Anesthesiology
80:1320–1327, 1994

Cerebral Blood Flow in Primates Is Increased by Isoflurane over Time and Is Decreased by Nitric Oxide Synthase Inhibition

Robert W. McPherson, M.D., Jeffrey R. Kirsch, M.D.,* Joseph R. Tobin, M.D.,†*
Ramsis F. Ghaly, M.D.,‡ Richard J. Traystman Ph.D.§

Background: **Cerebral blood flow (CBF) decreases over time in dogs and goats during volatile anesthesia. In the current study, we determined CBF during administration of isoflurane for 4 h in cynomolgus monkeys. In addition, we determined if nitric oxide (NO) contributes to cerebrovascular tone during isoflurane anesthesia by determining the CBF (microsphere) response to inhibition of NO synthase with N^{ω}-nitro-L-arginine methyl ester (L-NAME).**

Methods: **CBF was measured in five monkeys anesthetized with isoflurane (1.0% end-tidal). After 4 h of isoflurane (1.0% = 1 MAC), the effects of intravenous L-NAME (60 mg/kg over 10 min) followed by intravenous L-arginine (600 mg/kg over 10 min) on CBF were measured at constant cerebral perfusion pressure and arterial carbon dioxide tension.**

Results: **CBF was unchanged over time (4 h) in cerebellum but increased by 50 ± 18% in both forebrain and hindbrain ($P < 0.05$). CBF decreased by 41–48% ($P < 0.05$) 20 min after L-NAME in forebrain, cerebellum, and hindbrain, at which time brain NO synthase activity was less than 10% of baseline. Twenty minutes after L-arginine, CBF was increased in cerebellum by 32 ± 8% and in forebrain by 41 ± 9% ($P < 0.05$). The cerebral metabolic rate of oxygen consumption was unaffected by time or by L-NAME or L-arginine.**

Conclusions: **These data demonstrate that CBF increases over time during isoflurane anesthesia in primates. Tonic production of NO contributes to control of CBF in primates during isoflurane anesthesia. Increased CBF by L-arginine after L-NAME supports the hypothesis that L-NAME decreases CBF *via* a mechanism requiring NO synthesis. (Key words: Anesthetics, volatile: isoflurane. Blood vessels, vasoconstriction: N^{ω}-Nitro-L-arginine methyl ester. Brain: cerebral blood flow; cerebral metabolic rate of oxygen consumption. L-Arginine. Nitric oxide.)**

ISOFLURANE causes cerebral hyperemia in both pig and dog.[1–4] Inhalational anesthetic–induced cerebral hyperemia resolves spontaneously over time despite continued administration in dogs[5–8] and goats.[9] The mechanism for the decrease in cerebral blood flow (CBF) over time during inhalational anesthesia does not appear to be due to a change in cerebral metabolic rate of oxygen consumption (CMR_{O_2})[5] or alteration in cerebral spinal fluid *p*H.[8] We recently found that inhalational anesthetic–induced cerebral hyperemia is at least partially due to increased production of nitric oxide (NO)[1,2] and vasodilator prostanoids.[1] Therefore, we speculate that a decrease in basal NO-mediated tone during isoflurane anesthesia could account for the decrease in CBF that occurs over time during inhalational anesthesia. A recent study questioned whether inhalational anesthetics cause a transient increase in CBF in humans.[10]

Therefore, the goals of the current study were (1) to determine the effect of time on CBF during isoflurane anesthesia in primates, (2) to determine if NO contributes to basal CBF after 4 h of isoflurane anesthesia, and (3) to assess the efficacy of intravenous L-arginine in reversing any decrease in CBF that may be caused by inhibition of NO synthase with N^{ω}-nitro-L-arginine methyl ester (L-NAME).

Materials and Methods

This study was approved by the Animal Care and Use committee at the Johns Hopkins Medical Institutions. Five adult male cynomolgus monkeys (3.5–5.5 kg) were initially sedated with intraperitoneal thiopental

* Associate Professor, The Johns Hopkins Medical Institutions.

† Assistant Professor, The Johns Hopkins Medical Institutions.

‡ Staff Anesthesiologist, Cook County Hospital.

§ Professor, The Johns Hopkins Medical Institutions.

Received from the Department of Anesthesiology/Critical Care Medicine, The Johns Hopkins Medical Institutions, Baltimore, Maryland, and Cook County Hospital, Chicago, Illinois. Accepted for publication February 15, 1994. Supported in part by grants GM46764 and NS20020 from the National Institutes of Health, United States Public Health Service.

Address reprint requests to Dr. McPherson: Meyer 8-138, The Johns Hopkins Hospital, 600 North Wolfe Street, Baltimore, Maryland 21287.

in increments of 25 mg/kg until the animal was sufficiently sedated to allow induction of anesthesia by insufflation of isoflurane in oxygen. The total dose of thiopental was approximately 100 mg/kg. In these animals, emergence occurred when insufflation of isoflurane was not begun within 3–5 min of loss of consciousness that resulted from thiopental. After induction of anesthesia, a tracheostomy was performed and the lungs mechanically ventilated to normocapnia. Oxygen was administered to maintain arterial oxygen tension greater than 90 mmHg, and isoflurane was administered into the inspiratory limb of the ventilator circuit by an isoflurane vaporizer (TC-3, Ohio, Madison, WI). End-tidal isoflurane was monitored with an analyzer designed for halogenated agents (N-2500, Nellcor, Hayward, CA), and end-tidal isoflurane concentration was maintained at 1.0%. A single dose of pancuronium bromide (0.1 mg/kg, intravenous bolus), which is devoid of cerebrovascular effects,[11] was administered to all animals to minimize muscle contraction from electrocautery.

A balloon-tipped catheter was inserted *via* a femoral artery retrograde into the midthoracic aorta to prevent any decrease in cerebral perfusion pressure (CPP) during the experiment. A femoral vein catheter was placed for rapid removal of blood to prevent increases in CPP after L-NAME administration. Bilateral brachial artery catheters were inserted and advanced into the aortic arch for measurement of mean arterial blood pressure (MABP), withdrawal of microsphere reference blood, and arterial blood sampling. A catheter was inserted *via* left thoracotomy into the left atrial appendage for injection of radiolabeled microspheres.

The animal was turned prone, and a catheter was inserted in the sagittal sinus for withdrawal of cerebral venous blood and measurement of sagittal sinus pressure. Arterial and sagittal sinus pressure transducers were referenced to the right atrium. Pressures were measured with transducers (P23 Db, Statham, Oxnard, CA) and recorded on a polygraph (Gould-Brush, Cleveland, OH). A thermistor (Mon-A-Therm, LaBarge, St. Louis, MO) was placed into the parietal lobe to a depth of 3 mm *via* a burr hole, and temperature was maintained at 37.5 ± 0.2°C with a heating lamp and heating pads.

Arterial blood glucose was measured with a Yellow Springs (Yellow Springs, OH) Glucose Analyzer 2300. Glucose-containing solutions were administered when serum glucose concentration was less than 60 mg/dl. Arterial and sagittal sinus blood oxygen tensions, carbon dioxide tension, and *p*H (ABL3 analyzer, Radiometer, Copenhagen, Denmark) were measured at 37°C immediately after each CBF measurement. Hemoglobin oxygen saturation and hemoglobin concentration were measured spectrophotometrically (Hemoximeter OSM3, Radiometer).

Arterial and cerebral venous oxygen contents were calculated from the measured arterial and venous hemoglobin oxygen saturations and hemoglobin concentration and corrected for dissolved oxygen. CMR_{O_2} was calculated by multiplying the arterial to cerebral venous oxygen content difference by hemispheric CBF. CPP was calculated as MABP minus sagittal sinus pressure. Cerebrovascular resistance (CVR) was calculated by dividing CPP by CBF. CBF was measured with radiolabeled microspheres (16 ± 0.5 μm in diameter; DuPont–New England Nuclear Products, Boston, MA) using the reference withdrawal method.[12] Six radiolabels (^{153}Gd, ^{114m}In, ^{113}Sn, ^{103}Ru, ^{95}Nb, and ^{46}Sc) were used and injected in random sequence. Regional CBF was determined in forebrain (cerebral hemispheres), hindbrain (medulla, pons, midbrain, and diencephalon), and cerebellum.

NO synthase activity was determined by a modification of the techniques described by Dwyer *et al.*[13] and Bredt and Snyder[14] before and after L-NAME administration. In four monkeys, a 20-mm craniectomy was created with a diamond-tipped drill bilaterally over the parietal cortex. The first brain tissue sample (approximately 10 × 5 × 5 mm) was removed immediately after the 4-h measurement of CBF (before L-NAME). The post–L-NAME measurement was made from the contralateral cerebral hemisphere immediately after the post–L-NAME CBF measurement (approximately 25 min after L-NAME). Immediately after harvesting, each brain sample was placed in 20 volumes (weight/volume) ice-cold buffer containing 50 mM Tris and 2 mM ethylenediamine tetraacetate (*p*H 7.4) and ultrasonicated for 10 s.

In this technique, with [^{14}C]arginine as a substrate, NO synthase generates equimolar products of NO and [^{14}C]citrulline. NO synthase activity was confirmed to be dependent on reduced nicotinamide adenine dinucleotide phosphate and calcium ion. The brain homogenate was centrifuged at 10,000*g* × 15 min and the supernatant kept on ice until use. In each assay, 25 μl supernatant (homogenized tissue) was added to 100 μl reaction mixture containing 1 mM reduced nicotinamide adenine dinucleotide phosphate, 1.25 mM calcium chloride, and 1 μM [^{14}C]arginine. Incubation

was carried out at 22°C for 30 min and terminated with 2 ml buffer containing 30 mM hydroxyethylpiperazineethanesulfonic acid and 3 mM ethylenediamine tetraacetate, *p*H 5.5. Each sample was applied to a chromatography column of 0.5 ml Dowex AG50WX-8 (sodium ion form, Sigma, St. Louis, MO), which was then eluted with 2 ml water. [^{14}C]Citrulline was quantified by liquid scintillation spectroscopy, with an efficiency of 90–94%. Protein added to each tube was measured according to the method described by Bradford.[15] Results are expressed as activity per milligram protein (picomoles per milligram protein per minute). [^{14}C]Arginine (339.4 mCi/mmol; 1 Ci = 37 GBq) was obtained from DuPont–New England Nuclear Products. All other chemicals were obtained from Sigma Chemical (St. Louis, MO).

Protocol

Surgical preparation required approximately 45 min and was followed by a 15-min stabilization period during which blood gases were adjusted. Baseline measurements were obtained after 1 h of anesthesia.

Baseline data (hemodynamic and regional CBF) were obtained after 1 h of anesthesia and at hourly intervals for an additional 3 h. L-NAME (60 mg/kg) was then administered intravenously over 10 min, and CBF was determined after 20 min. L-Arginine (600 mg/kg) was then administered intravenously, and CBF was measured after an additional 20 min. Removal of blood was required to prevent increases in CPP after L-NAME administration. Reinfusion of all shed blood and inflation of the intraaortic balloon were required to maintain MABP and CPP after administration of L-arginine.

Data Analysis

The study was designed to determine whether regional CBF changes over 4 h of isoflurane anesthesia and to determine whether NO plays a role in basal cerebral vascular tone during isoflurane anesthesia. To assess the first point, we used analysis of variance for repeated measures to compare regional CBF, blood gases, and hemodynamic variables during the measurement periods in which all systemic variables were maintained constant (hours 1–4). To assess the second point, analysis of variance for repeated measures was used to compare regional CBF, blood gases, and hemodynamic variables during the following measurement periods: pre–L-NAME, post–L-NAME, and post–L-arginine. $P < 0.05$ was considered significant. The Student–Newman–Keul test was used to correct for multiple comparisons. Because standard deviation increased with the mean value, a logarithmic transformation[16] was performed on regional CBF and CVR data before statistical analysis. To evaluate the effect of the cortical biopsy and temperature probe insertion, the difference between left and right hemispheric blood flows in all animals for all CBF determinations was evaluated and the 95% confidence interval for the difference determined. Data in text, tables, and figures are presented as means ± standard error.

Results

Table 1 lists physiologic variables at each time point. MABP, CPP, arterial oxygen tension, arterial carbon dioxide tension, and arterial oxygen content were constant over time (4 h). L-NAME decreased sagittal sinus pressure, *p*H, and arterial oxygen content but did not alter MABP, CPP, arterial oxygen tension, or arterial carbon dioxide tension. Removal of 20 ± 6 ml/kg blood was required to prevent increases in CPP after L-NAME administration. Reinfusion of all shed blood and inflation of the intraaortic balloon were required to maintain MABP and CPP after administration of L-arginine. MABP was slightly greater after L-arginine (5 mmHg; $P < 0.05$), but CPP was unchanged. L-Arginine restored sagittal sinus pressure to the pre–L-NAME level, but *p*H and arterial oxygen content remained lower than before L-NAME ($P < 0.05$).

Figure 1 shows CBF during the first 4 h of the protocol in forebrain, hindbrain, and cerebellum. Flow was unchanged over time in cerebellum but increased at hours 3 and 4 in forebrain and hindbrain ($P < 0.05$).

Figure 2 depicts CMR_{O_2} and CVR during the first 4 h of the protocol. Baseline CMR_{O_2} (hour 1) was 2.3 ± 0.2 ml · min^{-1} · 100 g^{-1} and was unchanged over time. CVR was decreased compared with baseline at hours 3 and 4 ($P < 0.05$).

Twenty-five minutes after administration of L-NAME, cortical NO synthase activity was reduced by more than 90% (4.9 ± 1.1 to 0.5 ± 0.1 pmol · mg protein^{-1} · min^{-1}). Figure 3 shows regional CBF before and after L-NAME and L-arginine. L-NAME decreased flow in forebrain, hindbrain, and cerebellum ($P < 0.05$) by 41 ± 9, 48 ± 8, and 46 ± 6%, respectively. L-Arginine after L-NAME increased flow in forebrain (41 ± 9%) and cerebellum (32 ± 7%) ($P < 0.05$). Although the average increase in flow in hindbrain was similar to that in cerebellum (26 ± 6%), one of the five monkeys

Table 1. Hemodynamic Values and Arterial Blood Gases during 4 h of Isoflurane Administration, After Inhibition of Nitric Oxide Synthase and Administration of L-Arginine

Time (h)	MABP (mmHg)	P_{ss} (mmHg)	CPP (mmHg)	pH	P^{O2} (mmHg)	P^{CO2} (mmHg)	Arterial O_2 Content (ml/dL)
1	75 ± 3	7 ± 1	68 ± 4	7.39 ± 0.01	143 ± 21	43 ± 1	13.7 ± 0.3
2	73 ± 3	6 ± 1	67 ± 3	7.38 ± 0.01	143 ± 12	40 ± 1	13.4 ± 0.3
3	77 ± 3	7 ± 1	71 ± 4	7.35 ± 0.01*	147 ± 12	43 ± 1	13.3 ± 0.3
4	74 ± 5	7 ± 1	67 ± 5	7.35 ± 0.01*	149 ± 6	42 ± 0	13.3 ± 0.3
L-Name	76 ± 4	4 ± 1†	72 ± 5	7.29 ± 0.02	141 ± 13	39 ± 2	11.3 ± 0.1†
L-Arg	81 ± 4†,‡	6 ± 1‡	75 ± 2†	7.29 ± 0.03	151 ± 13	40 ± 1	10.6 ± 0.2†

Values are mean ± SE. Values were recorded at hourly intervals during 4 h of isoflurane administration, after inhibition of nitric oxide synthase with N^{ω}-nitro-L-arginine methyl ester (L-Name, 60 mg/kg, intravenous) and after administration of L-arginine (L-Arg, 600 mg/kg, intravenous).

MABP = mean arterial blood pressure; P_{ss} = sagittal sinus pressure; CPP = cerebral perfusion pressure; P^{O2} = arterial partial pressure of oxygen; P^{CO2} = arterial partial pressure of carbon dioxide.

* $P < 0.05$ *versus* 1 h.

† $P < 0.05$ 4 h *versus* L-Name or L-Arg.

‡ $P < 0.05$ L-Arg *versus* L-Name.

did not demonstrate increased flow in this region in response to L-arginine, and therefore statistical significance was not achieved.

Figure 4 illustrates CMR_{O_2} and CVR in response to L-NAME followed by L-arginine. L-NAME increased CVR ($P < 0.05$) and was restored to baseline level by L-arginine. CMR_{O_2} was not altered by either L-NAME or L-arginine.

The mean difference between left and right hemispheric CBFs for all animals during all treatments was 2 ± 1 ml·min^{-1}·100 g^{-1}, with a 95% confidence interval of -0.7–4.7 ml·min^{-1}·100 g^{-1}.

Discussion

We found that in primates, regional CBF remains constant (in cerebellum) or increases (in forebrain and hindbrain) rather than decreases over time during isoflurane anesthesia. This result conflicts with previous studies in dogs[5–8] and goats.[9] We also found that suppression of NO synthase (to approximately 10% of baseline) with L-NAME decreases regional CBF in forebrain, hindbrain, and cerebellum by approximately 50% even after 4 h of isoflurane administration. L-Arginine increased CBF after L-NAME in forebrain and cerebellum, but despite a percentage increase similar to that of cerebellum, the increase in hindbrain did not reach significance.

Because a large dose of thiopental was required in the current study to produce an adequate level of sedation before inhalational induction, it is possible that baseline CBFs were relatively less than what would be observed in monkeys with the use of only inhalational anesthetics. Likewise, if baseline CBF was reduced by residual thiopental, it is possible that the observed increase in CBF over time during isoflurane anesthesia may have been due simply to resolution of residual thiopental anesthesia. We believe that this is unlikely, however, because any increase in CBF due to a resolution of thiopental anesthesia would be associated with an increase in CMR_{O_2}. In the current study, CMR_{O_2} did not change over time. In addition, baseline CBF was similar between monkeys in the current study and dogs in our previous study[5] in all three brain regions (forebrain, hindbrain, and cerebellum), and CMR_{O_2} also was similar between the two species. Likewise, in both studies rapid administration of isoflurane was required to prevent the animals from awakening immediately, and the time course of CBF measurements in relationship to thiopental was identical. Therefore, we believe that differences between the two studies in regional CBF over time are unlikely to relate to differences either in experimental conditions (*e.g.*, level of baseline anesthesia) or in physiologic status at the beginning of the experiments. An important methodologic difference, however, between this study in primates and our previous studies in dogs[5,17] is the monitoring and careful control of brain temperature in the current primate study. In the dog studies, rectal temperature was closely controlled, but brain temperature was not monitored. However, we believe that in our studies in dogs it is unlikely that there were any significant

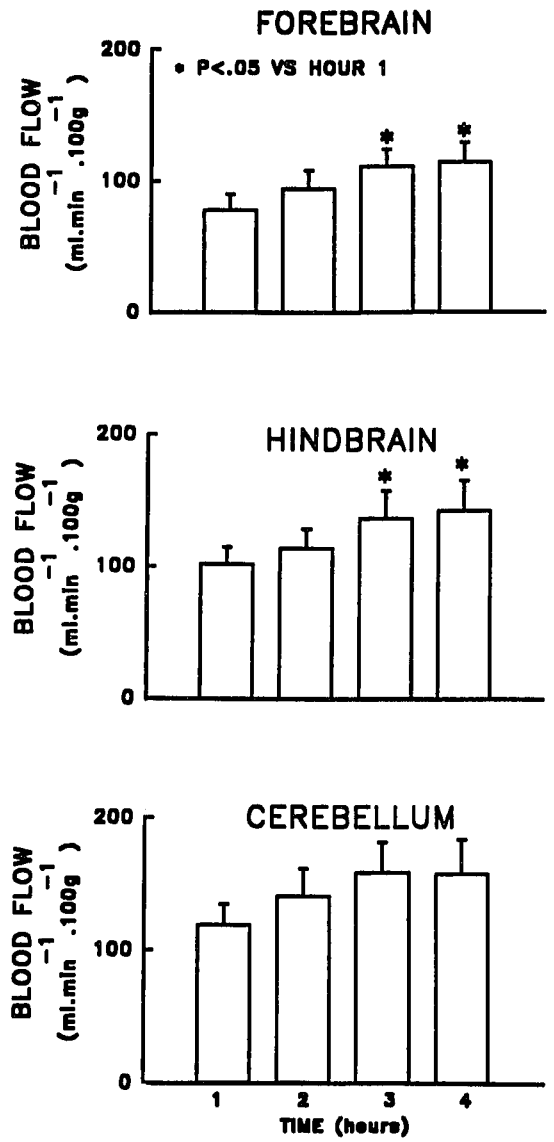

Fig. 1. Flow to forebrain, hindbrain, and cerebellum in five monkeys during 4 h of isoflurane (1.0% end-tidal) administration with constant cerebral perfusion pressure, arterial carbon dioxide tension, and arterial oxygen content. Values are means ± standard error. *$P < 0.05$ *versus* 1 h of isoflurane administration point.

changes in brain temperature, because there were no changes in CMR_{O_2} over time, which would be expected if brain temperature decreased.[18]

The mechanism of isoflurane-induced hyperemia in monkeys was not specifically tested in this study. However, isoflurane-induced hyperemia *in vivo* is mediated, at least in part, by a mechanism involving stimulation of NO synthase to produce NO[1,2] and stimulation of cyclooxygenase to produce vasodilator prostanoids.[1,19] In the current study we did not specifically evaluate whether isoflurane causes cerebral hyperemia in the monkey; rather, we evaluated the effect of time of isoflurane exposure on CBF and determined if a component of cerebral vascular tone during isoflurane anesthesia is mediated by NO. Our data are consistent with

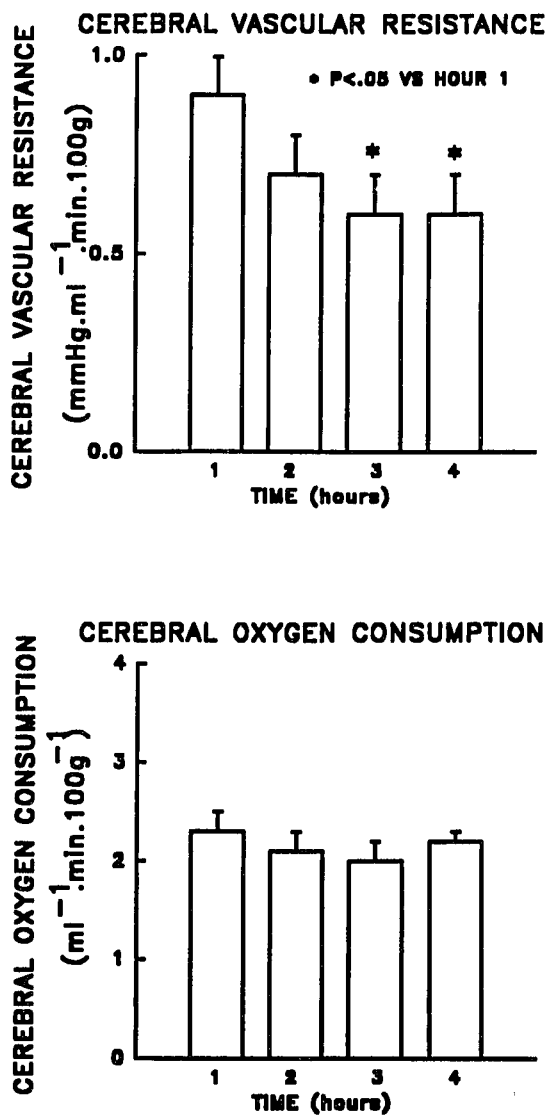

Fig. 2. Cerebrovascular resistance (CVR) and cerebral metabolic rate of oxygen consumption (CMR_{O_2}) in five monkeys during 4 h of isoflurane (1.0% end-tidal) administration with constant cerebral perfusion pressure, arterial carbon dioxide tension, and arterial oxygen content. Values are means ± standard error. *$P < 0.05$ *versus* 1 h of isoflurane administration point.

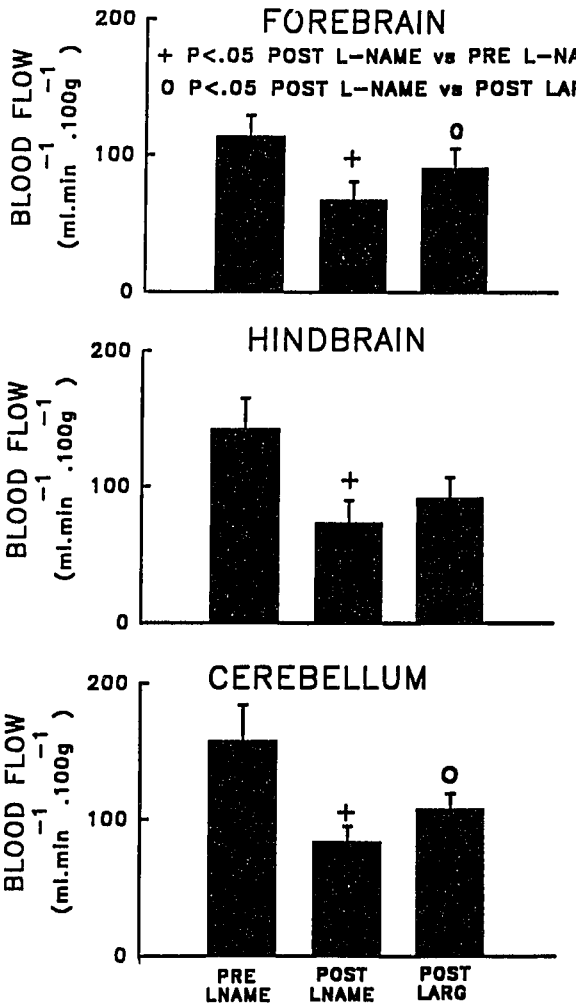

Fig. 3. Flow to forebrain, hindbrain, and cerebellum in five monkeys before and after inhibition of nitric oxide synthase with N$^{\omega}$-nitro-L-arginine methyl ester (L-NAME; 60 mg/kg intravenously) and after L-arginine (600 mg/kg intravenously). Values are means ± standard error.

the hypothesis that NO (or an NO-containing compound) is an important mediator of cerebral vascular tone during isoflurane anesthesia. Because CBF increased rather than decreased over time during isoflurane anesthesia in the monkey, we speculate that in the monkey, unlike the dog and the goat, there is sustained release or production of the vasodilating mediator for isoflurane-induced alteration in cerebral vascular tone.

The mechanism of inhalational anesthetic–induced vasodilation *in vitro* does not appear to be mediated by NO.[20] However, studies *in vitro* may not predict the effect of isoflurane on CBF *in vivo*. For example, NO is an important mediator of cerebral vasodilation produced by perivascular nerves in dogs, primates, and human cerebral arteries,[21–23] and it may be produced in large quantities by NO synthase, which is present in astrocytes[24] and parenchyma.[25] Any contribution of these neural elements to cerebrovascular tone is excluded from a vessel preparation *in vitro*.

The CBF response to NO synthase inhibition is different from that observed in chloralose- and urethane-anesthetized cats,[26] in which there is little or no re-

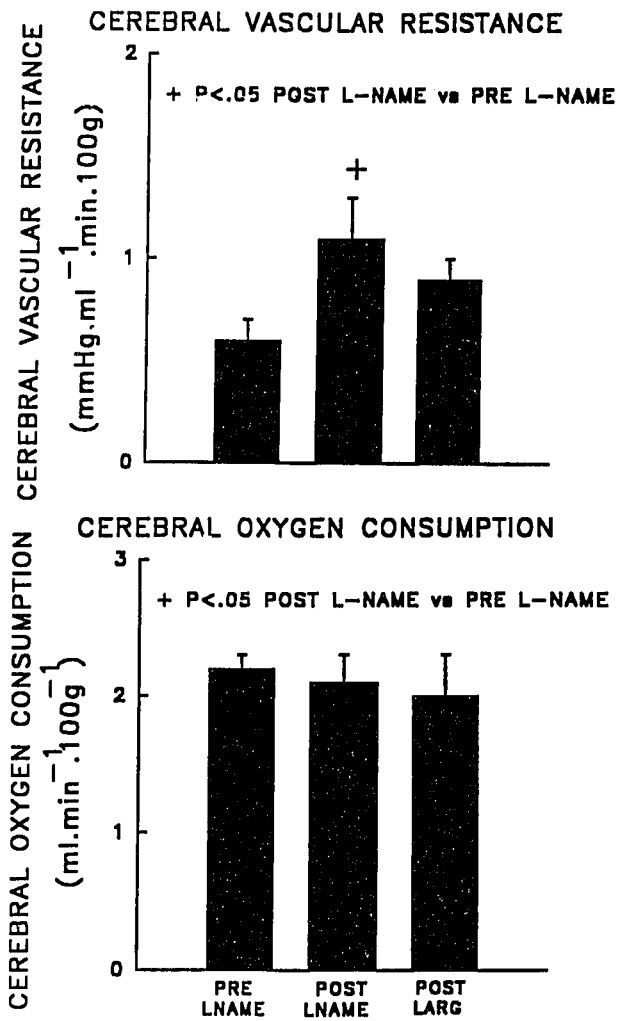

Fig. 4. Cerebrovascular resistance (CVR) and cerebral metabolic rate of oxygen consumption (CMR_{O_2}) in five monkeys before and after inhibition of nitric oxide synthase with N$^{\omega}$-nitro-L-arginine methyl ester (L-NAME; 60 mg/kg intravenously) and after L-arginine (600 mg/kg intravenously). Values are means ± standard error.

duction in CBF to cerebral cortex but significant reduction in flow to most subcortical regions. NO synthase inhibition resulted in no reduction in CBF in nonanesthetized sheep[27] and in little[28] or no[29] reduction in CBF in pentobarbital-anesthetized dogs. In contrast, NO synthase inhibition resulted in diffuse reduction in regional CBF in rats anesthetized with oxygen, nitrous oxide, and fentanyl[30] and in rats studied without anesthesia.[31] Differences in the CBF response to NO synthase inhibition may be a species effect. For example, it is not known whether regional distribution of NO synthase is similar among species or whether sensitivity to NO synthase inhibition differs among species. Alternatively, there may be differences among studies, based on anesthetic conditions. For example, use of anesthetics that decrease CBF, such as pentobarbital or chloralose–urethane, either prevents change[29] or limits change in CBF.[26,28]

Another possible reason for the differences from other studies may relate to the degree of NO synthase inhibition produced with the inhibitor we used. In our study, we measured NO synthase activity and found that a very large dose of L-NAME produced greater than 90% inhibition of NO synthase. This is in contrast with the much smaller dose (10 mg/kg) that was needed to produce a similar degree (greater than 90%) of NO synthase inhibition for 4 h in the cat.[32] Thus, it is difficult to compare studies regarding the effect of NO synthase inhibition because the efficacy of NO synthase–inhibiting agents is not known.

In addition to its effect on NO synthase, L-NAME has been shown to have actions as a muscarinic antagonist.[33] In our study, L-NAME reduces CBF primarily by a specific effect of NO synthase rather than an effect on the muscarinic receptor, because the reduction in CBF can be significantly increased by subsequent treatment with L-arginine. Although blood flow to forebrain returned to pre–L-NAME values after administration of L-arginine, blood flow in hindbrain and cerebellum was restored only partially. We believe that complete reversal was not obtained in these regions because L-arginine could not effectively compete with the strong interaction that L-NAME has with NO synthase.[13] In addition, if an antimuscarinic effect was a major contributor to the decreased CBF produced by L-NAME, other antimuscarinic agents also would decrease CBF under baseline conditions. Consistent with our hypothesis that the antimuscarinic effect of L-NAME is not responsible for alteration in cerebral vascular tone is the observation that atropine does not alter cerebral vascular tone during baseline conditions or during induced dilation.[34]

Our findings agree with those of a previous study[35] that showed that L-arginine *in vivo* effectively antagonizes vasoconstriction produced by L-NAME. Our results concerning L-arginine are somewhat inconsistent: we found that L-arginine increased blood flow in cerebellum and forebrain, but the increase in flow was not statistically significant in hindbrain according to analysis of variance for repeated measures in the comparison of pre–L-NAME, post–L-NAME, and post–L-arginine periods. We believe that this is an effect of sample size, because hindbrain flow increased substantially in response to L-arginine in four of the five animals, and increased only slightly in the fifth.

L-NAME caused systemic vasoconstriction in isoflurane-anesthetized monkeys. Peripheral vasoconstriction by NO synthase blockade appears to be both anesthetic- and species-dependent. In conscious and pentobarbital-anesthetized rats, NO synthase inhibition increases MABP by 50 mmHg, whereas with halothane anesthesia MABP does not change with N^G-monomethyl-L-arginine.[36] In the current study, we prevented increases in MABP by removal of 20 ml/kg blood. Had we not maintained MABP and CPP constant, changes in regional CBF may have been incorrectly attributed to altered CPP and failed autoregulation.

Although it is possible that the decrease in CBF with L-NAME administration was due to intense sympathetic stimulation during hemorrhage, we believe that this is unlikely because even intense hemorrhage, during normoxia, does not decrease CBF.[37] Likewise, we have previously demonstrated that electrical stimulation of the sympathetic nervous system does not cause a reduction in CBF.[38] In addition, if the decrease in CBF with L-NAME were due to a sympathetically mediated decrease in CBF, we would not have anticipated a return in CBF toward baseline values after administration of L-arginine.

We conclude that in primates, CBF increases over time during isoflurane anesthesia. Therefore, in primates, unlike in other species, production or release of the mediator for isoflurane-induced vasodilation is sustained for at least 4 h. Furthermore, even after 4 h of isoflurane anesthesia, there is significant basal NO tone throughout the brain. These findings are consistent with the hypothesis that sustained production of NO during isoflurane anesthesia in monkeys prevents the gradual decrease in CBF that has been observed during isoflurane anesthesia in dogs[5–8] and goats.[9]

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