The Influence of Isoflurane on the Vascular Reflex
Response to Lung Inflation in Dogs

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Positive pressure ventilation can affect hemodynamic stability by neuroreflex-mediated activity. Inhalational anesthesia is known to attenuate the arterial baroreflex function; however, little information is known about the effect of volatile anesthetics on the lung inflation reflex. The influence of isoflurane on static lung inflation reflex-induced changes in venous capacitance and systemic resistance was investigated in dogs. After controlling carotid sinus pressure at 50 mmHg and initiating total cardiopulmonary bypass, the lungs were inflated to tracheal pressures of 10 and 20 mmHg. The systemic vascular resistance index (SVRI) decreased by 0.04 ± 0.03 and 0.13 ± 0.03 mmHg · kg⁻¹ · min⁻¹ · ml⁻¹ during tracheal inflation pressures of 10 and 20 mmHg, respectively. There was an accompanying change in systemic vascular capacitance index (SVCI) by 1.0 ± 0.65 and 3.3 ± 0.82 ml · kg⁻¹ during tracheal inflation pressures of 10 and 20 mmHg. The addition of isoflurane decreased the reflex vascular response to lung inflation in a dose-dependent manner. A concentration of 1 MAC isoflurane administered via the cardiopulmonary bypass machine attenuated the change in SVRI to tracheal inflation pressures of 10 and 20 mmHg by 75% and 67%, respectively. Isoflurane at 1 MAC also reduced the reflex capacitance response to tracheal pressures of 10 and 20 mmHg by 36% each. Lung inflation-induced changes in SVRI and SVCI were abolished at isoflurane concentrations of 2 MAC. We conclude that under the conditions of this study, 1 MAC isoflurane was shown to attenuate lung reflex-induced changes in SVRI and SVCI and that at higher isoflurane concentrations (2 MAC) these reflex-induced changes were not seen. (Key words: Anesthetics, volatile: isoflurane. Blood pressure. Circulation: central venous pressure. Reflex: lung inflation. Species: dog.)

Positive pressure ventilation can have a significant effect on the cardiovascular system. The increase in intrathoracic pressure is known to decrease venous return,¹ ² right and left ventricular end-diastolic transmural pressures,³⁴ and right and left atrial end-diastolic transmural pressures⁵⁶ and to shift the interventricular septum to the left.⁷ Lung inflation by positive pressure ventilation can also affect hemodynamic stability by neuroreflex-mediated activity. Animal experiments have shown that low levels of static lung inflation can induce an excitatory reflex with tachycardia and vasconstriction⁸⁹ and that higher levels of lung inflation produce a depressor reflex on heart rate,⁰¹ contractility,¹²¹³ and systemic vascular resistance¹²¹⁴¹⁵ and an increase in systemic vascular capacitance.¹⁵ Because almost all patients undergoing a general anesthetic receive positive pressure ventilation, often with large tidal volumes, the lung inflation reflex may be an important factor affecting perioperative hemodynamics.

Inhalational anesthesia is known to attenuate the arterial baroreflex function, which is an important neural control system for maintaining hemodynamic stability.¹⁶¹⁷¹⁸ However, little is known about the effect of volatile anesthetics on lung inflation–neuromediated changes in the systemic circulation. In the general healthy patient, alteration of baroreflex by an inhalational anesthetic is not a major problem because larger variations in blood pressure can be tolerated, but in the elderly and other patients with perioperative hemodynamic instability, changes in any baroreceptor activity may compromise tissue perfusion.

A frequently used volatile anesthetic, isoflurane, has been shown to have minimal effect on the arterial baroreceptor reflex at common clinical concentrations.¹⁸¹⁹ This study was designed to analyze the effect of isoflurane on the static lung inflation reflex influence on the systemic circulation. Changes in mean arterial pressure (MAP), central venous pressure (CVP), systemic vascular resistance index (SVRI), and systemic vascular capacitance index (SVCI) were assessed in anesthetized dogs during cardiopulmonary bypass and positive pressure ventilation.

Materials and Methods

All experimental procedures and protocols in this investigation were reviewed and approved by the Animal Care Committee of the Medical College of Wisconsin, and all conformed to the Guiding Principles in the Care and Use of Animals of the American Physiologic Society. Seven mongrel dogs (23 ± 1.6 kg) were anesthetized with sodium pentobarbital (Nembutal, Abbott) 30 mg · kg⁻¹ intravenously. Supplemental doses of anesthetic were given as necessary throughout the experiment in order.
to maintain the anesthetic state as determined by observation of changes in autonomic responses to painful stimuli associated with surgery or spontaneous body movement. After tracheal intubation with a cuffed endotracheal tube, the lungs were ventilated using a Bird Mark 7 respirator. Systemic arterial pressure and CVP were measured through catheters placed in the thoracic aorta and the inferior vena cava via the right axillary artery and right femoral vein, respectively, and connected to Statham pressure transducers. Prior to cardiopulmonary bypass, body temperature was maintained by external warming with a heating pad and a heating lamp.

Closed loop perfusion of the carotid sinus was performed by isolating the left common carotid artery and ligating all branches, including the internal and external carotid arteries, thyroid artery, occipital artery, and lingual artery. The common carotid artery was then cannulated in a rostral direction while the external carotid artery was cannulated in a caudal direction, effectively isolating the carotid sinus between the two cannulae. The carotid chemoreceptors were eliminated from the isolated perfused side by ligating the occipital artery at its junction with the carotid artery, as described by Stephenson and Donald.80 The right carotid sinus and body were denervated by direct compression and the local application of alcohol. Denervation was verified by the absence of heart rate or systemic arterial blood pressure response to occlusion of the right common carotid artery.

The left and right cervical vagi and the aortic nerves were isolated in the neck using the method of Edis and Shepherd.81 The cervical aortic nerves were visually identified and then verified by nerve recording with bipolar electrodes, after which the aortic depressor nerves were cut just distal to the nodose ganglion.

**Cardiopulmonary Bypass and Left Carotid Artery Perfusion**

The chest was opened through a median sternotomy, and cardiopulmonary bypass was instituted using a Sarns roller pump (model 3500) and Harvey reservoir oxygenator. The reservoir was primed with a 2:1 mixture of lactated Ringers solution and dextran 40 with 10,000 U sodium heparin (Upjohn) added to prevent coagulation. In addition, sodium heparin (500 U·kg⁻¹) was administered to the animals before cannulation of arteries and veins and the start of perfusion. Arterial perfusion cannulae were placed in the femoral arteries. The inferior vena cava was ligated below the renal veins. Four large-bore cannulae were placed so that venous return from the splanchnic and extrasplanchnic vascular beds could be separated and individually measured (fig. 1). One cannula was placed into the inferior vena cava to the level of the diaphragm to collect the splanchnic and renal venous outflow. In all subsequent references to splanchnic venous outflow, it must be understood that this technique includes both splanchnic and renal venous outflow. The other three cannulae were placed into the superior vena cava, right ventricle through the right atrial appendage, and femoral vein to collect the extrasplanchnic venous outflow. The left ventricle was vented using a left ventricular cannula to collect venous return to the left side of the heart. The splanchnic and extrasplanchnic venous outflows were separately connected to the reservoir to avoid an interaction between venous outflows. The azygos vein was then ligated. The cardiopulmonary bypass pump was adjusted so that arterial blood pressure was approximately equal to that before bypass and maintained constant throughout the experiment. The pump rollers were ad-
justed to be completely occlusive to maintain constant cardiac output during conditions in which peripheral resistance was changing. The height of the opening of the tube draining venous return into the reservoir was adjusted so that mean CVP measured 5–6 mmHg. Zero pressure was referenced at the junction of the inferior vena cava and the right heart under direct inspection.

The vascularly isolated left carotid sinus was perfused by a Sarns roller pump, using blood drawn from the reservoir oxygenator, through the common carotid artery with outflow collected from the external carotid artery and returned to the reservoir oxygenator. Intrasinus pressure was measured by means of a lingual artery catheter advanced into the sinus. A screw-clamp resistor was placed on the outflow cannula to permit adjustments in carotid sinus pressure during constant flow perfusion.

Extrapulmonary and splanchnic–renal venous outflows were measured using a multichannel Biotronix electromagnetic flowmeter and in-line flow probes. Zero-flow signals were recorded using a bypass circuit to enable calibration before and during the experiments if necessary. The flow signal was displayed on a Grass polygraph and diverted into a filter–amplifier that permitted expansion of the peak flow signal into an amplified trace from which the changes could be measured more accurately. The mean flow and amplified flow recording were calibrated at the end of the experiment using the Sarns pump and timed collected flow.

Blood gases were measured before and during cardiopulmonary bypass using blood samples obtained from the brachial artery cannula or the pump reservoir during perfusion. A Radiometer ABL-1 blood gas machine was used for blood gas and pH analysis. Blood gases and pH were adjusted using carbon dioxide and oxygen flowmeters attached to the oxygenator and by the addition of sodium bicarbonate to the reservoir as required to maintain PaO₂ > 200 mmHg, PaCO₂ 30–40 mmHg, and pH 7.40–7.45.

**Lung Inflation and Anesthetic Administration**

After cardiopulmonary bypass was initiated, a solenoid ventilator as used by Nilsestuen et al.²² provided baseline mechanical ventilation of 10 breaths·min⁻¹ at tracheal pressures of 5 mmHg. The ventilator was programmed to inflate the lungs to constant tracheal pressures of 10 or 20 mmHg in random order for 70 s with approximately 5 min of baseline ventilation between static inflations of the lung during each testing period.

The baseline peak lung inflation pressure of 5 mmHg was chosen to keep pressures below the threshold of stimulation of receptors with C-fiber afferents.¹⁴,²² Static inflation pressures of 10 and 20 mmHg were chosen to ensure that large tidal volumes were delivered. The mean inspired volume at a tracheal pressure of 10 mmHg was 10.8 ± 4.5 ml/kg and at 20 mmHg was 42.4 ± 5.9 ml/kg. A 70-s inflation time was chosen to allow venous outflow to return to a steady state.

A precalibrated vaporizer was used in-line on the cardiopulmonary bypass circuit to provide 1- and 2-MAC concentrations of isoflurane. Blood concentrations of isoflurane were determined by gas chromatography (Perkin-Elmer, St. Louis, MO). Blood samples were drawn from the brachial artery cannula just before the inhalation anesthetic was started, after 20 min exposure to each concentration of isoflurane, and 30 min after isoflurane was discontinued.

**Procedure and Measurements**

Aortic blood pressure, carotid sinus pressure, extrapulmonary and splanchnic venous pressures, extrapulmonary and splanchnic venous outflows, and amplified venous return flows were recorded simultaneously on a Grass model 7 polygraph and a Vetter eight-channel tape recorder for later analysis.

Changes in blood volume within the animal were measured during increases or decreases in venous outflow (constant cardiac output) by integrating the area under the total venous return flow, which was obtained as the sum of the extrapulmonary and splanchnic venous outflows, using a Science Accessories sonic digitizer.

After 20 min of extracorporeal circulation, with MAP maintained similar to prebypass values and at a set carotid sinus pressure of 50 mmHg, which was selected because low carotid sinus pressures augment changes in vascular capacitance and resistance due to lung inflation,¹³ the lungs were inflated in random order to static tracheal pressures of 10 and 20 mmHg for 70 s each, with approximately 5 min of baseline mechanical ventilation between static lung inflations. Isoflurane was then started at 1 MAC. This concentration was maintained for approximately 20 min with at least 3 min of steady-state hemodynamics before the lungs again were inflated to tracheal pressures of 10 and 20 mmHg. The isoflurane concentration was then increased to 2 MAC for approximately 20 min with at least 3 min of steady-state hemodynamics before the lungs were inflated to tracheal pressures of 10 and 20 mmHg. The isoflurane was discontinued for 30 min, and static lung inflations were repeated.

**Data Analysis**

The data are expressed as mean ± SEM. Changes in SVRI and SVCI secondary to lung inflation at 10 or 20 mmHg of tracheal pressure at a carotid sinus pressure of 50 mmHg were assessed using two-way analysis of vari-
ance. After a significant F test, Duncan’s test was performed. A level of \( P < 0.05 \) was chosen as significant.

**Results**

The average perfusion flow rate during cardiopulmonary bypass was 161 ± 17 ml·min\(^{-1} \)·kg\(^{-1} \). The mean baseline flows in the splanchnic and extraplanchnic vascular beds were 95.6 ± 11.7 and 66.0 ± 8.2 ml·min\(^{-1} \)·kg\(^{-1} \), respectively. The baseline SVRI was 0.64 ± 0.1 mmHg·kg·min·ml\(^{-1} \), and MAP was 99 ± 5.9 mmHg.

A typical response to lung inflation at a tracheal pressure of 20 mmHg and with exposure to 2 MAC isoflurane is shown in figure 2. Lung inflation to a pressure of 20 mmHg caused a decrease in MAP and splanchnic venous outflow and an increase in extraplanchnic venous outflow. Both venous outflows and MAP returned to baseline 70 s after lung inflation was initiated. With the administration of 2 MAC isoflurane, those changes produced by lung inflation were essentially eliminated.

The effect of isoflurane on the lung inflation–induced changes in systemic blood volume are shown in figure 3. Without isoflurane, systemic blood volume significantly increased, by 1 ± 0.65 and 3.3 ± 0.82 ml·kg\(^{-1} \) at tracheal pressures of 10 and 20 mmHg, respectively. One MAC isoflurane significantly attenuated this reflex increase in systemic blood volume, to 0.64 ± 0.45 ml·kg\(^{-1} \) at lung inflation to a tracheal pressure of 10 mmHg and to 2.11 ± 0.64 ml·kg\(^{-1} \) at a tracheal pressure of 20 mmHg.

Static lung inflation to tracheal pressures of 10 and 20 mmHg decreased SVRI by 0.04 ± 0.03 and 0.18 ± 0.05 mmHg·kg·min·ml\(^{-1} \), respectively, with a significant change reached with the higher inflation pressure (fig. 4). With the addition of 1 MAC isoflurane there

**Fig. 2.** Effect of isoflurane on the responses to lung inflation. The controls show the dramatic decrease in arterial pressure, decrease in splanchnic flow, and increase in extraplanchnic flow coincident with lung inflation (20 mmHg). There were no changes in blood pressure (BP), splanchnic flow, or extraplanchnic flow at the same lung inflation pressure in the same animal when 2 MAC isoflurane was added. CSP = carotid sinus pressure; CVP = central venous pressure; SVC = superior vena cava; IVC = inferior vena cava.

**Fig. 3.** The effect of lung inflation on systemic blood volume with increasing isoflurane concentration. The transitory volume change caused by lung inflation to 20 mmHg is less at 1 MAC (*\( P < 0.05 \)) and 2 MAC (**\( P < 0.05 \)) isoflurane compared to control (no inhalational anesthetic). With lung inflation to 10 mmHg, there were no differences in systemic blood volume changes between control and 1 or 2 MAC isoflurane.

**Fig. 4.** The effect of lung inflation on systemic vascular resistance (SVR) with increasing isoflurane concentration. Lung inflation to 20 mmHg resulted in significant decreases in systemic vascular resistance changes between control and 1 MAC (*\( P < 0.05 \)), control and 2 MAC (**\( P < 0.05 \)), and 1 MAC and 2 MAC isoflurane (**\( P < 0.05 \)). With lung inflation to 10 mmHg, there were no differences in SVR changes between control and 1 or 2 MAC isoflurane.
was no noticeable decrease in SVRI (0.01 ± 0.01 mmHg·kg·min·ml⁻¹) at a lung inflation pressure of 10 mmHg. However, when the lung was inflated to a tracheal pressure of 20 mmHg, there was a smaller but still significant decrease in SVRI of 0.06 ± 0.2 mmHg·kg·min·ml⁻¹. No lung inflation changes in SVCI or SVRI were seen when isoflurane was administered at 2 MAC. When isoflurane was discontinued for 30 min and the lungs were inflated to tracheal pressures of 10 and 20 mmHg, the changes in SVCI and SVRI were not significantly different from preanesthetic values. Lung inflation caused a dose-dependent change in MAP, as shown in figure 5. At an isoflurane concentration of 2 MAC, minimal change in MAP is seen with lung inflation to tracheal pressures of 10 and 20 mmHg. There were no significant differences in isoflurane blood levels among animals. The mean isoflurane concentrations at 1 and 2 MAC were 0.09 ± 0.03 and 1.3 mM, respectively. Isoflurane was undetectable in blood 30 min after administration of the anesthetic was stopped.

Discussion

The cardiovascular system is influenced by many neural- and humoral-mediated reflexes. The decrease in vascular capacitance and resistance from sustained lung inflation is believed to be caused by a decrease in sympathetic efferent nerve activity.\textsuperscript{15,24} This lung inflation reflex is probably mediated through receptors whose afferent pathways are primarily in the vagus nerve, because bilateral sectioning of pulmonary branches of the vagus nerves or pulmonary hilar denervation abolishes this response, as has been shown by other investigators\textsuperscript{25} and a previous study in our laboratories.\textsuperscript{15}

Volatile anesthetics can depress the arterial baroreflex, which is an important mechanism in maintaining hemodynamic stability.\textsuperscript{19,26-28} It has been suggested that of the currently available volatile anesthetics, isoflurane has the least depressive effect on baroreceptor function.\textsuperscript{18,29} However, little is known about volatile anesthetic effects on circulatory responses to the lung inflation reflex in the anesthetized dog. The results of this study indicate that the lung inflation reflex is also inhibited by isoflurane, as seen by decreased changes in SVCI and SVRI, with sustained lung inflation at 1 MAC isoflurane and with the reflex circulatory changes essentially eliminated at 2 MAC isoflurane.

When studying the circulatory changes due to a sustained lung inflation reflex, it is important to eliminate or minimize mechanical effects and the influence of different components of the baroreceptor reflex arc, including the receptors, afferent and efferent nerve pathways, central integratory centers, peripheral ganglia, and the heart. Moreover, there is a potential for obstructing venous outflow with large sustained lung inflation. To minimize this obstruction of venous outflow, we used large-bore venous catheters to collect venous outflow and performed a long median sternotomy with maximal separation of sternal split. There were no significant changes in CVPs with lung inflation to tracheal pressures of 10 mmHg. However, tracheal pressures of 20 mmHg caused CVP to increase by 2–4 mmHg, after which CVP returned to baseline after discontinuation of static lung inflation. This increase in venous pressure, in itself, can cause an increase in SVCI. However, when isoflurane is added, a dose-dependent decrease in SVCI and SVRI responses to lung inflation occur without any diminution in the elevation of CVP. Therefore, even though the change in CVP may influence the effect of the lung reflex on SVCI and SVRI, it is clear from our data that isoflurane attenuates the reflex response. Although large lung inflations may also shift blood from the pulmonary to systemic circulation, in this preparation, this volume of blood would be small because the pulmonary bed was not perfused and thus would have minimal, if any, influence on SVCI change.

Large lung inflations can change cardiac filling pressure, resulting in alteration in cardiac receptor stimulation mediated through vagal afferents. The decrease in blood flow to the left atrium\textsuperscript{30,31} and ventricle\textsuperscript{32} could decrease receptor stimulation, whereas an increase in right heart pressure\textsuperscript{30,31,33} can increase cardiac receptor stimulation. This contribution is insignificant in our preparation because hilar denervation of vagal afferents eliminated the majority of the reflex response to lung inflation.\textsuperscript{15}

Activation of carotid sinus baroreceptor reflex could
alter the SVCI and SVRI changes seen with lung inflation. This interaction was removed by denervating one carotid sinus and producing a steady state pressure in the opposite carotid sinus with a regulated perfusion pump. The increased intrathoracic pressure from a large lung inflation could cause a decrease in aortic transmural pressure and an unloading of aortic receptors. This possible interaction was eliminated by transection of the aortic depressor nerve at the level of the nodose ganglia. Also, any potential influence by the carotid body chemoreceptors was eliminated by mechanical and chemical denervation.

A subthreshold carotid sinus perfusion pressure was selected because it was found that the responses of SVCI and SVRI to lung inflation were augmented at low carotid sinus pressure and attenuated at high carotid sinus pressure. Sustained lung inflation could induce venous congestion and stimulate abdominal low-pressure baroreceptors, causing a reflex sympathetic activation. This sympathetic excitation was shown not to occur when arterial or cardiac receptors were selectively denervated. Therefore, in this preparation, abdominal venous baroreceptor activity was not an important factor.

The specific receptor mechanisms responsible for the change in SVRI and SVCI from sustained lung inflation from positive pressure ventilation is unclear. At low levels of static lung inflation, the reflex response is probably mediated by pulmonary A-fibers with a threshold positive airway pressure for activation of approximately 5 cmH2O. Larger airway volumes with higher airway pressures can stimulate other pulmonary afferents, such as pulmonary stretch receptors and pulmonary C-fibers. The clinical impact of the lung inflation reflex would be difficult to determine because other neural, humoral and myogenic reflexes remain intact and can influence systemic circulation. It can be hypothesized that an increase in SVCI and decrease in SVRI is one mechanism to help maintain adequate splanchnic blood volume during periods of increased respiratory efforts, which stimulate the release of catecholamines and other humoral amines that can compromise tissue perfusion. We can only speculate on the mechanism of the effect of isoflurane on inhibition of the cardiovascular reflex responses to lung inflation. It is likely that this inhibition occurs at multiple sites, as has been shown for isoflurane with regard to inhibition of cardiovascular responses to other reflexes. It has been established that isoflurane may inhibit ganglionic transmission and neurally mediated changes in vascular resistance and capacitance and may have direct inhibitory actions on the heart and vascular smooth muscle.

In summary, sustained lung inflation reflexly increases SVCI and decreases SVRI. Isoflurane inhibits this reflex in a dose-dependent manner. The specific receptor or afferent pathway affected remains to be elucidated.

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