Wade JC, Stevens WC: Isoflurane: An anesthetic for the eighties? *Anesth Analg (Cleve)* 60:666-682, 1981
 14. Wilhelm BJ, Jacobsen E: The protective action of different barbituric acid derivatives against anoxia in mice. *Acta Pharmacol Toxicol* 28:203-208, 1970
 15. Artru AA, Michenfelder JD: Cerebral protective, metabolic, and vascular effects of phenytoin. *Stroke* 11:377-382, 1980
 16. Steen PA, Michenfelder JD: Barbiturate protection in tolerant and nontolerant hypoxic mice: comparison with hypothermic protection. *ANESTHESIOLOGY* 50:404-408, 1979
 17. Astrup J, Sorensen PM, Sorensen MR: Oxygen and glucose consumption related to Na^+ - K^+ transport in canine brain. *Stroke* 12:726-731, 1981
 18. Nugent M, Artru AA, Michenfelder JD: Cerebral metabolic, vascular, and protective effects of midazolam maleate. Comparison to diazepam. *ANESTHESIOLOGY* 56:172-176, 1982
 19. Cucchiara RF, Theye RA, Michenfelder JD: The effects of isoflurane on canine cerebral metabolism and blood flow. *ANESTHESIOLOGY* 40:571-574, 1974
 20. Artru AA, Michenfelder JD: Canine cerebral metabolism and blood flow during hypoxemia and normoxic recovery from hypoxemia. *Journal of Cerebral Blood Flow and Metabolism* 1:277-283, 1981
 21. Astrup J, Skovsted P, Gjerris F, Sorensen HR: Increase in extracellular potassium in the brain during circulatory arrest. Effects of hypothermia, lidocaine, and thiopental. *ANESTHESIOLOGY* 55:256-262, 1981
 22. Hossman KA: Treatment of experimental cerebral ischemia. *Journal of Cerebral Blood Flow and Metabolism* 2:275-297, 1982
 23. Flamm ES, Demopoulos HB, Seligman ML, Posner RG, Ransohoff J: Free radicals in cerebral ischemia. *Stroke* 9:445-447, 1978
 24. Shiu GK, Nemoto EM: Barbiturate attenuation of brain free fatty acid liberation during global ischemia. *J Neurochem* 37:1448-1456, 1981
 25. Simeone FA, Frazer G, Lawner P: Ischemia brain edema: Comparative effects of barbiturates and hypothermia. *Stroke* 10:8-12, 1979
 26. Feustel PJ, Ingvar MC, Severinghaus JW: Cerebral oxygen availability and blood flow during middle cerebral artery occlusion: Effects of pentobarbital. *Stroke* 12:858-863, 1981
 27. Shapiro HM, Wyte Sr, R, Loeser J: Barbiturate-augmented hypothermia for reduction of persistent intracranial hypertension. *J Neurosurg* 40:90-100, 1974
 28. Feustel PJ, Ingvar MC, Severinghaus JW: Cerebral oxygen availability and blood flow during middle cerebral artery occlusion: Effects of pentobarbital. *Stroke* 12:858-863, 1981