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# Effect of Isoflurane and Halothane on In Vivo Ischemia-induced Dopamine Release in the Corpus Striatum of the Rat

## A Study Using Cerebral Microdialysis

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**Background:** Dopamine is released in large quantities into the corpus striatum during cerebral ischemia and may exacerbate tissue damage.

**Methods:** Using cerebral microdialysis, the effect of isoflurane on *in vivo* ischemia-induced dopamine release was studied in rat corpus striatum. Reversible cerebral ischemia was induced using carotid ligatures and induced hypovolemia and was monitored with laser-Doppler flowmetry. Following baseline measurements, 28 normothermic, anesthetized rats were subjected to cerebral ischemia followed by reperfusion. The rats were divided into four groups. Group 1 (n = 10) was anesthetized using chloral hydrate. Groups 2 and 3 received 1.5% end-tidal isoflurane. In group 2 (n = 6), hypotension was left untreated during the reperfusion period, and in group 3 (n = 6), mean arterial pressure was maintained using phenylephrine. Group 4 (n = 6) received 1-1.2% end-tidal halothane.

**Results:** Compared with pre-ischemic levels, large quantities of dopamine (350 × baseline levels) were released in group 1 animals during cerebral ischemia. Compared with group 1, ischemia-induced dopamine release was significantly reduced in group 2 (by 58%) and in group 3 (by 56%), but not in group 4. Group 2 animals were uniformly hypotensive during reperfusion and continued to release substantial amounts of do-

pamine (8 × baseline levels). In groups 1, 3, and 4, dopamine release decreased to near baseline levels during reperfusion. In group 3, dopamine metabolite production was significantly increased during ischemia, suggesting that enzymatic function and neuronal reuptake of dopamine was preserved.

**Conclusions:** Isoflurane, compared with chloral hydrate and halothane, inhibits the release of the neurotransmitter dopamine during cerebral ischemia. (Key words: Anesthetics, volatile: halothane; isoflurane. Blood pressure: hypotension. Brain: corpus striatum; metabolism. Cerebral microdialysis. Sympathetic nervous system, catecholamines: dopamine.)

ALTHOUGH cerebral ischemia may have many etiologies, it generally is agreed that the triggering factor that eventually leads to irreversible neuronal damage is a rapid and severe depletion of cerebral energy metabolites. However, when circulation is restored after brief periods of ischemia, energy metabolite levels may recover, but neurologic deficit persists. It has been established that synaptic regions of the neuron are selectively vulnerable to anoxia.<sup>1</sup> A great deal of research has focused on the role played by neurotransmitters in cerebral ischemia. Energy failure causes massive release of catecholamines and excitatory amino acids. These substances have been implicated by a number of studies in the pathogenesis of ischemic brain damage.<sup>2-6</sup>

Inhibition of the release of catecholamines could prevent neuronal injury during global ischemia. The protection of dopamine terminals from ischemia-induced injury by catecholamine depletion supports this concept.<sup>7</sup> When the striatal dopamine content is depleted by unilateral destruction of nigrostriatal pathways, ipsilateral intrinsic striatal neurons are protected from damage during global ischemia in rats.<sup>8</sup> Pharmacologic reduction of dopamine release in ischemic striatum has been achieved using pentobarbital<sup>9</sup> and etomidate.<sup>10</sup>

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Inhibition of ischemia-induced striatal dopamine release may be a property of other anesthetic agents. Isoflurane is a potent volatile anesthetic agent with a depressant effect on cortical metabolism similar to that of barbiturates.<sup>11</sup> There is some evidence that isoflurane provides cerebral protection against global cerebral ischemia<sup>12</sup> and favorably affects the global oxygen supply-demand balance.<sup>13</sup> In humans, electroencephalographic evidence of cerebral ischemia occurs at significantly higher regional cerebral blood flows (CBFs) with halothane than with isoflurane ( $18\text{--}20\text{ ml} \cdot 100\text{ g}^{-1} \cdot \text{min}^{-1}$  vs.  $10\text{ ml} \cdot 100\text{ g}^{-1} \cdot \text{min}^{-1}$ ).<sup>14</sup> Of the potent volatile anesthetic agents, halothane decreases cerebral metabolic rate ( $\text{CMR}_{\text{O}_2}$ ) the least and increases CBF and cerebrospinal fluid pressure the most. The purpose of this study was to compare the effects of isoflurane, halothane, and choral hydrate on the release of dopamine into rat corpus striatum in response to reversible periods of induced cerebral ischemia. Similarly, we studied the effect of these agents on the formation of dopamine metabolites 3,4-dihydroxyphenylacetic acid (DOPAC) and homovanillic acid (HVA). The effect of anesthetic agents on neuronal dopamine release and/or metabolism may be one of the mechanisms by which they modify neuronal injury.

## Methods and Materials

### *Animal Preparation*

The study was approved by the Institutional Animal Care and Use Committee of The Mount Sinai Medical Center. Sprague-Dawley rats (250–300 g body weight) were studied. All animals were anesthetized initially with 400 mg/kg intraperitoneal chloral hydrate.

Using blunt dissection, the left subclavian vein was isolated and cannulated, and both common carotid arteries were exposed and carefully separated from the vagosympathetic trunks. Reversible ligatures consisting of loops of O nylon suture material with silicone elastomer tubing were placed loosely around each of the carotid arteries. The right femoral artery was cannulated with a 24-G Teflon catheter, and the arterial pressure was measured using a Sorenson Transpac transducer (Abbott, North Chicago, IL).

After the trachea was intubated *via* a tracheostomy, each animal was transferred to a stereotaxic frame for microdialysis probe insertion. The skull was exposed, and a hole was drilled according to the coordinates for the right corpus striatum (1 mm anterior to the bregma

and 2.5 mm lateral to the midline suture).<sup>15</sup> A microdialysis probe mounted on a probe clip and carrier then was lowered into the right corpus striatum to a depth of 5.5 mm ventral to the dura (fig. 1). Using analogous coordinates on the left, the area above the left corpus striatum and left middle cerebral artery also was exposed. A P-435 Softip Laser-Doppler probe (TSI, St. Paul, MN) was placed over the left middle cerebral artery, and continuous laser-Doppler flowmetry (LDF) was performed using the Laserflo BPM 403A Monitor (TSI).

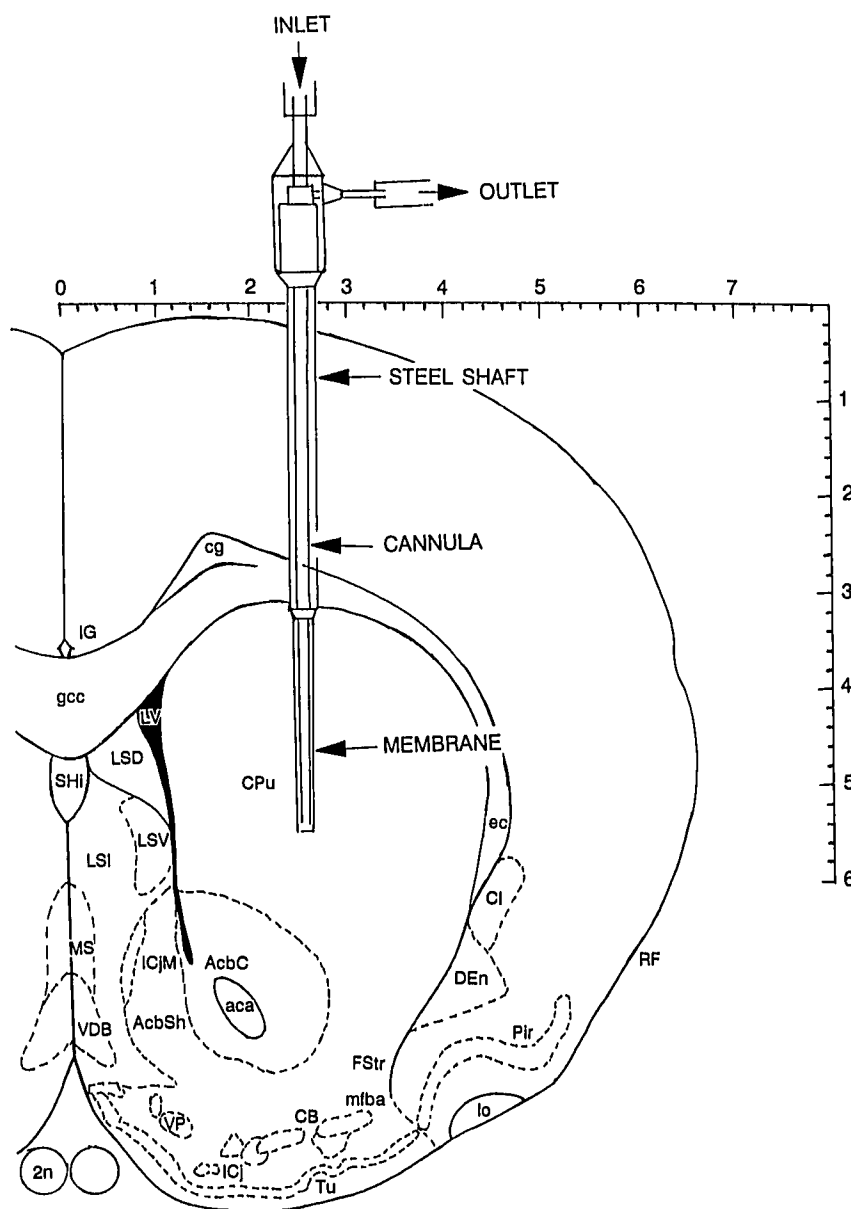
The lungs of each animal were mechanically ventilated (Loosco Amsterdam Infant Ventilator M.K.2, Hoekloos, The Netherlands) using 100%  $\text{O}_2$ , and paralysis was achieved with pancuronium bromide (0.6 mg/kg intravenously). Additional doses of pancuronium bromide (0.3 mg/kg) were administered as necessary to prevent respiratory movements. Arterial  $\text{P}_{\text{O}_2}$  was maintained  $>100$  mmHg,  $\text{Pa}_{\text{CO}_2}$  was maintained between 35 and 40 mmHg throughout the experimental procedure, and ventilatory parameters were adjusted according to the results of intermittent arterial blood gas analyses. Rectal temperature was monitored with a thermocouple probe (Mon-a-Therm, St. Louis, MO) and kept at  $36\text{--}37^\circ\text{C}$  throughout the experiment using an infrared lamp. Brain temperature was determined by insertion of a modified needle thermocouple probe into the left corpus striatum. Intravenous fluid administration consisted of 0.9% NaCl administered at a rate of  $5\text{ ml} \cdot \text{kg}^{-1} \cdot \text{h}^{-1}$ . A three-lead electrocardiograph was recorded using subcutaneous needle electrodes and a Spacelabs 414 Dual Pressure OPT 21 Monitor (Beulah, GA).

### *Experimental Protocol*

The animals were assigned randomly to four study groups. Each of the groups was subjected to an episode of cerebral ischemia, which was followed by a 45–75-min reperfusion period. Group 1 ( $n = 10$ ) received only choral hydrate anesthesia, with additional 100 mg/kg intraperitoneal chloral hydrate every 2 h, or sooner if hypertension or tachycardia was encountered ( $>20\%$  increases from baseline). Group 2 ( $n = 6$ ) animals received isoflurane to an end-tidal concentration of 1.5%, once the preparation had been completed and baseline measurements had been obtained. Isoflurane-induced hypotension (defined as a mean arterial pressure (MAP)  $<60$  mmHg) was not treated during the baseline and reperfusion periods in group 2. Group 3 ( $n = 6$ ) animals were treated identically to group 2, with the exception that phenylephrine was infused intravenously to maintain a MAP of 100–120 mmHg during the base-

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**Fig. 1.** A schematic representation of an adult rat brain with the stereotaxic coordinates of the coronal plane 1 mm anterior to the bregma. The location of the corpus striatum is as indicated. 2n = optic nerve; aca = anterior commissure, anterior; AcbC = accumbens nu, cor; AcbSh = accumbens nu, shell; CB = cell bridges ventral striatum; cg = cingulum; CI = claustrum; CPu = caudate putamen; DEn = dorsal endopiriform nu; ec = external capsule; FStr = fundus striati; gcc = genu corpus callosum; ICj = islands of calleja; ICjM = islands of calleja, major; IG = indusium griseum; lo = lateral olfactory tract; LSD = lateral septal nu, dorsal; LSI = lateral septal nu, intermediate; LSV = lateral septal nu, ventral; LV = lateral ventricle; mfba = medial forebrain bundle; a; MS = medial septal nu; Pir = piriform cortex; RF = rhinal fissure; SHi = septohippocampal nu; Tu = olfactory tubercle; VDB = nu vertical limb diagonal band; VP = ventral pallidum.



line and reperfusion periods. Group 4 ( $n = 6$ ) animals received halothane to an end-tidal concentration of 1.0–1.2%. No additional chloral hydrate was administered to animals receiving volatile anesthetic agents. Both isoflurane and halothane were administered from concentration-calibrated agent-specific vaporizers (Ohmeda, Madison, WI), commencing 30 min before induction of cerebral ischemia and continuing to the end of the experiment. The concentration of volatile

anesthetic agents was monitored using an anesthetic gas analyzer (Puritan-Bennett, Wilmington, MA). As determined from the literature, MAC for isoflurane was taken as 1.4 (vol%), and for halothane, 1.1.<sup>16</sup>

In all animals, a 60-min stabilization period was allowed after insertion of the microdialysis probe before the initiation of the experiment. Samples of the microdialysis perfusate, representing extracellular fluid, were collected automatically at 15-min intervals and ana-

lyzed by high performance liquid chromatography (HPLC) throughout the experiment. A 20-min episode of global cerebral ischemia was induced by tightening the bilateral carotid ligatures and withdrawing 10–20% of the estimated blood volume (6–12 ml/kg) into a heparinized syringe until the MAP was reduced to 40–50 mmHg. The MAP was maintained at this level by gradual withdrawal or reinfusion of blood as needed. This rat model of transient global cerebral ischemia is similar to the technique of Smith *et al.*<sup>17</sup>

LDF was recently validated as a quantitative measure of blood flow in central nervous system tissue.<sup>18,19</sup> Ischemia was confirmed by LDF when the CBF decreased to approximately 20% of the pre-ischemic value. LDF results are presented as arbitrary units. Previous work has determined this to be the ischemic flow threshold for dopamine release.<sup>20,21</sup> The onset of the ischemic interval was timed to precede the beginning of a HPLC cycle by 4 min. Thus, one 15-min HPLC cycle covered minutes 4–19 of cerebral ischemia.

Postischemic reperfusion was accomplished by removing the carotid ligatures and reinfusing the warmed (36° C) autologous blood. The length of the reperfusion period was determined by the interval required for dopamine levels to decrease to baseline or steady-state levels (at least two cycles). The initial cycle included the end of the preceding ischemic period, and the data from this cycle were not included in the analysis. The second (and subsequent cycles, if necessary) confirmed that dopamine release had reached steady-state levels. This was defined as two cycles with dopamine release values with <50% variability.

Data for heart rate, MAP, CBF, rectal temperature, cerebral temperature,  $pH_a$ ,  $PaO_2$ ,  $PaCO_2$ , and blood glucose were collected at the following study points: baseline, cerebral ischemia, and reperfusion period. Values for dopamine and its metabolites DOPAC and HVA were recorded at 15-min intervals and averaged at each study point. Values were determined from the chromatographic tracing as described below and expressed as pg/15 min.

#### Cerebral Dialysis and HPLC Measurements

Extracellular dopamine release into the corpus striatum was analyzed using cerebral microdialysis techniques as previously described.<sup>5,9</sup> The dialysis probes (Bioanalytical Systems CMA/10, West Lafayette, IN) had a tip length of 3 mm. The probes were perfused with artificial cerebrospinal fluid at a rate of 1.4  $\mu$ l/min. The perfusate was fed directly into the injector port of the

HPLC system. Samples were analyzed at 15-min intervals. The HPLC system consisted of a Brownlee C-18 Velosep 3- $\mu$  reverse phase cartridge (3.2 mm ID  $\times$  10 cm; Rainin Instrument, Woburn, MA), a 20- $\mu$ l injection loop, a LC-4B electrochemical detector and cell (Bioanalytical Systems), and a multichannel recorder. The electrochemical detector was set at +0.7 V *versus* an Ag/AgCl electrode.

Before each experiment, the uniformity and efficiency of each microdialysis probe was tested by immersing it in 0.1 N perchloric acid and 40 mg/l diethylethylenetriamine-pentaacetic acid containing 20  $\mu$ g/ml DOPAC, HVA, and dopamine. This procedure resulted in final concentrations that ranged between 20% and 30% of the standard concentrations for each of the compounds under study. Probes falling significantly below 20% were discarded. The mobile phase consisted of 93 parts 150 mM monochloroacetic acid ( $pH$  3.0) with 0.7 mM EDTA and 2.0 mM sodium octyl sulfate (paired ion reagent) and 7 parts acetonitrile. Peak heights and retention times of standard solutions were compared with the reference samples and used to calculate concentrations of constituents.

#### Statistical Analysis

Data were analyzed using repeated measures two-way analysis of variance and Scheffé's multiple contrasts.  $P < 0.05$  was considered statistically significant. All analyses were two-tailed, and data are presented as mean  $\pm$  SD.

#### Results

Group 1 animals remained normotensive during both the baseline and the reperfusion period of the experiment. Group 2 animals were normotensive during the baseline period but remained hypotensive (MAP < 60 mmHg) during the reperfusion period. Group 3 animals received continuous infusions of phenylephrine (1–3  $\mu$ g  $\cdot$  kg<sup>-1</sup>  $\cdot$  min<sup>-1</sup>), titrated to a MAP of 80–100 mmHg during the baseline and reperfusion periods. Group 4 animals were normotensive during the baseline and reperfusion period. The MAP data are summarized in table 1. There were no significant differences between the groups with respect to baseline levels of dopamine, DOPAC, or HVA, or in CBF (figs. 2–4, table 1). Similar degrees of cerebral ischemia, as evidenced by diminished CBF, were obtained in all groups during cerebral ischemia, and all groups had mean CBF values  $\leq$  11 units (table 1). More blood had to be withdrawn from

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Table 1. Mean Arterial Pressure, Cerebral Blood Flow, and Brain Temperature

	Baseline	Ischemia	Reperfusion
<b>Group 1</b>			
Mean arterial pressure (mmHg)	113 ± 18	41 ± 4	98 ± 15
Cerebral blood flow (units)	81 ± 15	11 ± 4	73 ± 12
Brain temperature (° C)	36.6 ± 0.3	33.4 ± 0.5	36.3 ± 0.3
<b>Group 2</b>			
Mean arterial pressure (mmHg)	84 ± 26	37 ± 2	58 ± 3
Cerebral blood flow (units)	73 ± 27	8 ± 4	70 ± 13
Brain temperature (° C)	36.3 ± 0.3	33.7 ± 0.6	36.1 ± 0.1
<b>Group 3</b>			
Mean arterial pressure (mmHg)	108 ± 21	34 ± 17	100 ± 13
Cerebral blood flow (units)	87 ± 35	8 ± 4	87 ± 36
Brain temperature (° C)	36.6 ± 0.3	33.4 ± 0.5	36.3 ± 0.3
<b>Group 4</b>			
Mean arterial pressure (mmHg)	81 ± 11	42 ± 1	98 ± 12
Cerebral blood flow (units)	96 ± 16	3 ± 1	90 ± 15
Brain temperature (° C)	36.3 ± 0.3	33.6 ± 0.6	36.3 ± 0.3

Values are mean ± SD.

group 1 animals to achieve the desired degree of ischemia than from each of groups 2 and 3 ( $7 \pm 2$  ml in group 1,  $2 \pm 1$  ml in each of groups 2 and 3, and  $3 \pm 2$  ml in group 4). Brain temperature decreased by approximately  $4^{\circ}$  C in all four groups during cerebral ischemia (table 1). The brain temperature returned to baseline during reperfusion. Brain temperature did not differ among the groups at any time period.

Large amounts of dopamine were released in group 1 during cerebral ischemia (fig. 2). The dopamine level during cerebral ischemia was approximately 350 times higher than baseline level. Dopamine release was significantly decreased in groups 2 (by 58%) and 3 (by 56%) at cerebral ischemia compared to group 1 ( $P < 0.01$ ; fig. 2). There was no significant difference in do-

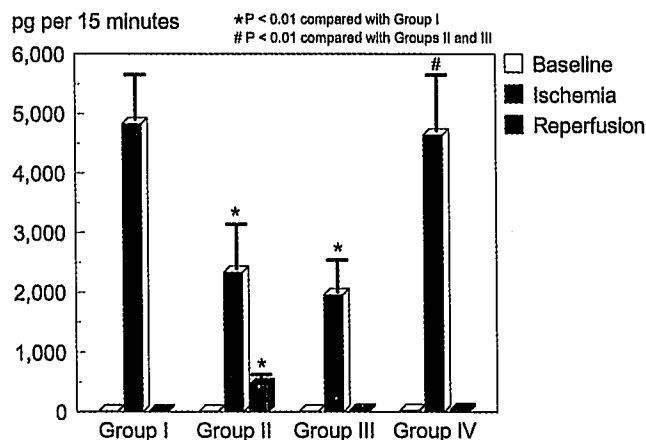


Fig. 2. Dopamine recovery by cerebral microdialysis (pg/15 min, mean ± SD) is presented for baseline (B), cerebral ischemia (IS), and reperfusion (R). Group 1 ( $n = 10$ ) received only chloral hydrate anesthesia. Group 2 ( $n = 6$ ) received 1.5% isoflurane and remained hypotensive during reperfusion. Group 3 ( $n = 6$ ) received 1.5% isoflurane and received phenylephrine to maintain normal blood pressure during baseline and reperfusion. Group 4 ( $n = 6$ ) received 1–1.2% halothane.

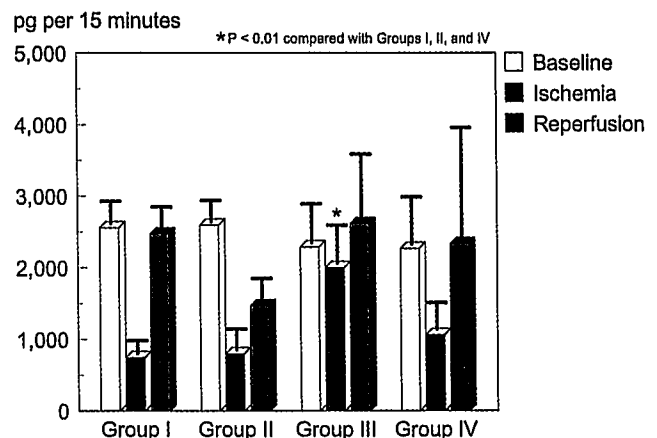


Fig. 3. 3,4-Dihydroxyphenylacetic acid recovery by cerebral microdialysis (pg/15 min, mean ± SD) is presented for baseline (B), cerebral ischemia (IS), and reperfusion (R). Group 1 ( $n = 10$ ) received only chloral hydrate anesthesia. Group 2 ( $n = 6$ ) received 1.5% isoflurane and remained hypotensive during reperfusion. Group 3 ( $n = 6$ ) received 1.5% isoflurane and received phenylephrine to maintain normal blood pressure during baseline and reperfusion. Group 4 ( $n = 6$ ) received 1–1.2% halothane.

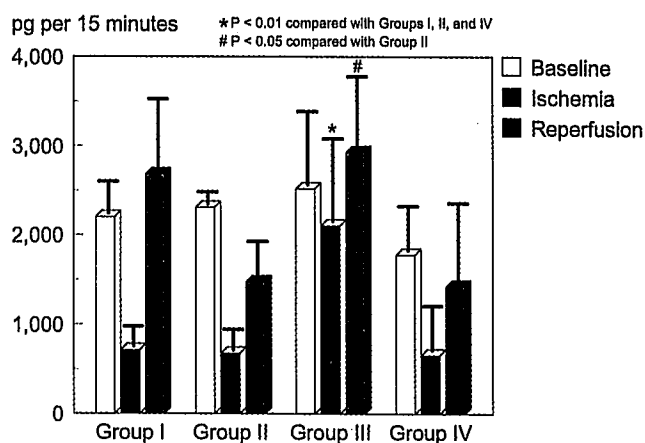


Fig. 4. Homovanillic acid recovery by cerebral microdialysis (pg/15 min, mean  $\pm$  SD) is presented for baseline (B), cerebral ischemia (IS), and reperfusion (R). Group 1 ( $n = 10$ ) received only chloral hydrate anesthesia. Group 2 ( $n = 6$ ) received 1.5% isoflurane and remained hypotensive during reperfusion. Group 3 ( $n = 6$ ) received 1.5% isoflurane and received phenylephrine to maintain normal blood pressure during baseline and reperfusion. Group 4 ( $n = 6$ ) received 1–1.2% halothane.

pamine release during cerebral ischemia between groups 2 and 3. Large amounts of dopamine were released in group 4 during cerebral ischemia (approximately 200 times higher than the baseline level). This did not statistically differ from cerebral ischemia in group 1 but was significantly greater than cerebral ischemia in groups 2 and 3 ( $P < 0.01$ ; fig. 2).

A marked reduction in dopamine release was observed in groups 1, 3, and 4 during reperfusion, to levels that were not distinguishable from baseline. In group 2, however, dopamine release remained approximately eight times greater during reperfusion than baseline levels ( $P < 0.01$ ; fig. 2). The levels of the dopamine metabolites DOPAC and HVA were reduced by 40–70% during cerebral ischemia (compared to baseline) in groups 1, 2, and 4. DOPAC and HVA levels were significantly greater during cerebral ischemia in group 3 compared with groups 1, 2, and 4 ( $P < 0.01$ ; figs. 3 and 4).

## Discussion

The current study describes an animal model of reversible cerebral ischemia as evidenced by normal levels of dopamine release during normotensive reperfusion periods. Isoflurane decreased dopamine release during global cerebral ischemia (groups 2 and 3) com-

pared to control animals (group 1) and halothane animals (group 4). However, isoflurane administration was associated with hypotension that led to continued neuronal dopamine release during the reperfusion period (in group 2). Correction of the hypotension during the reperfusion period (using phenylephrine) in group 3 prevented the continued release of dopamine. Halothane did not decrease ischemia-induced dopamine release (group 4) compared to group 1.

Biotransformation of dopamine proceeds with reuptake into the cytoplasm of the neuron, where monoamine oxidase converts dopamine to DOPAC. DOPAC subsequently is metabolized by membrane-bound catechol-O-methyltransferase to HVA. Dopamine metabolite production was significantly reduced during global cerebral ischemia in group 1, which is consistent with depressed enzymatic function and/or decreased neuronal reuptake. During reperfusion in group 1, metabolite levels were markedly increased, which is consistent with recovery of reuptake and enzymatic function. Group 2 was not distinguishable from group 1 in this respect. In group 3, dopamine metabolite production was significantly increased compared with group 2 during ischemia. This suggests that the combination of isoflurane and an adequate mean arterial pressure during baseline partially preserved enzymatic function and neuronal reuptake of dopamine during ischemia.

When cerebral ischemia occurs, intracellular energy stores are depleted rapidly.<sup>22</sup> This is associated with a rapid increase in intracellular calcium concentration and a marked release of neurotransmitters from nerve terminals into the extracellular fluid of the synaptic cleft.<sup>23,24</sup> Since tyrosine hydroxylase, dopamine- $\beta$ -hydroxylase, and monoamine oxidase require molecular oxygen for their activities, both dopamine synthesis and degradation are believed to be depressed during ischemia.<sup>25–27</sup>

The marked increase in extracellular dopamine during cerebral ischemia in the control animals of our study is consistent with numerous previous reports<sup>5,17,28,29</sup> and suggests that dopamine accumulated in the extracellular spaces of the striatum. Currently, the exact mechanisms responsible for this accumulation are not clear but probably involve a combination of accelerated dopamine release due to active release and/or cell membrane dysfunction, impaired dopamine reuptake, and diminished enzymatic dopamine breakdown. It is possible that the dopamine accumulating in the brain during ischemia may be involved in neuronal injury. Selective vulnerability to

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ischemia has been demonstrated in highly dopamine-dependent areas of the brain such as the striatum and the dorsal hippocampus.<sup>30,31</sup>

The cerebral metabolic and cortical electrical depression caused by isoflurane at clinically useful concentrations suggests that it may offer some degree of cerebral protection. Yet repeated studies examining neurologic and/or histopathologic outcome after global and focal ischemia have failed to demonstrate protection by isoflurane.<sup>32,33</sup> Hence, the importance of  $\text{CMR}_{\text{O}_2}$  reduction as the main mechanism responsible for brain protection has been questioned recently.<sup>34</sup> The protective efficacy of different anesthetics does not parallel their ability to suppress the electroencephalogram or depress  $\text{CMR}_{\text{O}_2}$ . Our data suggest that isoflurane significantly diminishes, but does not abolish, the release of dopamine during ischemia. Furthermore, isoflurane, combined with adequate perfusion pressures during reperfusion, appears to preserve dopamine metabolism. This is an important mechanism whereby the brain protects itself from the toxic effects of excess dopamine. These phenomena are intriguing and warrant further study.

For approximately 10 yr, halothane enjoyed the status of the most commonly used anesthetic agent for neurosurgical procedures. Since 1969, it commonly has been condemned by most for the same procedures.<sup>35</sup> Although halothane is a potent cerebral vasodilator and can cause noticeable increases in intracranial pressure, these effects are obliterated by hyperventilation to a  $\text{PaCO}_2$  of approximately 25 mmHg. Within clinically relevant concentrations, halothane causes a moderate decrease in  $\text{CMR}_{\text{O}_2}$  (20–30%) with no evidence of toxicity as reflected by normal brain metabolites in experimental animals.<sup>36</sup> Our data suggest that halothane is not as effective in attenuating ischemia-induced neuronal dopamine release as is isoflurane.

In studying the effect of ischemia on dopamine release from the corpus striatum, investigators have used many different animal models. The current model incorporated many features of previously described models, while attempting to control most variables potentially influencing dopamine release. These included duration of ischemia, degree of ischemia, and  $\text{PaCO}_2$ . Although CBFs measured by LDF during cerebral ischemia in group 4 were significantly lower compared to the other groups, we believed similar degrees of ischemia nevertheless had been achieved in all groups. The threshold for loss of cellular ion homeostasis requires a reduction of CBF to below  $10\text{--}12 \text{ ml} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1}$ ,

a threshold originally established by workers measuring extracellular  $\text{K}^+$  concentration.<sup>37</sup> At this threshold,  $\text{K}^+$  is released from cells, and  $\text{Ca}^{2+}$ ,<sup>38</sup>  $\text{Na}^+$ , and  $\text{Cl}^-$  with osmotically obligated water enter the cells.<sup>39</sup> Presynaptically, these  $\text{Na}^+$  and  $\text{Ca}^{2+}$  events trigger the release of neurotransmitters.<sup>40</sup> Disruption of cellular energy homeostasis occurs at the same threshold CBF as dissipation of the ion gradients described above. In fact, energy failure and loss of membrane homeostasis are mutually reinforcing events. This threshold flow rate (below 20% of control CBF) is relatively unambiguous and may apply to cats, monkeys, and humans.<sup>41</sup> In rats, the threshold is reportedly higher,<sup>38</sup> probably reflecting the higher neuronal packing density and higher  $\text{CMR}_{\text{O}_2}$ . However, because the control CBF is also higher, the percentage of reduction required to reach a certain threshold is most likely similar. The reductions in CBF attained during cerebral ischemia were well below threshold in all our groups, and it is unlikely that any variability in CBF below this threshold would have influenced dopamine release dramatically.

A recent study has demonstrated that 0.75 MAC isoflurane significantly prolongs the time to terminal depolarization during cardiac arrest in rats ( $77 \pm 7 \text{ s}$  for halothane *vs.*  $102 \pm 19 \text{ s}$  for isoflurane).<sup>38</sup> Over a 15-min ischemic period, the halothane group would have been subjected to a slightly longer period of ischemic depolarization, perhaps partially explaining the larger amounts of dopamine released in this group. The brain temperature decreased equally in all experimental groups. It is therefore unlikely that the results were biased by this factor.

In conclusion, dopamine is released in large quantities into the corpus striatum during cerebral ischemia. Using cerebral microdialysis, the authors studied the effect of isoflurane and halothane on *in vivo* ischemia-induced dopamine release in rat corpus striatum in a controlled fashion. Ischemic dopamine release was significantly reduced by isoflurane but not halothane. During reperfusion, hypotension exacerbated dopamine release in the isoflurane-treated animals, but this was averted when hypotension was prevented using phenylephrine. The data indicate that isoflurane inhibits the release of the neurotransmitter dopamine during cerebral ischemia in a reversible fashion. This phenomenon is consistent with one hypothesis of cerebral protection mediated by anesthetic agents. However, other studies have shown that neuronal death in ischemia may be regulated by the interaction of a variety of neurotransmitters and neuromodulators.<sup>42,43</sup> Further

studies are needed to establish the effect of anesthetic agents on the interrelationship between dopamine and other neurotransmitters and the correlation between these findings and histopathologic outcome.

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