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The Inhibitory Action of Halothane on Reflex Constriction in Mesenteric Capacitance Veins

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Potent inhalational anesthetics depress autonomic reflex responses at multiple sites. Most studies emphasize cardiac chronotropic changes and changes in systemic blood pressure. Recently, active reflex venoconstriction of 500–1,000 μm O.D. mesenteric veins has been demonstrated. In the current study, the effects of halothane on the reflex responses of similar mesenteric veins were measured. Mesenteric vein diameter and intravenous pressure were measured in 500–1,000 μm O.D. veins from the mesentery of segments of terminal ileum externalized *in situ* from 27 New Zealand white rabbits anesthetized with alpha-chloralose. Mean arterial pressure was measured with femoral arterial cannulation, and heart rate was determined from the arterial pressure signal. In a separate group of six animals, sympathetic efferent nerve activity was measured from a postganglionic splanchnic nerve. Reflex venoconstriction and increases in mean arterial pressure and mesenteric vein pressure in response to bilateral carotid occlusion were attenuated by 0.5% and 1% inhaled halothane but not by superfusate equilibrated with 3% halothane. Decreases in mesenteric vein diameter and increases in mesenteric vein pressure in response to celiac ganglion stimulation were unaffected by both 0.5% inhaled halothane and superfusate equilibrated with 5% halothane. The bilateral carotid occlusion reflex-mediated increase in sympathetic efferent nerve activity was depressed by both 0.5% and 1% inhaled halothane. The effect of inhaled halothane on prestimulation baseline vein diameter was inconsistent. Superfusate equilibrated with 5% but not 3% halothane caused baseline venodilation. These results suggest a mechanism whereby control of venous tone is inhibited by halothane proximal to the postganglionic neuron. This could involve central or ganglionic inhibition. (Key words: Anesthetics, volatile: halothane. Sympathetic nervous system: baroreflex; reflex venoconstriction; sympathetic efferent nerve activity.)

HALOTHANE has been shown to cause dose-dependent vasodilation of arterioles and venules in various tissues.[1] In addition, halothane inhibits autonomic reflex responses at multiple sites, including baroreceptors, afferent and efferent nerves, and the autonomic ganglia.[2–4] Most studies on the effects of anesthetics on end-organ responses to autonomic stimuli emphasize cardiac chronotropic changes and changes in systemic blood pressure. Muldoon *et al.*[5] and Rorie *et al.*[6] demonstrated an inhibition of norepinephrine release from electrically stimulated *in vitro* dog saphenous vein segments superfused with physiologic salt solution containing halothane. However, few studies have been reported describing effects of anesthetics on sympathetic activation of capacitance veins that are known to regulate preload and cardiac output.[7] Epstein *et al.* provided some information about this question.[8] They demonstrated a halothane-induced attenuation of the increase in caudal central venous pressure evoked by a decrease in carotid sinus pressure in anesthetized dogs. However, changes in venous pressure do not always accurately reflect venous capacitance changes because the former are also a function of alterations in both arterial inflow and resistance to venous outflow.[9] Recently, Ozono *et al.*[10] demonstrated active sympathetic reflex-mediated venoconstriction of mesenteric capacitance vessels by direct measurement of vein diameter and intravenous pressure.

Clinically, patients having halothane anesthesia tend to demonstrate increased tolerance to, and occasionally a requirement for, volume expansion to maintain hemodynamic stability. This suggests a change in venous capacitance. Although the mechanisms responsible for this change have not been defined, our overall hypothesis is that halothane increases capacitance by reducing neurally mediated venous tone. Therefore, the objective of this study was to examine the effects and sites of action of halothane on sympathetically mediated venoconstriction in mesenteric capacitance vessels.

Materials and Methods

Animal Preparation

The animal preparation has been described in detail by Ozono *et al.*[10] Briefly, after approval by the Animal Care Committee, 33 New Zealand white rabbits (1–2 kg) were anesthetized with thiamylal (10–20 mg/kg) through the ear vein. Anesthesia was maintained with an infusion of alpha-chloralose (12.5–37.5 mg/h). Surgical preparation consisted of local anesthesia by subcutaneous infiltration of 1% lidocaine (3–5 ml) in incision sites, followed by tracheostomy, femoral arterial cannulation for arterial

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pressure measurement, femoral venous cannulation for intravenous infusion, and midline laparotomy. In one group of 15 rabbits, both carotid arteries were isolated *in situ* for subsequent occlusions. In a second group of 12 rabbits, bipolar silver stimulating electrodes were sewn to the celiac ganglion after disruption of central input to the plexus. Postganglionic neuron stimulation was confirmed by the administration of the ganglionic blocker hexamethonium at the end of the experiment. In a third group of six rabbits, a postganglionic splanchnic nerve was isolated *in situ* and sectioned distal to the ganglion. Bipolar recording electrodes composed of two single-strand coated stainless steel wires (0.25 mm O.D.) in Silastic® tubing were fixed to the nerve with Wacker silgel (Wacker-Chemie, Munich, Germany).

Blood Vessel Preparation

In the 27 rabbits subjected either to carotid occlusion or celiac ganglion stimulation, a 13-cm loop of terminal ileum was externalized through a laparotomy incision and mounted in a temperature-regulated plastic tissue chamber placed on a movable microscope stage. The ileum and associated mesentery were continuously superfused with physiologic salt solution originally formulated by Bohlen[11] to simulate the environment of the peritoneal cavity. The composition was as follows (millimolar): NaCl 118.4, KCl 5.9, $NaHCO_3$ 25.0, and $CaCl_2$ 3.3. Temperature was maintained at 37–38° C, and *p*H was kept at 7.35–7.45 by continuous slow bubbling with a gas mixture composed of 5% O_2, 5% CO_2, and 90% N_2. To prevent movement artifact, the mesentery was pinned down to a layer of clear Silastic® rubber that coated the chamber floor. Small *in situ* lengths of mesenteric vein (500–1,000 μm O.D.), cleared of excess fat tissue when necessary, were used for all diameter and pressure measurements. During the surgical preparation the animals breathed spontaneously. After the animals were placed onto the stage, ventilation was controlled with a Harvard model 665® (Harvard Apparatus, South Natick, MA) animal respirator. Periodic arterial blood samples were taken to measure blood gases with the use of a Radiometer Copenhagen ABL I® blood gas device. Normocarbia and normal *p*H were maintained by means of ventilator adjustments and 1–2-ml boluses of 1 mEq/ml of $NaHCO_3$. Rectal temperature was measured with a thermistor probe and maintained at 36.5–37.5° C with a heating blanket. After completion of the animal preparation, an equilibration period of approximately 30 min was provided before measurements were made.

Measurements

In each animal, arterial blood pressure was measured directly through the femoral arterial catheter. The interval between sequential systolic arterial pressure signals was measured in conjunction with a timer module, and a digital to analog converter was used to produce a continuous measurement of heart rate. Mesenteric vein diameter was measured continuously by means of an RCA (Lancaster, PA) model TC2011® video camera focused through the side arm of a Modified Reichert Stereo Star Zoom® (Cambridge Instruments, Buffalo, NY) dissecting microscope and an on-line video micrometer system. The video signal was monitored on an RCA TC1910® television monitor. A comparator signal was generated from the video signal, and its duration was determined by an eight-bit timer module with a digital to analog converter to produce an analog signal proportional to the vessel diameter. This has been described by Bell *et al.*[12] Mesenteric intravenous pressure was determined simultaneously with mesenteric vein diameter in the same vessel segment by means of a World Precision Instruments model 900 Servo-null® (World Precision Instruments, New Haven, CT) Pressure Measuring System. Glass micropipettes were sharpened by beveling to a 5–10-μm tip diameter by means of a specially designed pipette sharpener with the use of a diamond particle disk. The pipettes were filled with 2 M NaCl and used as sensing electrodes.[10] Sympathetic efferent nerve activity was recorded from a postganglionic splanchnic nerve with the bipolar electrodes described above. Directly recorded nerve activity was amplified 40,000× and passband filtered between 0.1–2 kHz. A fourth-order Bessel filter and a moving time averager with a digital to analog converter using 200-ms time increments produced an analog output proportional to sympathetic efferent nerve activity.[13] All data were recorded on a Vetter model 820® (A. R. Vetter Co., Rebersburg, PA) digital video cassette recorder and subsequently printed on an Astromed® Model 9500 eight-channel recorder. Halothane concentrations in blood and physiologic salt solution were measured with the use of a Perkin Elmer (Norwalk, CT) model Sigma 3B gas chromatography system.

Experimental Protocols

Bilateral Carotid Occlusion

Three groups were studied. For the first group (n = 9), changes in mesenteric vein diameter, mesenteric intravenous pressure, mean arterial pressure, and heart rate were measured simultaneously in response to bilateral carotid occlusion at inhaled halothane concentrations of 0%, 1%, 0%, and 0.5%, delivered sequentially for 30 min through a North American Draeger vaporizer using O_2 as a carrier gas at flows of 5 l/min. Inhaled halothane in subsequent protocols (described below) was delivered in similar fashion. For the second group (n = 6), mesenteric vein diameter and mesenteric intravenous pressure were measured simultaneously during superfusion of the vessel preparation with physiologic salt solution equilibrated with

0%, 3%, and 0% concentrations of halothane. Throughout the superfusion experiment, ventilation was controlled and the animals breathed room air occasionally supplemented with 1–2 l/min O_2 to keep Pa_{O_2} levels at or slightly above baseline room air Pa_{O_2} levels. Physiologic salt solution equilibrated with 3% halothane was produced by bubbling a gas mixture of 3% halothane vapor in 5% O_2, 5% CO_2, and 90% N_2 through a container of physiologic salt solution at a flow of 2 l/min for 30 min. Pilot data had previously shown that 15 min was sufficient for equilibration at a particular halothane concentration. For the celiac ganglion stimulation protocol described below, physiologic salt solution equilibrated with 5% halothane was produced in similar fashion.

For the third group (n = 6), changes in sympathetic efferent nerve activity were recorded from a postganglionic splanchnic nerve in response to bilateral carotid occlusion at inhaled halothane concentrations of 0%, 1%, 0%, and 0.5% delivered sequentially.

Celiac Ganglion Stimulation

Two groups were studied. For the first group (n = 6), simultaneous measurements were made of mesenteric vein diameter, and mesenteric intravenous pressure in response to sequential 30-s celiac ganglion stimulations at 5, 10, and 20 Hz (1-ms duration, 3–8-mA current) during sequentially inhaled halothane concentrations of 0%, 0.5%, and 0% for 30 min. For the second group (n = 6), similar measurements were made sequentially during superfusion of the vessel preparation with physiologic salt solution equilibrated with 0%, 5%, and 0% halothane. Again the animals breathed room air with occasional O_2 supplementation. The celiac ganglion stimulation parameters described above were similar to those used by Ozono *et al.*,[10] and are within the maximal physiologic range described by Nishi.[14]

Both 0.5 and 1% inhaled halothane (for 30 min) significantly attenuated the observed reflex venoconstriction in response to bilateral carotid occlusion (see results). In view of the significant attenuation resulting from 0.5% inhaled halothane, this dose was also used in the celiac ganglion stimulation protocol. However, no such attenuation resulted and no other inhaled halothane doses were tested. In the second portion of the bilateral carotid occlusion protocol (*i.e.*, when halothane was administered as a superfusate), 3% halothane vapor was equilibrated with physiologic salt solution for 30 min to produce a proper superfusate concentration. The resulting mean superfusate halothane concentration, which was 0.91 ± 0.06 mM, was chosen because it closely approximated the blood halothane concentration resulting from inhalation of 1% halothane for 30 min (0.88 ± 0.03 mM, see table 1). Pilot studies had indicated that superfusate equilibrated with 3% halothane had no effect on the venoconstrictor response to celiac ganglion stimulation. Diameter reductions resulting from this stimulus are two to five times greater than those resulting from bilateral carotid occlusion. Therefore, to examine the possibility that venoconstriction resulting from celiac ganglion stimulation would be affected by large doses of halothane, a concentration of 1.97 ± 0.11 mM was used when halothane was administered as a superfusate. This was produced by the equilibration of 5% halothane with the physiologic salt solution.

TABLE 1. Concentrations of Halothane in Blood and PSS

Vapor Concentration	Concentration in Blood (mM)	Concentration in PSS (mM)
0.5% inhaled	0.51 ± 0.03	—
1% inhaled	0.88 ± 0.03	—
3% dissolved in PSS	0	0.91 ± 0.06
5% dissolved in PSS	0.004 ± 0.004	1.97 ± 0.11

Mean ± SEM. For inhaled halothane protocols, concentrations in blood are reported. For superfused halothane protocols, concentrations in superfusate and in blood (absorbed halothane) are reported. n = 6–9 for each protocol.

Statistics

Percentage changes in mean arterial pressure, heart rate, mesenteric vein diameter, and mesenteric intravenous pressure were calculated for each animal at each halothane dose in the bilateral carotid occlusion protocol. Percentage change in mesenteric vein diameter was calculated for each animal at each halothane dose in the celiac ganglion stimulation protocols. Percentage change in total activity was calculated for each animal in the sympathetic efferent nerve activity protocol. Arc-sine transformations were performed on all percentages to ensure normal distribution of their values, and data were analyzed by multiple analysis of variance for repeated measures with planned comparisons using the Stats Plus® statistical software produced by Stat Soft Corporation (Tulsa, OK) for IBM PC® computers.

Results

Bilateral Carotid Occlusion Studies

During bilateral carotid occlusion before halothane inhalation, mean arterial pressure increased by 44.0 ± 2.60% from a mean initial value of 75.8 ± 1.95 mmHg (fig. 1*A*), and heart rate increased by 3.91 ± 1.01% from a mean initial value of 251 ± 13.4 beats per min (fig. 1*B*). Simultaneously, mesenteric vein diameter decreased by 4.95 ± 0.95% from a mean initial value of 660 ± 39.9 μm (fig. 2*A*), and mesenteric intravenous pressure increased by 13.6 ± 1.77% from a mean initial value of 9.27 ± 0.38 mmHg (fig. 2*B*). All values reported are mean

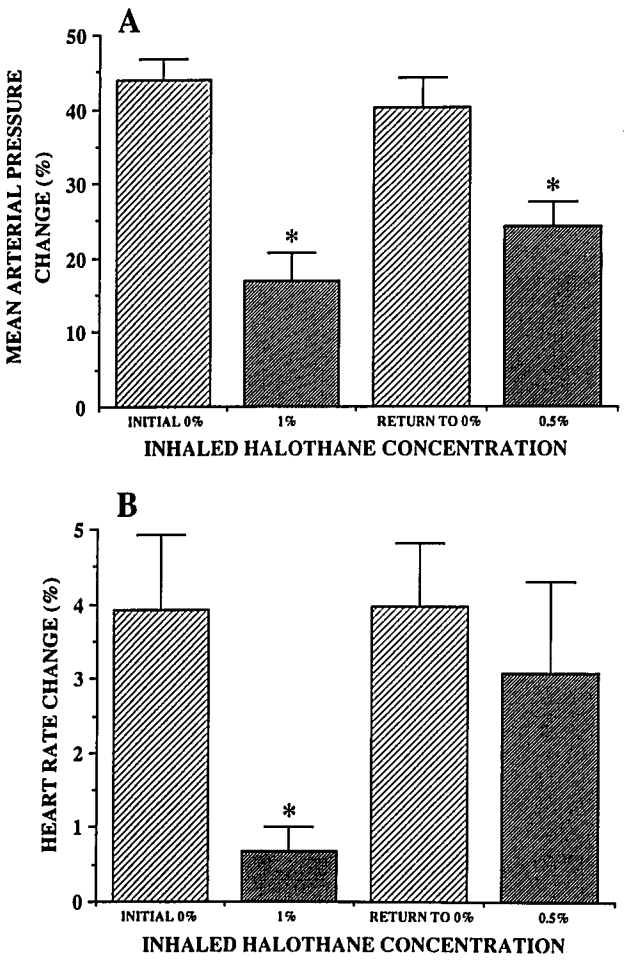

FIG. 1. (*A*) Equal attenuation of mean arterial pressure increase in response to bilateral carotid occlusion by both 1.0 and 0.5% inhaled halothane. (*B*) Attenuation of heart rate increase in response to bilateral carotid occlusion by 1.0 but not 0.5% inhaled halothane. Each column represents mean ± SEM. $*P \leq 0.05$, relative to 0% inhaled halothane controls, n = 6–9.

± standard error of the mean (SEM). Both 0.5% and 1.0% inhaled halothane significantly attenuated the increases in mean arterial pressure, mesenteric intravenous pressure, and mesenteric venoconstriction in response to bilateral carotid occlusion. There were no significant differences between the two halothane concentrations in their attenuation of mean arterial pressure, mesenteric vein diameter, and mesenteric intravenous pressure response to bilateral carotid occlusion. However, 1% but not 0.5% inhaled halothane significantly reduced the heart rate response to bilateral carotid occlusion. Representative traces of these responses are illustrated in Figures 3*A* and *B*. Physiologic salt solution equilibrated with 3% halothane and superfused locally over the mesenteric vein preparation, in contrast to physiologic salt solution without halothane, had no effect on systemic mean arterial pressure or heart rate response to bilateral carotid occlusion. A lack of systemic effect was expected because essentially no halothane was absorbed systemically from the superfusate (see table 1). Three percent halothane in the physiologic salt solution superfusate also had no effect on mesenteric vein diameter or mesenteric intravenous pressure, as is shown in figures 4*A* and *B*, respectively.

Celiac Ganglion Studies

During celiac ganglion stimulation, the magnitude of mesenteric venoconstriction was frequency dependent. Neither 0.5% inhaled halothane (fig. 5*A*) nor physiologic salt solution equilibrated with 5% halothane vapor (fig. 5*B*) had any significant effect on mesenteric venoconstriction in response to stimulation of the celiac ganglion at frequencies of 5, 10, or 20 Hz. Pilot data in the celiac ganglion stimulation protocol indicated that systemically administered hexamethonium, 10 mg/kg, did not block

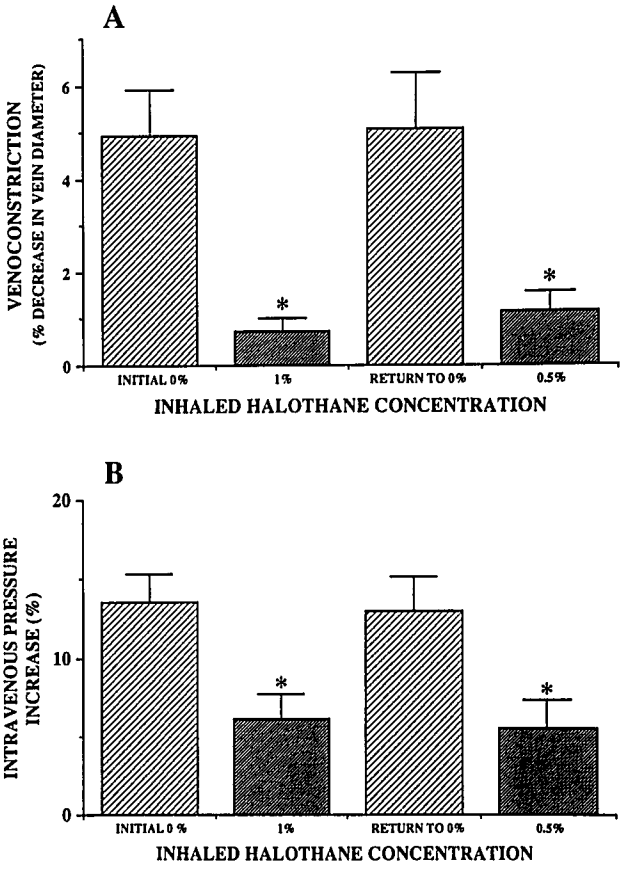

FIG. 2. (*A*) Equal attenuation of mesenteric venoconstriction response to bilateral carotid occlusion by both 1.0 and 0.5% inhaled halothane. (*B*) Equal attenuation of mesenteric intravenous pressure increase by both 1.0 and 0.5% inhaled halothane. Columns, asterisks, and number of vessel preparations as in figure 1.

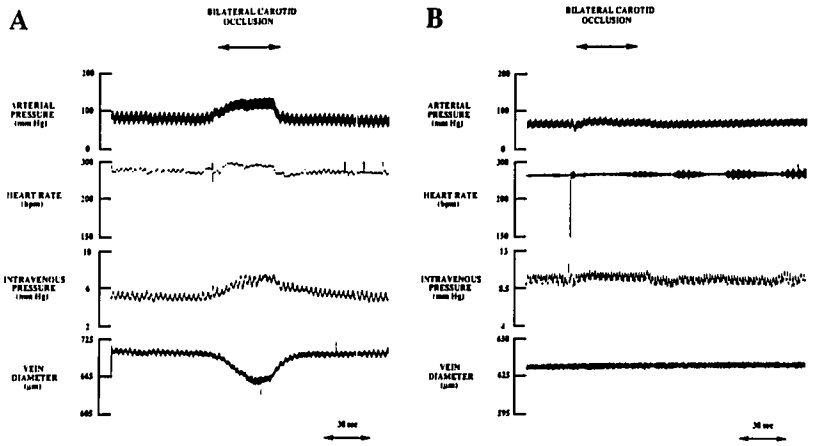

FIG. 3. Representative recording of arterial pressure, heart rate, venous diameter, and intravenous pressure responses to bilateral carotid occlusion prior to and during (A) 0% inhaled halothane and (B) 1.0% inhaled halothane. Note: Recordings were similar for the 1.0 and 0.5% (not shown) inhaled halothane.

venoconstriction in response to celiac ganglion stimulation. This verified that delivery of the ganglionic stimulation was to the postganglionic neuron.

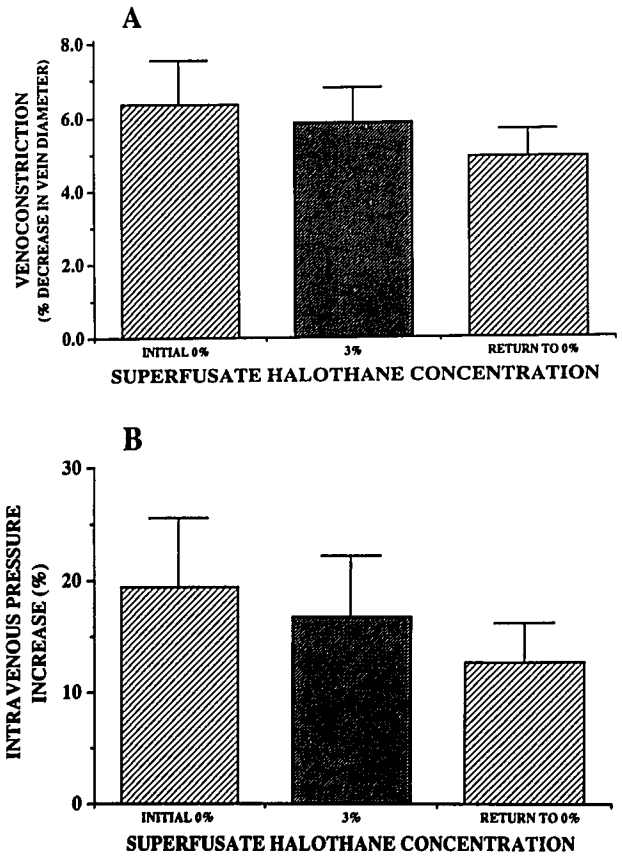

FIG. 4. (*A*) Lack of attenuation of mesenteric venoconstriction in response to bilateral carotid occlusion by physiologic salt solution superfusate equilibrated with 3% halothane. (*B*) Lack of attenuation of mesenteric intravenous pressure increase in response to bilateral carotid occlusion by physiologic salt solution superfusate equilibrated with 3% halothane. Columns, asterisks, and number of vessel preparations as in figure 1.

SYMPATHETIC EFFERENT NERVE ACTIVITY STUDIES

Both 0.5% and 1% inhaled halothane equally reduced the baseline sympathetic efferent nerve activity below that of the 0% halothane control (fig. 6). These concentrations also equally attenuated the respective sympathetic efferent nerve activity increases in response to bilateral carotid occlusion.

EFFECT OF HALOTHANE ON PRESTIMULATION BASELINE DIAMETER

The effects of halothane alone on baseline mesenteric vein diameters (*i.e.*, before maneuvers to produce sympathetic activation such as bilateral carotid occlusion and celiac ganglion stimulation) were not definite. Neither 0.5% nor 1.0% inhaled halothane had any effect on baseline mesenteric vein diameter in the bilateral carotid occlusion study (fig. 7*A*). On the other hand, 0.5% inhaled halothane in the celiac ganglion stimulation study did cause a small but statistically significant venodilation (fig. 7*B*). Physiologic salt solution superfusate equilibrated with 3% halothane in the bilateral carotid occlusion study did produce a tendency toward venodilation (fig. 7*C*), but the difference was statistically significant only when compared with the washout diameters and not with the initial diameters. Physiologic salt solution superfusate equilibrated with 5% halothane in the celiac ganglion stimulation study produced a statistically significant venodilation when compared with both initial and washout diameters (fig. 7*D*).

HALOTHANE CONCENTRATIONS

Mean halothane concentrations ± SEM for each protocol are tabulated in table 1. Units are millimolar in blood when halothane was administered as an inhaled vapor, and when the halothane was administered in the superfusate the units are millimolar in both physiologic salt

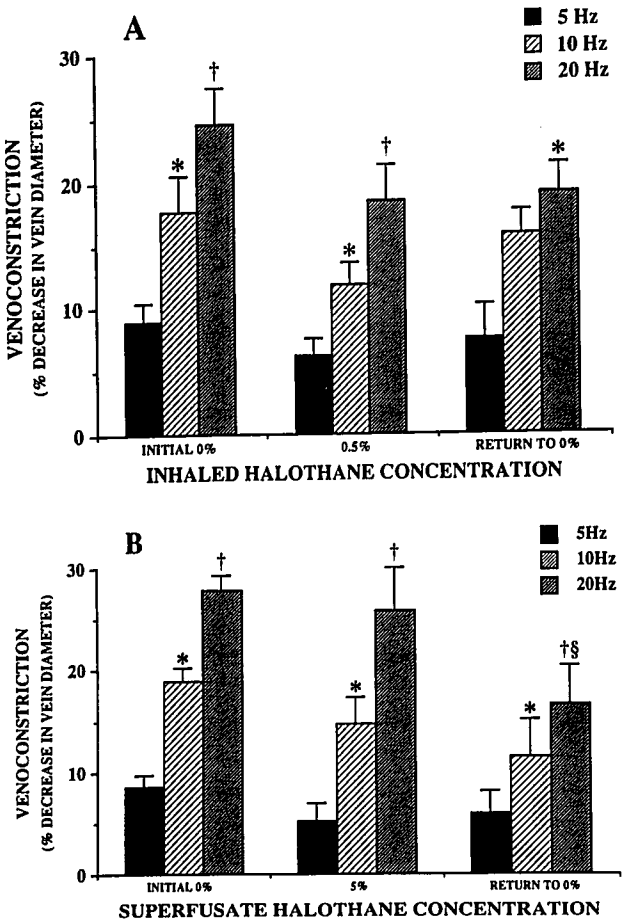

FIG. 5. (*A*) Lack of attenuation by 0.5% inhaled halothane on the step increases in mesenteric venoconstriction due to frequency step increases in celiac ganglion stimulation. (*B*) Lack of effect of physiologic salt solution superfusate equilibrated with 5% halothane on increasing mesenteric venoconstriction in response to frequency step increases in celiac ganglion stimulation. Columns as in figure 1. (*10 or 20 Hz > 5 Hz; †20 Hz > 10 Hz; §20 Hz of return to 0% < 20 Hz of initial 0%; n = 6 vessels, one per animal).

solution and blood. Concentrations in blood during the halothane physiologic salt solution superfusion protocols are very nearly 0 and reflect little or no systemic uptake of halothane from the superfusate.

Discussion

We have shown that active reflex mesenteric venoconstriction in response to decreasing carotid sinus pressure was equally reduced by blood halothane concentrations resulting from 0.5% and 1.0% inhaled halothane vapor. Equivalent or greater physiologic salt solution superfusate concentrations had no effect on this response. Direct celiac postganglionic activation of these mesenteric veins was unaffected by blood concentrations resulting from inhaled halothane vapor sufficient to block reflex responses. Equal or greater superfusate concentrations of halothane also had no effect on such direct postganglionic activation. Inhaled and consequent blood halothane concentrations that inhibited baroreflex-mediated venoconstriction also inhibited reflex-induced increases in postganglionic sympathetic efferent nerve activity.

A possibility exists in the current study for a time-dependent attenuation in mean venoconstrictor response to bilateral carotid occlusion during sequentially administered superfusate equilibrated with 0%, 3%, and return to 0% halothane (fig. 4). A similar possibility exists for the mean venoconstrictor responses to celiac ganglion stimulation before, during, and after inhaled and after superfused halothane (fig. 5). Although a trend seems to exist for such a time-dependent reduction, its statistical significance could not be established. Furthermore, such an effect, if real, would appear to exaggerate any halothane-mediated reduction in venoconstriction. Because superfused halothane did not significantly reduce the venoconstrictor response to bilateral carotid occlusion and neither superfused nor inhaled halothane significantly reduced the venoconstrictor response to celiac ganglion stimulation, such a time-dependent attenuation would not alter our interpretation of these results. All of the attenuated reflex responses described above were inhibited by halothane administration that was superimposed on a basal level of alpha-chloralose anesthesia. This was obviously necessary so that surgical anesthesia could be provided under control conditions. Alpha-chloralose is recognized

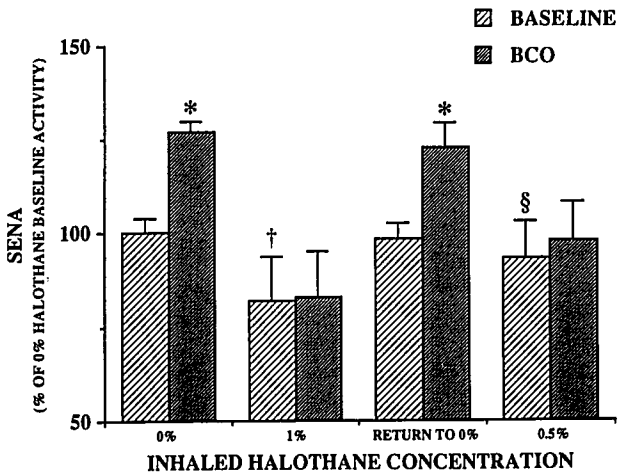

FIG. 6. Equal attenuation by 1.0 and 0.5% inhaled halothane of both prestimulation baseline sympathetic efferent nerve activity and its response to bilateral carotid occlusion. Columns as in figure 1. (*Greater than baseline; †less than 0% pre-bilateral carotid occlusion baseline; §less than pre-bilateral carotid occlusion baseline in 0% and in return to 0%; n = 6 rabbits).

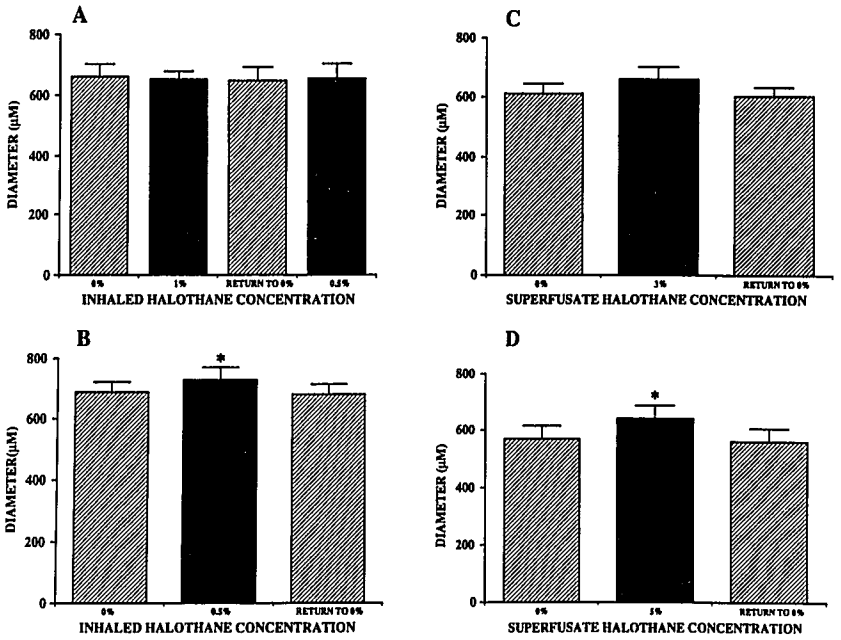

FIG. 7. Prestimulation baseline mesenteric vein diameter response to halothane. (*A*) Lack of effect of 1.0% and succeeding 0.5% inhaled halothane used in bilateral carotid occlusion protocol. (*B*) Small but significant venodilation by the 0.5% inhaled halothane used in celiac ganglion stimulation protocol. (*C*) Tendency for venodilation by the 3% halothane physiologic salt solution superfusate used in the bilateral carotid occlusion protocol. (*D*) Small but significant venodilation by the 5% halothane physiologic salt solution superfusate used in the celiac ganglion stimulation protocol. Columns, asterisks, and number of vessel preparations as in figure 1.

to provide such a level of anesthesia while having minimal effects on cardiovascular reflex responses, in particular, baroreflexes.[15,16] Therefore, the halothane-mediated inhibition of reflex responses in the current study is believed to be independent of any effect of the baseline anesthesia.

The venoconstriction observed in these studies may be the net effect of both active and passive forces acting on the mesenteric veins. The two factors that have been proposed to influence mesenteric vein diameter by passive effects are as follows: 1) the arteriolar inflow of blood into the mesenteric bed; and 2) the venous outflow from the bed, both of which are dependent on the intraluminal pressure and tone of vessels leading to and from the bed. Reduction of venous diameter by active venoconstriction would result from direct neural stimulation of vascular smooth muscle through innervation of the veins.[9,17] Although the exact contribution of each factor is not known for certain, in general the passive influences are known to be opposite in effect. The following evidence supports active venoconstriction. First, second-order mesenteric veins similar in location to those used in these studies, but in the rat, have been shown to be innervated.[18] Postganglionic sympathetic efferent nerve activity measured in the mesenteric bed increases during baroreflex activation. Second, in the current study mesenteric venoconstriction was accompanied by a simultaneous increase in intravenous pressure that can be explained only by active constriction. Third, reflexly mediated mesenteric venoconstriction similar to venoconstriction observed in this study has been shown to be abolished by locally applied tetrodotoxin, a specific blocker of neural activity.[10] Thus, the mesenteric venoconstriction in these studies is most likely the result of neurally mediated active smooth muscle contraction.

The active venoconstriction in response to bilateral carotid occlusion observed in the current study was clearly inhibited by halothane. Hemodynamic depression resulting from halothane administration has been extensively investigated, and many studies report a halothane-mediated arterial vasodilation with consequent reduction in total peripheral resistance[19] as well as myocardial depression. The former may be the result of the direct action of halothane on vascular smooth muscle or halothane-mediated inhibition of the neural control over vascular tone.[19] The results of the current study are consistent with the second of these two possible mechanisms. Although systemically administered (inhaled) halothane inhibited venoconstriction in response to bilateral carotid occlusion, no such inhibition occurred when halothane was locally administered directly to mesenteric veins through the physiologic salt solution superfusate. Venoconstriction resulting from stimulation of the postganglionic neuron at the level of the celiac ganglion was unaffected by either systemically or locally administered halothane. The fact that the reflex response is blocked only by systemically administered halothane, whereas the response to electrical stimulation at the level of the ganglion is entirely unaffected by halothane, strongly suggests an inhibition of neurally mediated control of venous tone proximal to the postganglionic neuron.

The mechanism of attenuation may involve central or ganglionic inhibition and alterations in baroreceptor organ activity and neurotransmission centrally.[19] Numerous lines of evidence support this view. Price *et al.* demonstrated central inhibition of baroreflexes by confining halothane administration to the cephalic circulation in the dog.[20] Subsequently, Price *et al.* again demonstrated central inhibition of reflex changes in heart rate and blood pressure by injecting solutions containing halothane directly into the medullary pressor and depressor areas in dogs.[21] Inhibition of ganglionic transmission has also been demonstrated[2,3,22] and appears to involve a reduction in neurotransmitter release from the presynaptic terminal.[2,23] In halothane-treated isolated ring preparations taken from vessels identical to those in the current study, endogenous neurotransmitter release was shown to be decreased but response to exogenous neurotransmitter was preserved.[24] In addition to the central and ganglionic effects described above, halothane has also been shown to inhibit neurotransmitter release from postganglionic sympathetic neurons in dog saphenous vein strips *in vitro*.[5] Based on results with field stimulation, Rorie *et al.* suggested that the mechanism of this inhibition may involve halothane-mediated activation of prejunctional muscarinic receptors that, on activation, reduce the norepinephrine release from the postganglionic nerve terminal.[6] However, such a proposed mechanism is not supported by the results of our study, because both inhaled and superfused halothane failed to inhibit mesenteric venoconstriction in response to celiac ganglion stimulation in intact animals. Nevertheless, because celiac ganglion stimulation is normally much more effective than field stimulation, it is possible that the celiac ganglion stimulations in the current study were suprathreshold even at the lowest (5 Hz) frequency. If so, this would produce a neural activation that would overshadow halothane-mediated inhibition acting at the postganglionic nerve ending site. Although halothane-mediated inhibition of neurovascular control may occur at any or all of these levels, as well as by baroreceptor sensitization,[3] central and ganglionic inhibition in the intact animal would be most consistent with our results. Additional support from our study for a mechanism involving central inhibition by halothane is the observed reduction in baseline mesenteric sympathetic efferent nerve activity and an inhibition of reflex-mediated increase in sympathetic efferent nerve activity during bilateral carotid occlusion when halothane was administered systemically. These reductions were correlated with the inhibition of reflex-mediated mesenteric venoconstriction that we observed when inhaled halothane was administered in the same sequence.

The decrease in baseline and reflex sympathetic efferent nerve activity that we observed in the current study is in contrast with the data of Millar and Biscoe, who reported an increase in both preganglionic and postganglionic sympathetic efferent nerve activity during inhaled halothane administration in the rabbit.[25,26] Our results, however, are in agreement with those from many other previous studies.[3,4,27] Some of the discrepancy between the results of Millar and Biscoe and those in the current study may be explained by the fact that much higher doses of inhaled halothane were administered in the former. Although serum halothane concentrations were not reported, most of the animals were reported to have received 3% inhaled halothane for periods of 6–24 min in one of the two studies,[25] and 8–16 min in the other.[26] By design, mean arterial blood pressure decreased from 30% to 60%, reaching values of 20 mmHg in some cases. Possibly at such low blood pressures and presumably low levels of cerebral perfusion there is some protective sympathetic activation that overshadows halothane-mediated central inhibition.

Also in contrast to the data supporting a central inhibition by halothane is the work by Epstein and Wang *et al.*[8,28] In one study[28] they used cross-perfusion techniques in the dog and reported decreases in baseline arterial blood pressure and pressor responses in the recipient body when halothane was administered to the body alone (which was vascularly separate from the head). However, these decreases were not seen when halothane was administered to the head alone. In a second study,[8] surgical ligation and cross clamps were used to separate the cephalic circulation from the caudal circulation in the dog by major vessel occlusion. When systemic halothane was administered and then the two circulations were separated, both cephalic and caudal arterial pressor responses and caudal central venous pressor responses to bilateral carotid occlusion were diminished. However, when halothane was administered only to the cephalic circulation, only attenuation of the cephalic pressor response was observed. Their conclusion from both of these studies was that the circulatory depression produced by inhaled halothane was the result of an action peripheral to the central nervous system.[8] Nevertheless, in some of the cross-circulation experiments, these authors did observe a decrease in blood pressure in the body when halothane was being delivered to the isolated head. It is possible that this pressure decrease may have partially resulted from some central inhibition. Also, in the major vessel occlusion experiments,[8] a 0.4–1% inhaled halothane concentration was administered for 10–20 min when it was delivered to the entire circulation, but a 3–10% inhaled halothane concentration was administered for 1–2 min when it was delivered to the cephalic circulation alone. The apparent lack of central effects of halothane may have been related to these sharply different dosing schemes. Regardless of

the extent of central inhibition or direct peripheral vasodilation that existed in these studies, the results can also be explained on the basis of ganglionic inhibition, which has been discussed previously.

In the current study, inhaled halothane had no effect in the bilateral carotid occlusion group and produced dilation in the celiac ganglion stimulation group. Locally applied (superfused) halothane caused significant venodilation when 5% was given, but the observed dilation was not significant when 3% was given (see results, and halothane concentrations in blood and physiologic salt solution in table 1). This inconsistency is in contrast to other studies in the literature that have demonstrated halothane-mediated vasodilation in the resting state, as a result of withdrawal of neural regulatory tone or direct smooth muscle relaxation.[1,19] The reason for this discrepancy may well be that the current study was designed to measure the effect of halothane on *responses* to neural activation rather than on control baseline venous diameters. As such, it is entirely possible that the baseline diameters or their changes in response to halothane may have been altered by one or more of the neurally mediated constrictions before halothane was administered. The halothane-mediated baseline venodilation that was observed in the current study corresponds with the baseline reductions in sympathetic efferent nerve activity that were also observed (see fig. 5). Nevertheless, these baseline changes could also be the result of a direct inhibition by halothane of smooth muscle tone, as suggested above.

In summary, many data exist to support multiple sites of action of halothane to explain its well-established hemodynamic depression. In addition, the role of neural regulation of venous tone in maintaining hemodynamic stability has recently been reviewed by Rothe.[7,29] Clinically, hypotension may develop in patients maintained on cardiopulmonary bypass (and therefore with a fixed cardiac output) during increased halothane administration, which results from a decrease in venous return caused by an increase in venous capacitance.[29] However, the mechanism and relative importance of such a change have not been substantiated. Our findings indicate that inhaled halothane inhibits reflex responsiveness of mesenteric capacitance veins. In addition, they suggest a mechanism of action whereby control of venous tone is inhibited by halothane proximal to the postganglionic neuron. This could involve central or ganglionic inhibition. We observed some evidence for a direct venodilatory action of halothane, but only when it was administered locally to the venous preparation in higher concentrations. Although there is evidence to show an inhibitory effect of halothane on vascular smooth muscle,[30,31] the contribution of this mechanism to observed changes in mesenteric venous capacitance is less well established. Additional studies of the direct effects of halothane on venous smooth muscle, as well as the reflex and direct actions of the other potent inhalational anesthetics, and the responses in various other venous beds are indicated.

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