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Intracoronary Isoflurane Causes Marked Vasodilation in Canine Hearts

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Previous studies of coronary vasomotor effects of isoflurane were complicated by changes in systemic hemodynamic conditions and in global cardiac work demands. Accordingly, in the current study, the left anterior descending coronary artery (LAD) of 11 open-chest dogs anesthetized with fentanyl and pentobarbital was cannulated and perfused with isoflurane-free arterial blood or with arterial blood equilibrated in an extracorporeal oxygenator with isoflurane (0.5, 1.0, 2.0% in 95.5% oxygen–4.5% carbon dioxide). Steady-state changes in coronary blood flow (CBF) in LAD were measured electromagnetically, and their transmural distribution (endocardium: epicardium ratio) was evaluated with 15-μm radioactive microspheres. Venous blood was obtained from the anterior interventricular vein and analyzed for oxygen tension (P_{O_2}) and oxygen content. Myocardial oxygen consumption ($M\dot{V}_{O_2}$) was calculated using the Fick equation. Cardiac responses during isoflurane were compared to those during maximal vasodilation with intracoronary adenosine. Perfusion pressure was maintained at 100 mmHg. CBF increased 271, 279, and 503% with 0.5, 1.0, and 2.0% isoflurane, respectively, with no change in the endocardium:epicardium ratio. With 2.0% isoflurane, the increase in CBF was 80% of the maximal, adenosine-induced response. The increases in CBF caused by isoflurane were accompanied by greater than proportional increases in venous P_{O_2} and decreases in the arteriovenous oxygen difference, reflecting the reduction (approximately 40%) in $M\dot{V}_{O_2}$. In conclusion, isoflurane has a direct, concentration-dependent relaxing effect on coronary vascular smooth muscle in the canine heart *in situ*. The ability of isoflurane to increase CBF nearly maximally while also significantly reducing local myocardial oxygen requirements attests to the potency of isoflurane's direct vasodilator action. (Key words: Anesthetics, volatile: isoflurane. Heart: coronary blood flow; myocardial oxygen consumption.)

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IN 1983, Reiz *et al.* reported that isoflurane anesthesia caused myocardial ischemia in patients with a history of coronary artery disease, as evidenced by ST–T-segment depressions or T-wave inversions in combination with markedly decreased myocardial lactate extraction.[1] Since this myocardial ischemia occurred in the presence of a decrease in calculated left coronary vascular resistance, the investigators suggested that a direct coronary vasodilator action of isoflurane was responsible for diverting flow away from myocardium receiving only marginal perfusion and limited vasodilator reserve, toward myocardium already receiving adequate flow—the "coronary steal" phenomenon.[2]

Studies performed in dogs have yielded inconsistent findings concerning the coronary dilator action of isoflurane suggested by Reiz *et al.*[1] Sill *et al.*[3] reported that isoflurane administration caused uncoupling of coronary blood flow (CBF) from the prevailing myocardial oxygen demands, which is the hallmark of a direct coronary vasodilator.[4] On the other hand, Merin[5] reported that CBF remained proportional to the myocardial oxygen demands during progressive isoflurane administration, indicating intact metabolic control of myocardial perfusion and lack of a direct vasodilator effect. A definitive explanation for these inconsistent findings is uncertain, but a major factor may have been the significant systemic hemodynamic changes accompanying inhalation of isoflurane (*e.g.*, aortic hypotension), which themselves may have profound influence on CBF and its transmural distribution.[4]

The current study was designed to evaluate direct coronary vasomotor effects of isoflurane systematically, under controlled hemodynamic conditions, by perfusing selectively a portion of the left anterior descending coronary artery (LAD) in the *in situ*, working canine heart with arterial blood equilibrated extracorporeally with clinically relevant concentrations of isoflurane. Since the remainder of the dog, including most of the heart, was naturally supplied *via* the aorta with arterial blood free of isoflurane, this approach avoided the systemic hemodynamic instabilities that complicated interpretation of previous studies.

Materials and Methods

Canine Preparation

The study was conducted in compliance with the Institutional Animal Research Committee. Experiments were performed on 11 conditioned, heartworm-free mongrel dogs of either sex (weight range 20–29.5 kg). Anesthesia was induced with intravenous bolus injection of fentanyl (40 $\mu g \cdot kg^{-1}$) and sodium pentobarbital (10 $mg \cdot kg^{-1}$). Anesthesia was maintained by continuous intravenous infusion of fentanyl and sodium pentobarbital at rates of 20 $\mu g \cdot kg^{-1} \cdot h^{-1}$ and 1 $mg \cdot kg^{-1} \cdot h^{-1}$, respectively. In four dogs, supplementary bolus injections of fentanyl were given as needed to maintain heart rate near 100 beats per min. After tracheal intubation and left thoracotomy in the fourth intercostal space, the lungs were mechanically ventilated (Air Shields) with fractional inspired oxygen concentration (FIO_2) equal to 1.0. The volume and rate of the ventilator were established to maintain arterial carbon dioxide tension (P_{CO_2}) at physiologic levels. Oxygen tension (P_{O_2}), P_{CO_2}, and *p*H of arterial blood samples and of coronary perfusate and venous samples (see below) were measured electrometrically (model 413, Instrumentation Laboratories, Lexington, MA). Muscle paralysis was obtained with an intravenous injection of vecuronium bromide 0.1 $mg \cdot kg^{-1}$ with supplements of 0.05 $mg \cdot kg^{-1} \cdot h^{-1}$.

Polyethylene cannulas were inserted into 1) the thoracic aorta *via* the right carotid artery for measuring arterial blood pressure, 2) the left atrium for measuring left atrial pressure, 3) the right brachial artery for collecting samples of arterial blood for gas analysis, and 4) the right femoral vein for administration of heparin (400 $U \cdot kg^{-1}$ with supplementation) for anticoagulation and administration of supplementary anesthetic. A micromanometer-tip pressure transducer (Millar Instruments, Houston, TX) was inserted into the left ventricle *via* the left atrium and mitral valve to measure left ventricular pressure. The maximum rate of increase of left ventricular systolic pressure (dP/dt_{max}) was obtained from the left ventricular pressure pulse with an electronic differentiator. The left ventricular pressure signal was used to drive a cardiotachometer. Arterial, left atrial, and coronary (see below) blood pressures were measured with Statham pressures transducers (model P23ID, Gould, Cleveland, OH). A continuous record of blood pressures, left ventricular dP/dt_{max}, heart rate, and CBF (see below) was obtained on an eight-channel physiologic recorder (model 2800S, Gould).

The LAD was isolated approximately 2 cm from its origin for cannulation. A thin-wall stainless-steel cannula (2.5 mm inside diameter) was introduced into the isolated segment of the LAD, so that the artery could be perfused selectively by an extracorporeal perfusion system (fig. 1). LAD cannulation required that blood flow was interrupted for less than 60 s. The perfusion system used two reservoirs (500-ml aspirator bottles); one contained normal, well-oxygenated blood equilibrated with no isoflurane, while the other contained similar blood equilibrated with isoflurane. The coronary reservoirs were connected to a large (20-l) air chamber that was pressurized with compressed air. Because of the large volume of the air chamber, small changes in the blood volume of the reservoirs had only negligible effect on coronary perfusion pressure. The normal blood reservoir was supplied by a peristaltic pump with blood from the left femoral artery. The isoflurane-equilibrated blood reservoir was supplied with blood from the right femoral artery that was passed through a bubble oxygenator (Bentley-5 pediatric blood oxygenator) supplied with a 95.5% oxygen–4.5% carbon dioxide gas mixture. Isoflurane, provided by a calibrated Fortec vaporizer, was added to the gas mixture supplying the extracorporeal oxygenator, so that direct coronary and myocardial effects of graded concentrations of isoflurane could be studied. Blood supplied to the isoflurane

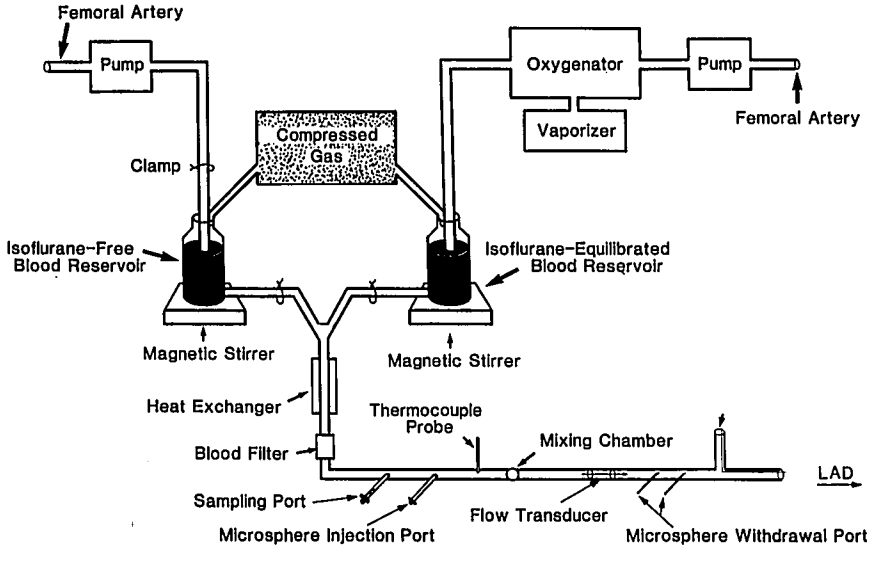

FIG. 1. Extracorporeal perfusion system permitting selective exposure of left anterior descending coronary artery (LAD) to arterial blood equilibrated with isoflurane.

reservoir was recirculated at least three times through the extracorporeal oxygenator to ensure complete equilibration at the desired isoflurane concentration.

A common perfusion tubing connected the perfusion reservoirs with the LAD cannula. This perfusion tubing was equipped with 1) a heat exchanger to maintain coronary perfusate temperature at 38° C, 2) an electromagnetic flow transducer to measure CBF, 3) ports for collecting samples of coronary perfusate, for injecting radioactive microspheres, and for withdrawing reference samples for the microsphere technique, and 4) a mixing chamber for microspheres and drugs administered into the coronary perfusion tubing. LAD perfusion pressure was sensed through a small-diameter tube positioned at the orifice of the perfusion cannula. To minimize blood depletion in experimental animals, blood from donor dogs was used to prime the perfusion system.

Experimental Measurements

Myocardial Oxygen Consumption

In seven dogs, measurements of oxygen consumption in the LAD-perfused myocardium were made in order to distinguish changes in CBF due to direct effects of isoflurane on coronary vascular smooth muscle from those secondary to variations in myocardial metabolic demand. The anterior interventricular vein was cannulated at the same level as the LAD. The venous cannula was allowed to drain freely into a beaker to prevent venous stagnation and interstitial edema. This venous blood was returned intermittently to the dog to maintain isovolemic conditions. At specified times in the study, 1-ml blood samples were collected from the coronary venous cannula under mineral oil to maintain anaerobic conditions. These venous blood samples were paired with 1-ml arterial blood samples obtained from the LAD perfusion tubing, so that the arteriovenous oxygen difference for oxygen could be determined. Hemoglobin concentration and percent hemoglobin oxygen saturation of the coronary blood samples was measured with a CO-Oximeter (model 482, Instrumentation Laboratories), and used to calculate oxygen bound to hemoglobin assuming an oxygen carrying capacity for hemoglobin of 1.39 ml oxygen per g.[6] The oxygen dissolved in the blood was computed (oxygen dissolved = 0.003 ml oxygen · 100 ml blood^{-1} · mmHg^{-1}) and added to the bound component to compute total oxygen content. Myocardial oxygen consumption ($M\dot{V}_{O_2}$) was computed from the product of the coronary arteriovenous oxygen difference and the LAD blood flow (measured electromagnetically) at the time that blood samples were obtained.

Regional Myocardial Blood Flow

In seven dogs, the transmural distribution of blood flow in the LAD-perfused myocardium was determined from tissue uptake of 15 μm microspheres injected into the perfusion tubing proximal to the mixing chamber. In each heart, microsphere injections were made under four conditions—control condition, 1.0% isoflurane, 2.0% isoflurane, and adenosine. These conditions corresponded to the four differently labeled microspheres that were available—^{46}Sc, ^{85}Sr, ^{113}Sn, and ^{141}Ce (New England Nuclear, Boston, MA; 3M, St. Paul, MN). Prior to injection, 50,000 microspheres labeled with a particular radionuclide were dispersed in a solution of 10% dextran and agitated in a vortex mixer and in an ultrasonic bath. Beginning simultaneously with each microsphere administration, duplicate reference samples of labeled blood were withdrawn at a constant rate (6 ml · min^{-1}) for 3 min from the perfusion tubing at different points distal to the mixing chamber. Comparison of radioactivities of the duplicate reference samples was used to assess adequacy of mixing of microspheres in the perfusion tubing. At the completion of the each experiment, Evans blue dye was injected into the LAD, with perfusion pressure maintained at the normal level, to identify the LAD perfusion field. After the heart was stopped with KCl, it was removed, trimmed, and frozen to facilitate sampling. The dyed myocardium was excised and weighed so that electromagnetic blood flow could be expressed on a per-100-g basis. A 3-g transmural sample of myocardium was obtained from the center of the LAD perfusion field. This sample was divided into thirds to yield subepicardial, midmural, and subendocardial samples, which were weighed. Radioactivity of myocardial samples and reference blood samples obtained from the perfusion tubing were measured with a gamma spectrometer equipped with a multichannel analyzer (model 1282-002, LKB, Turku, Finland). Isotope separation was accomplished by standard techniques of gamma spectroscopy with the aid of a minicomputer (model 91499-70, IBM, Boca Raton, FL). Myocardial blood flow (MBF, in milliliters per minute per 100 grams) was calculated from the equation

$$MBF = AF \times (MC/AC) \times 100$$

where AF is the coronary arterial sampling rate (milliliters per minute), MC is microsphere radioactivity (counts per minute per gram) in tissue samples, and AC is coronary arterial blood sample radioactivity (counts per minute). The endocardium:epicardium flow ratio was calculated by dividing blood flow values for the subendocardial and subepicardial samples.

During each microsphere injection samples of blood were collected continuously from the anterior interventricular vein so that arteriovenous shunting of the microspheres could be assessed from the equation[7]

$$\text{percent shunt flow} = (VC/VF)/(AC/AF) \times 100$$

where VC is coronary venous blood sample radioactivity (counts per minute), VF is coronary venous sample rate (milliliters per minute), AC is coronary arterial blood sample radioactivity (counts per minute), and AF is coronary arterial sampling rate (milliliters per minute).

Blood Isoflurane Concentration

In four dogs, isoflurane concentration in LAD arterial perfusate was determined using a modification of the equilibration method described in detail by Yamamura *et al.*[8] Briefly, a 2-ml sample of arterial blood was obtained from the LAD perfusion tubing using an air-tight glass syringe and was introduced into a 5-ml glass vial. The vial was placed in a constant-temperature chamber at 38° C for 30 min. After equilibration, 100 μl of the gas in the vial was introduced into a gas chromatograph (model 5890, Hewlett Packard) equipped with a flame ionization detector, and the area under the curve was measured. Anesthetic concentration in blood was determined by means of a calibration curve derived from appropriate standards. All analyses were performed in triplicate and the mean values calculated.

Experimental Protocols

To rule out time-dependent factors, each exposure of the LAD to isoflurane or adenosine was immediately preceded by control period of normal blood perfusion. Initial control measurements were not obtained until at least 45 min was permitted for recovery from surgical preparation. Control measurements of CBF and other parameters, including $M\dot{V}_{O_2}$, were obtained during perfusion with isoflurane-free arterial blood with LAD perfusion pressure set equal to mean aortic pressure. With perfusion pressure maintained at the control level, the LAD perfusate was then switched to arterial blood equilibrated with 0.5% isoflurane, and when stable values for monitored local hemodynamic parameters (*e.g.*, electromagnetic measurements of CBF) indicated that steady-state conditions prevailed (5–10 min after the change in LAD perfusate), values were again obtained. This protocol was repeated for isoflurane concentrations of 1.0 and 2.0%. The order of isoflurane concentrations was randomized to avoid biasing the results. With perfusion from the isoflurane-free reservoir at normal pressure, adenosine (15 mM in isotonic saline) was infused intracoronarily at a dose sufficient to cause steady-state, maximal vasodilation (11.1 ± 2.5 μmol/min, corresponding to an infusion rate of 0.74 ± 0.17 ml/min). Maximal vasodilation was indicated when an increase in the rate of infusion of adenosine failed to cause a further increase in CBF. The CBF rate during adenosine infusion was used as a reference to assess the extent of coronary vasodilation by intracoronary isoflurane.

In four dogs, two maneuvers were carried out to determine whether vasoactive substances originating in the oxygenator, *e.g.*, old blood or structural contaminant, may have contributed to the increased blood flow during isoflurane administration. First, perfusion of the LAD was switched from the normal blood reservoir to a reservoir containing blood that passed through the oxygenator while the oxygenator was supplied with isoflurane-free gas. Second, while the LAD was being perfused by the isoflurane-equilibrated blood reservoir, the vaporizer was turned off so that isoflurane concentration in the coronary perfusate progressively declined.

Statistical Analyses

A completely randomized analysis of variance in combination with the Student-Newman-Keuls test[9] was used to evaluate effects of isoflurane and adenosine and effects of differences among preisoflurane and preadenosine control values. The Student's *t* test for paired samples[9] was used to evaluate effects of the switch from the normal blood reservoir to the oxygenator-supplied blood reservoir with the vaporizer off. A $P < 0.05$ was considered significant throughout this study.

Results

Figure 2 presents a representative tracing of monitored hemodynamic parameters during selective exposure of the LAD to 0.5% isoflurane; *A* indicates the point at which perfusion was switched to the isoflurane reservoir and *B* the point at which it was returned to the isoflurane-free reservoir. After a modest delay due to dead space in the inflow circuit, isoflurane caused a rapid and significant increase in CBF. Of note was that monitored hemodynamic parameters, including coronary perfusion pressure, were essentially constant during intracoronary administration of isoflurane. Figure 2 shows a rapid decline to the control level of CBF upon return to isoflurane-free blood.

Because supplementary bolus injections of fentanyl to decrease heart rate did not affect responses to intracoronary isoflurane, data for all studies were combined. Preisoflurane and preadenosine control values did not differ significantly. Therefore, for the sake of simplicity and brevity, the pooled means for all controls are presented in tables 1–3.

Table 1 summarizes changes in CBF during intracoronary administration of isoflurane and adenosine. Composition of the coronary arterial blood perfusate remained similar to control under all conditions, except that P_{O_2} was increased during isoflurane administration. Intracoronary isoflurane caused essentially dose-dependent increases in CBF, although the value at 1.0% was not significantly greater than that at 0.5%. Because the flow in-

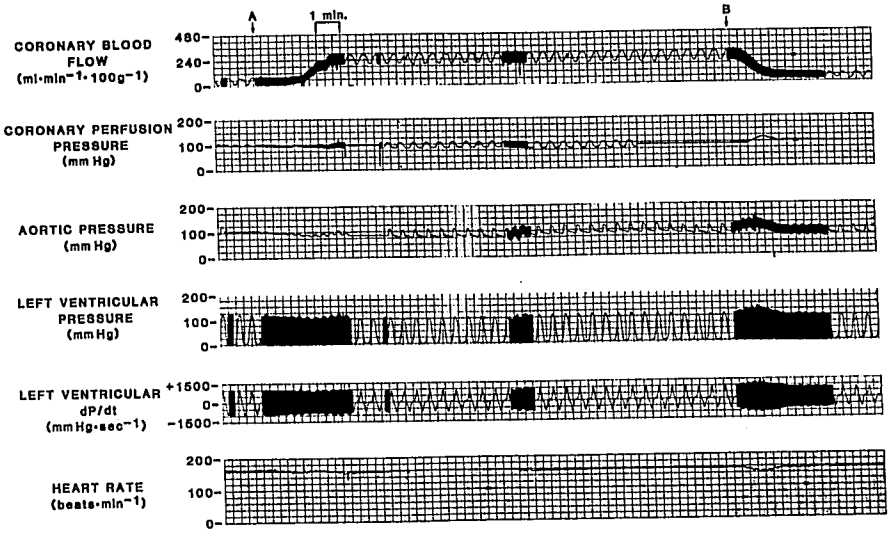

FIG. 2. Tracing demonstrating effect of intracoronary administration of arterial blood equilibrated with 0.5% isoflurane on monitored hemodynamic parameters. Note that the significant increase in coronary blood flow occurred in the presence of constant perfusion pressure and systemic hemodynamic parameter.

creases occurred in the presence of constant perfusion pressure, they reflected proportional decreases in vascular resistance. With 2.0% isoflurane, the increases in CBF were approximately 80% of the maximal adenosine-induced increases in CBF. The increases in CBF caused by isoflurane and adenosine were transmurally uniform, as indicated by constant values for the endocardium:epicardium flow ratio. Radioactivity in venous samples indicated that arteriovenous shunting of microspheres was less than 2.0% under control conditions and remained at that level during coronary vasodilation with either isoflurane or adenosine.

Table 2 summarizes changes in $M\dot{V}_{O_2}$ and related parameters during intracoronary administration of isoflurane or adenosine. Intracoronary isoflurane caused increases in coronary venous P_{O_2} and decreases in coronary arteriovenous oxygen difference, both of which were disproportionate to the induced blood flow increases, reflecting reductions in $M\dot{V}_{O_2}$. The increases in coronary venous P_{O_2} were remarkable (exceeding 100 mmHg with 2.0% isoflurane) and were consistent with the bright red, arterial appearance of the collected venous blood samples. During intracoronary adenosine, the decreases in the coronary arteriovenous oxygen difference were proportional to the increases in CBF resulting in no change in $M\dot{V}_{O_2}$.

Table 3 demonstrates that monitored hemodynamic parameters (mean aortic pressure, mean left atrial pressure, left ventricular dP/dt_{max}, and heart rate), as well as aortic blood gases, were stable during intracoronary administration of isoflurane or adenosine.

Results indicating lack of release of vasodilator substance from the oxygenator are presented in figure 3 and table 4. Figure 3 is an original tracing that demonstrates that changing vaporizer setting from 2.0 to 0% isoflurane while maintaining perfusion from oxygenator-supplied reservoir (fig. 3B) caused progressive decline in CBF; CBF eventually decreased to a level similar to that evident dur-

TABLE 1. Changes in Coronary Blood Flow during Intracoronary Administration of Isoflurane or Adenosine

Parameter	Control	Isoflurane (%)			Adenosine
		0.5	1.0	2.0	
Blood flow (ml·min^{-1}·100 g^{-1})	91 ± 3	338 ± 64*	345 ± 61*	549 ± 67*	688 ± 95*
Endo/epi ratio	1.3 ± 0.1	—	1.3 ± 0.2	1.0 ± 0.1	1.3 ± 0.1
Perfusion pressure (mmHg)	103 ± 2	101 ± 5	101 ± 3	102 ± 3	102 ± 3
Coronary artery values					
P_{O_2} (mmHg)	170 ± 14	499 ± 52*	464 ± 37*	472 ± 52*	182 ± 30
P_{CO_2} (mmHg)	38 ± 1	41 ± 2	41 ± 1	38 ± 1	37 ± 1
*p*H	7.37 ± 0.01	7.35 ± 0.01	7.35 ± 0.01	7.37 ± 0.01	7.38 ± 0.01
O_2 saturation (%)	95 ± 1	96 ± 1	97 ± 1	97 ± 1	93 ± 2
O_2 content (vol %)	17.8 ± 0.4	19.1 ± 1.0	19.0 ± 0.5	18.9 ± 0.6	17.7 ± 0.9
Hemoglobin (g·100 ml^{-1})	13.0 ± 0.2	13.3 ± 0.6	13.1 ± 0.3	13.1 ± 0.4	13.0 ± 0.5

Values are mean ± SE in 11 dogs, except for the endo/epi ratio, which was obtained in 7 dogs.

* $P < 0.05$, compared to control.

TABLE 2. Changes in Myocardial Oxygen Consumption and Related Parameters during Intracoronary Administration of Isoflurane or Adenosine

Parameter	Control	Isoflurane (%)			Adenosine
		0.5	1.0	2.0	
Oxygen consumption (ml · min^{-1} · 100 g^{-1})	8.1 ± 0.4	5.1 ± 1.3*	5.4 ± 1.0*	4.3 ± 1.1*	8.1 ± 1.3
Blood flow (ml · min^{-1} · 100 g^{-1})	92 ± 4	230 ± 33*	269 ± 37*	446 ± 48*	550 ± 79*
A-V O_2 difference (vol %)	8.8 ± 0.4	2.7 ± 1.0*	2.3 ± 0.5*	1.1 ± 0.4*	1.6 ± 1.3*
O_2 extraction (%)	50 ± 2	15 ± 6*	12 ± 3*	6 ± 2*	9 ± 2*
Coronary venous values					
P_{O_2} (mmHg)	32 ± 1	70 ± 8*	83 ± 9*	130 ± 21*	71 ± 7*
P_{CO_2} (mmHg)	45 ± 1	44 ± 2	45 ± 2	43 ± 1	39 ± 2
*p*H	7.34 ± 0.01	7.35 ± 0.02	7.32 ± 0.01	7.34 ± 0.01	7.36 ± 0.03
O_2 saturation (%)	48 ± 2	85 ± 7*	89 ± 3*	94 ± 2*	88 ± 3*
O_2 content (vol %)	8.8 ± 0.5	15.4 ± 1.5*	16.4 ± 0.9*	17.0 ± 0.9*	17.7 ± 0.9*

Values are mean ± SE in seven dogs. * $P < 0.05$, compared to control.

ing perfusion from normal blood reservoir (figure 3A). Table 4 shows that switching from normal blood reservoir to oxygenator-supplied reservoir with the vaporizer off had no effect on CBF.

The coronary arterial blood concentrations for isoflurane were 3.0 ± 0.2, 5.3 ± 0.8, and 10.8 ± 1.1 mg · 100 ml^{-1} for 0.5, 1.0, and 2.0% isoflurane, respectively.

Discussion

CRITIQUE OF METHODS

The canine preparation used to perfuse selectively the LAD at controlled pressure has been used previously to evaluate direct coronary vasomotor effects of drugs, including adenosine,[10] and effects of physiologic factors, including hypoxemia and hemodilution.[11,12] This preparation results in control values for hemodynamic, metabolic, and functional variables in the perfused bed that are similar to those in the naturally supplied coronary bed. Furthermore, normal responsivity to vasodilating stimuli (as evidenced by the marked increases in blood flow during adenosine or isoflurane in the present study) is preserved.

Validation studies demonstrated that the extracorporeal oxygenator used to equilibrate blood with isoflurane did not elaborate a substance with coronary vasomotor effects. Also, it was demonstrated that the isoflurane concentration of blood leaving the oxygenator was proportional to the percentage of isoflurane provided by the vaporizer and that they were in the range of values found in humans during general anesthesia.[13]

While the LAD was perfused with isoflurane-equilibrated blood venous effluent containing isoflurane (the portion not collected *via* the inplanted coronary venous catheter) returned directly to the systemic circulation. However, because of 1) the short duration of intracoronary administration of isoflurane, 2) the relatively small size of this coronary venous return compared to the total system venous return, and 3) the clearance of isoflurane at the lungs, we assumed that isoflurane concentration in the systemic arterial circulation was insufficient to affect findings in the LAD bed. Although measurements of end-tidal isoflurane were not available to confirm this as-

TABLE 3. Stability of Systemic Hemodynamic Parameters during Intracoronary Administration of Isoflurane or Adenosine

Parameter	Control	Isoflurane (%)			Adenosine
		0.5	1.0	2.0	
Mean aortic pressure (mmHg)	103 ± 16	106 ± 6	105 ± 5	110 ± 6	105 ± 6
Mean left atrial pressure (mmHg)	5.3 ± 0.4	5.1 ± 0.7	6.0 ± 0.9	6.1 ± 0.8	5.6 ± 0.8
dP/dt_{max} (mmHg · min^{-1})	905 ± 43	925 ± 88	897 ± 68	875 ± 88	890 ± 73
Heart rate (beats per min)	138 ± 7	138 ± 16	139 ± 10	129 ± 12	122 ± 10
Aortic values					
P_{O_2} (mmHg)	185 ± 16	180 ± 30	205 ± 26	207 ± 39	172 ± 33
P_{CO_2} (mmHg)	37 ± 1	37 ± 1	38 ± 1	39 ± 2	38 ± 2
*p*H	7.38 ± 0.01	7.38 ± 0.01	7.36 ± 0.01	7.36 ± 0.02	7.36 ± 0.02
Hematocrit (%)	38 ± 1	38 ± 2	39 ± 1	38 ± 2	38 ± 2

Values are mean ± SE in 11 dogs.

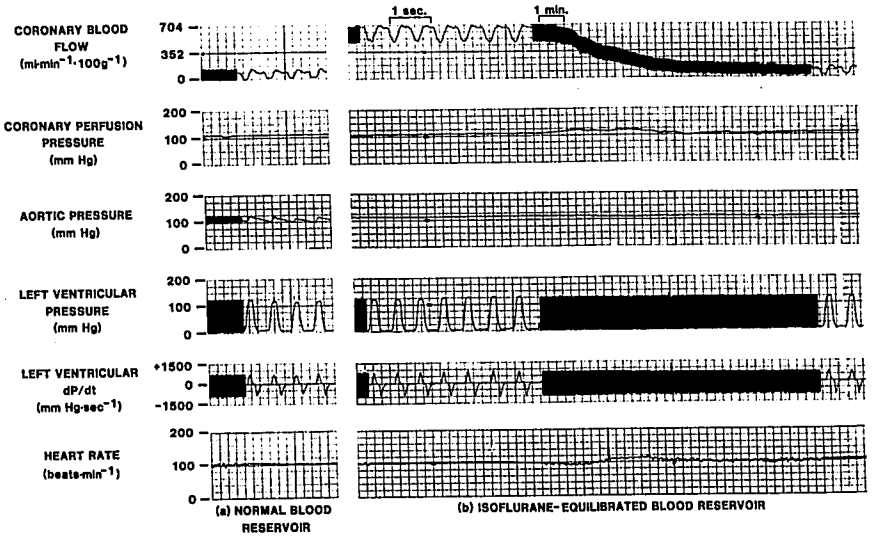

FIG. 3. Tracing demonstrating that changing vaporizer setting from 2.0 to 0% isoflurane while maintaining perfusion from oxygenator-supplied reservoir (*b*) caused progressive decline in coronary blood flow from the peak isoflurane-induced level; coronary blood flow eventually decreased to a level similar to that evident during perfusion from normal blood reservoir (*a*). In this dog, heart rate was maintained at 100 beats per min by bolus injections of fentanyl.

sumption, two lines of evidence are consistent with maintained low systemic isoflurane concentrations. First, systemic hemodynamic parameters did not vary during intracoronary administration of isoflurane. Second, CBF declined rapidly to control values when perfusion was returned to the normal blood reservoir (see fig. 2), which received blood directly from the aorta.

The validity of the determinations of coronary arteriovenous shunting of microspheres and of $M\dot{V}_{O_2}$ required that the blood samples from the anterior interventricular vein be representative of the venous effluent from LAD-dependent myocardium. This was verified by previous studies that mapped left coronary venous drainage patterns using inert gas tracers[14] or chromium-labeled red blood cells.[15]

The radioactive microsphere method can provide reliable measurements of regional blood flow if 1) the microspheres and reference sample collection themselves do not influence coronary hemodynamics; 2) the microspheres are well mixed in the coronary blood supply; 3) coronary arteriovenous shunting of microspheres is insignificant; and 4) at least 400 microspheres are present in tissue samples.[16] With regard to the first condition, no changes in total CBF measured by electromagnetic flowmeter or other continuously monitored hemodynamic parameters accompanied the microsphere injections or withdrawal of reference samples, implying unaltered local coronary flow conditions. Second, radioactivity of the duplicate reference samples differed by less than 5%, indicating that the microspheres were well mixed. Third, samples of venous blood obtained from the LAD perfusion field contained negligible radioactivity, indicating no significant shunting of microspheres. Finally, sufficient numbers of microspheres were recovered in all samples for low-error, high-precision flow measurements. Sufficient counts were accumulated for all samples to maintain counting statistical errors of less than 3%.

TABLE 4. Lack of Influence of Switch from Normal Blood Reservoir (Reservoir 1) to Oxygenator-supplied Blood Reservoir with Vaporizer off (Reservoir 2) on Coronary Blood Flow

Parameter	Reservoir 1	Reservoir 2
Blood flow (ml · min^{-1} · 100 g^{-1})	105 ± 9	111 ± 8
Perfusion pressure (mmHg)	100 ± 9	99 ± 10
Mean aortic pressure (mmHg)	94 ± 12	94 ± 14
Heart rate (beats per min)	120 ± 10	120 ± 7
Coronary artery values		
P_{O_2} (mmHg)	177 ± 71	522 ± 69*
P_{CO_2} (mmHg)	35 ± 1	34 ± 1
*p*H	7.42 ± 0.03	7.42 ± 0.03
Hematocrit (%)	40 ± 3	41 ± 2

Values are mean ± SE in four dogs.
* $P < 0.05$, compared to control.

A limitation of the current animal model was that coronary effects of isoflurane were superimposed on baseline anesthesia. The decision to combine pentobarbital with fentanyl was based on the large quantities of fentanyl alone required to achieve a stable anesthetic state in dogs.[17] The high heart rates in our earlier studies prompted the use of supplementary bolus injections of fentanyl to achieve heart rates closer to basal levels. Coronary vasodilator responses during intracoronary isoflurane were not altered by the increased doses of fentanyl (fig. 3).

CORONARY EFFECTS OF ISOFLURANE

To attribute the observed increases in CBF to a direct relaxing effect of isoflurane on vascular smooth muscle, it is necessary to rule out other factors. First, it appears that reduced coronary vascular resistance was not due to

changes in composition of coronary arterial blood, since values for P_{CO_2}, *p*H, and hematocrit were constant. Although P_{O_2} was higher during administration of isoflurane because of more efficient gas exchange in the extracorporeal oxygenator compared to the canine lung, it was sufficient under all conditions for essentially complete oxygen saturation of hemoglobin, resulting in similar coronary arterial oxygen content in the absence and presence of isoflurane. Furthermore, hyperoxia has been demonstrated to cause coronary vasoconstriction rather than vasodilation.[4] Thus, increased coronary arterial P_{O_2} would have been expected to antagonize rather than to augment vasodilator responses during isoflurane. Second, because $M\dot{V}O_2$ decreased rather than increased during isoflurane administration, coronary vasodilation was not maintained by a metabolic mechanism. Indeed, the potency of isoflurane's coronary vasodilator effect is evidenced by its ability to cause significant increases in CBF despite a reduced metabolic requirement for blood flow. Third, heart rate and left atrial pressure (a reflection of left ventricular end-diastolic pressure) were constant during isoflurane administration, which suggests that reduced extravascular compression of subendocardium did not contribute to the observed increases in CBF.[4] This was reflected in the constant values for the endocardial:epicardial flow ratio.

Inhaled isoflurane has been demonstrated to reduce $M\dot{V}O_2$ in human patients[1] and in canine models.[5] However, since in these studies isoflurane caused changes in other hemodynamic determinants of myocardial oxygen demand, including heart rate and aortic pressure, it was not possible to identify the contribution of reduced myocardial contractility to the decrease in $M\dot{V}O_2$. In the current study, the isoflurane-induced decreases in $M\dot{V}O_2$ occurred in the presence of constant heart rate and aortic pressure, and thus they were due presumably to decreases in myocardial contractility in the LAD perfusion field. Since this region comprised a limited portion of the total left ventricular free wall, dP/dt_{max} was unaffected. This ability of isoflurane to cause direct cardiac depression in the intact heart is consistent with previous observations in isolated preparations of cardiac muscle *in vitro*[18] and in isolated, paced, working rat hearts.[19]

Intracoronary isoflurane caused remarkable increases in local venous P_{O_2}. These high venous P_{O_2} values were not due to redirection of blood flow through large arteriovenous anastomoses, *i.e.*, nonexchange vessels, since no increase in the number of shunted microspheres was observed. They were also not due to inadequate time for oxygen unloading of hemoglobin resulting from the isoflurane-induced flow increases, since extraction was not reduced further at the higher rate of flow during adenosine infusion. The high venous P_{O_2} values during isoflurane apparently simply reflected the combined effects of significantly increased oxygen supply, *i.e.*, CBF, and reduced myocardial oxygen demand.

The current study provided no information on the vascular site for isoflurane-induced coronary dilation. However, several lines of evidence suggest that the primary site for coronary dilation was probably the arteriolar resistance vessels rather than the large epicardial conduit vessels. First, Sill *et al.* found no effect of 0.75–2.25% end-tidal isoflurane on epicardial coronary diameter assessed by computerized analysis of arteriograms in dogs.[3] Second, Blaise *et al.* reported no reduction in basal tension of isolated canine coronary arterial strips exposed in vitro to 2.3% isoflurane.[20] Finally, in normal coronary circulations, basal vascular tone of the epicardial arteries is low, and these vessels contribute only 2–5% of the total coronary vascular resistance.[21] This implies that even if isoflurane caused relaxation of vascular smooth muscle in large coronary arteries, this action could account for only a small portion of the increases in CBF observed.

The mechanism by which isoflurane relaxes vascular smooth muscle is unknown, although impairment to Ca^{2+} movement or availability at the cell membrane, sarcoplasmic reticulum, and contractile proteins has been proposed.[22] Recent *in vitro* studies of canine arterial segments preconstricted pharmacologically have suggested that isoflurane-induced coronary relaxation requires an intact endothelium, suggesting a role for endothelium-derived relaxing factor released from these vessels.[20]

The lack of change in local $M\dot{V}O_2$ during intracoronary adenosine has been demonstrated previously,[10] and it supports the recent report that intravenous adenosine had no direct inotropic effect in the left ventricular myocardium of the conscious dog.[23]

In conclusion, the current study demonstrated that isoflurane causes significant dose-dependent coronary vasodilation *via* a direct effect of vascular smooth muscle, most likely at the arteriolar level. The potency of the coronary vasodilator action of isoflurane was evidenced by an increase in CBF that was nearly equal to that during maximal vasodilation, a prototype small-vessel coronary dilator.

The current findings are qualitatively consistent with those of Sill *et al.*, who demonstrated significant coronary vasodilation during inhalation of isoflurane by fentanyl–pentobarbital-anesthetized dogs,[3] although the magnitude of the flow increases we observed were greater. An explanation for this quantitative difference has not been determined with certainty. One factor may be the reductions in aortic pressure and global cardiac work demand in the study of Sill *et al.*,[3] which complicated identification of changes in CBF due to isoflurane itself. Another factor may be the different time at which blood flow measurements were obtained in the two studies. In the current

study, measurements of CBF were obtained immediately upon achieving steady-state conditions following abrupt exposure of the coronary circulation to blood equilibrated with isoflurane, whereas in the study of Sill *et al.* measurements were made 25–30 min after beginning inhalation of isoflurane, which provided time for moderating mechanisms to attenuate the direct dilator effects of isoflurane.

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References

1. Reiz, S, Balfors E, Soreson MB, Ariola S, Jr, Friedman A, Trudesson H: Isoflurane: A powerful coronary vasodilator in patients with coronary artery disease. ANESTHESIOLOGY 59:91–97, 1983
2. Gross GJ, Warltier DC: Coronary steal in four models of single or multiple vessel obstruction in dogs. Am J Cardiol 48:84–92, 1981
3. Sill JC, Bove AA, Nugent M, Blaise GA, Dewey JD, Graban C: Effects of isoflurane on coronary arteries and coronary arterioles in the intact dog. ANESTHESIOLOGY 66:273–279, 1987
4. Feigl EM: Coronary physiology. Physiol Rev 63:1–205, 1983
5. Merin RG: Are myocardial functional and metabolic effects of isoflurane really different from those of halothane and enflurane? ANESTHESIOLOGY 55:398–408, 1981
6. Weibel ER: The Pathway for Oxygen. Cambridge, Harvard University Press, 1984, pp 138–151
7. Crystal GJ: Coronary hemodynamic responses during local hemodilution in canine hearts. Am J Physiol 254:H525–H531, 1988
8. Yamamura H, Wakasugi B, Sato S, Takebe Y: Gas chromatographic analysis of inhalational anesthetics in whole blood by an equilibration method. ANESTHESIOLOGY 27:311–317, 1966
9. Zar JH. Biostatistical Analysis. Englewood Cliffs, Prentice-Hall, 1974
10. Crystal GJ, Downey HF, Bashour FA: Small vessel and total coronary blood volume during intracoronary adenosine. Am J Physiol 241:H194–H210, 1981
11. Downey HF, Crystal GJ, Bockman EL, Bashour FA: Non-ischemic myocardial hypoxia: Coronary dilation without increased tissue adenosine. Am J Physiol 243:H512–H516, 1981
12. Crystal GJ, Salem MR: Myocardial oxygen consumption and segmental shortening during selective coronary hemodilution in dogs. Anesth Analg 67:500–508, 1988
13. Holady DA, Fiserova-Bergerova V, Latto IP, Zumbiel MA: Resistance of isoflurane to biotransformation in man. ANESTHESIOLOGY 43:325–332, 1975
14. Roberts DL, Nakazawa HK, Klocke FJ. Origin of great cardiac vein and coronary sinus drainage within the left ventricle. Am J Physiol 230:486–492, 1976
15. Vinten-Johannsen J, Johnston WE, Crystal GJ, Mills SA, Santamore WP, Cordell AR: Validation of local venous sampling within at-risk left ventricular descending vascular bed in the canine left ventricle. Cardiovasc Res 21:646–656, 1987
16. Heyman MA, Payne BD, Hoffman JIE, Rudolph AM: Blood flow measurement with radionuclide-labelled particles. Prog Cardiovasc Dis 20:55–88, 1978
17. Stanley TH, Port JD: Fentanyl "anesthesia" in dogs. ANESTHESIOLOGY 62:837–838, 1985
18. Lynch C: Differential depression of myocardial contractility by halothane and isoflurane *in vitro*. ANESTHESIOLOGY 64:620–631, 1986
19. Sahlman L, Henriksson B-A, Martner J, Ricksten S-E: Effects of halothane, enflurane, and isoflurane on coronary vascular tone, myocardial performance, and oxygen consumption during controlled changes in aortic and left atrial pressure. ANESTHESIOLOGY 69:1–10, 1988
20. Blaise G, Still JC, Nugent M, Van Dyke RA, Vanhoutte PM: Isoflurane causes endothelium-dependent inhibition of contractile responses of canine coronary arteries. ANESTHESIOLOGY 67: 513–517, 1987
21. Young AA, Vatner SF: Regulation of large coronary arteries. Circ Res 59:579–596, 1986
22. Bollen BA, Tinker JH, Hermsmeyer K: Halothane relaxes previously constricted isolated porcine artery segments more than isoflurane. ANESTHESIOLOGY 66:748–752, 1987
23. Freeman GL, Colston JT, Hultman: Influences of adenosine on left ventricular performance in conscious dogs. Am J Physiol 258:H424–H430, 1990