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The Effects of Halothane on Cardiovascular Responses in the Neuraxis of Cats

Influence of Background Anesthetic State

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Background: **This study examined the effects of halothane on arterial pressure after central nervous system (CNS) pressor site stimulation in anesthetized cats, cats rendered unconscious by midcollicular transection, and conscious cats.**

Methods: **Two anesthetized groups and two nonanesthetized groups were used. Cats were anesthetized with either α-chloralose and urethane or pentobarbital. Nonanesthetized groups were cats with midcollicular transections or conscious cats with chronically implanted electrodes. Stimulating electrodes were placed into vasomotor areas of the hypothalamus (HYP), reticular formation (RF), and medulla, and arterial pressure responses to increasing stimulus currents were examined during different halothane concentrations. Two groups of cats were also anesthetized with either pentobarbital or urethane and underwent bilateral carotid artery occlusion.**

Results: **Stimulation at each CNS site produced increases in arterial pressure and heart rate. Halothane attenuated pressor responses evoked by stimulation of all loci in all groups of cats. The inhibition by halothane on these cardiovascular responses was greatest at HYP and RF sites, while the medulla was more resistant to the effects of halothane in the anesthetized animals. Midcollicular transection decreased this medullary resistance. The inhibition of pressor responses by halothane was also greater in pentobarbital- than chloralose urethane-anesthetized animals. In contrast, pressor responses elicited by bilateral carotid occlusion were attenuated by halothane similarly in both anesthetic groups. Reticular formation stimulation in conscious animals resulted in "alerting responses" in addition to pressor effects, both of which were attenuated by halothane.**

Conclusions: **Modulation of CNS cardiovascular control centers contribute to halothane-induced hemodynamic alterations. Baseline anesthesia, CNS stimulation site, and the suprabulbar system influence the effects of halothane. (Key words: Anesthetics, hypnotics: α-chloralose; pentobarbital; urethane. Anesthetics, volatile: halothane. Brain, hypothalamus, reticular formation, medulla: baroreflex; cardiovascular regulation; pressor sites.)**

THE volatile anesthetics, including halothane, have been previously demonstrated to disrupt normal cardiovascular function. Halothane has been shown to alter cardiovascular stability through multiple actions, including a direct negative inotropic and chronotropic action on the heart,[1] a direct vascular smooth muscle vasodilatation,[2] a ganglionic blocking action,[3,4] an alteration of sympathetic and parasympathetic tone to the heart,[5] and effects within the central nervous system (CNS).[6–10] Halothane has been postulated to alter CNS modulation of circulatory control by a variety of mechanisms, including a disruption of baroreflex function,[9–12] increase or decrease in vagal efferent activity,[5,6,13] and an alteration of central integration of afferent neuronal input and the generation of appropriate efferent responses.[7,8]

The action of halothane in altering CNS control of cardiovascular function has been unclear. Price[7] demonstrated that halothane attenuated both pressor and depressor responses to medullary stimulation in va-

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gotomized, decerebrate dogs. In contrast, using a variant of the isolated supported head technique, Wang *et al.* examined the effect of halothane on medullary pressor responses in pentobarbital anesthetized dogs and showed little or no reduction of reflex vascular responses to medullary stimulation when halothane was delivered to the cephalic perfusion.[8] In that study, there were marked alterations in responses to medullary stimulation when halothane was delivered caudally to the body only, indicating a peripheral, rather than central, action of halothane in modulating circulatory control.

A variety of vasomotor control areas exist within the CNS, including the hypothalamus, brainstem reticular formation, and medulla,[14–23] and their roles in regulating circulatory function have been relatively well delineated. However, few studies have examined the effects of the volatile anesthetics, including halothane, on these central vasomotor centers. Recently, Poterack *et al.*[23] examined the effects of isoflurane, midazolam, and etomidate on cardiovascular responses to stimulation of CNS pressor sites in chronically instrumented cats.[23] All three agents significantly attenuated the increase in arterial pressure after stimulation of hypothalamic or reticular formation vasomotor centers. In contrast to the work of Wang *et al.*,[8] these results indicate that these anesthetic agents may alter hemodynamic stability, at least in part, by disruption of cardiovascular control centers within the CNS.

Because the majority of investigations examining anesthetic actions on CNS function have been in acute studies in animals that had received a hypnotic agent (baseline anesthetic) and because all anesthetics disrupt ongoing cardiovascular function to some extent, the purpose of the first part of the current study was to use different hypnotic agents in analyzing the actions of halothane on CNS modulation of cardiovascular function. A separate group of cats with midcollicular transections was also used in the current study to eliminate disruptive influences of hypnotic agents. However, midcollicular transections produce a degree of CNS cardiovascular imbalance and, thus, an additional group of conscious, chronically instrumented cats was also used. Three previously well-delineated central vasomotor areas were chosen for electrical stimulation sites: the ventrolateral hypothalamus, the mesencephalic reticular formation, and the medulla. The hypothesis being tested was that the disruptive effects of halothane on CNS control of the cardiovascular system is influenced by the presence of a baseline anesthetic and by an intact suprabulbar region.

Materials and Methods

All experimental procedures were approved by the Medical College of Wisconsin Animal Use and Care Committee, and protocols were completed in accordance with the Guiding Principles in the Care and Use of Laboratory Animals of the American Physiological Society and were in accord with NIH guidelines.‖ All chronically instrumented animals were housed within the animal facilities of the Medical College of Wisconsin, accredited by the American Association for the Accreditation of Laboratory Care. Animals were randomly assigned to treatment groups.

General Preparation

Fifty-five cats were used for the CNS stimulation experiments. Successful experiments were performed in twenty-seven animals, as follows. Seven male and female cats were anesthetized by an intraperitoneal injection of α-chloralose (50–60 mg/kg) and urethane (500–600 mg/kg) and seven cats were given intraperitoneal sodium pentobarbital (30–35 mg/kg). Five cats were instrumented for midcollicular transections and eight chronically instrumented cats were used. The weight range was 1.9–3.6 kg with no significant differences in weights between the groups. All animals fasted overnight before experimentation, and 0.9% saline was used as fluid replacement.

In all animals, the right femoral artery and left cephalic vein were cannulated for measurement of arterial pressure and for fluid and drug administration, respectively. Animals were either tracheally intubated or a 6.5 mm O.D. metal cannula was inserted through a tracheostomy site and the animals' lungs were mechanically ventilated with 100% oxygen, using a ventilator and a semiclosed circuit. Arterial blood samples were determined for measurements of blood gas tensions (Model ABL-2; Radiometer, Copenhagen, Denmark). Ventilation was adjusted to maintain normal arterial p_{CO_2}. During anesthesia, animals were placed in a stereotaxic restraint and the skull was exposed through a midline incision. Burr holes were made through the calvarium with a high-speed, air-turbine

‖ Guide for the Care and Use of Laboratory Animals. DHEW (DHHS) Publication No. (NIH) 85-23, revised 1985.

drill (Midwest Dental products, Des Plaines, IL) and 23-G coaxial stimulating electrodes were stereotaxically placed in target sites in the right ventrolateral hypothalamus (Horsely-Clark, anterior 10 mm, lateral 2.5 mm, depth −3–−5 mm), the left reticular formation (Horsely-Clark, anterior 2 mm, lateral 2 mm, depth 0–−2 mm), and the right medulla (Horsely-Clark, posterior 11 mm, lateral 4.5 mm, depth −4–−6 mm) according to the atlases of Bleier# and Berman.** The electrode was advanced toward the target depth in 0.5-mm increments, with stimulation at each depth, to determine the optimal position for eliciting responses. The coaxial stimulating electrodes were constructed of epoxy insulated 23-G stainless steel tubing with formvar insulated wire cemented in the lumen. Total resistance in saline usually ranged from 60 to 80 kiloohms.

The stimulus for pressor responses was a 10-s train of 0.05–0.1-msec square wave pulses delivered at 100 Hz through a constant current unit using digital stimulators (WPI Electronics, New Haven, CT). Current intensity was monitored and adjusted continuously by means of an optically isolated current monitor. Thus, even if electrode impedance varied during the experiment, current density remained the same as at selected control values.

After determining the optimal depth for eliciting a pressor response, a "threshold" current, which elicited a small but consistent increase in pressure, was established. Threshold and two and four times threshold current was then accomplished at 3–5-min intervals, and at least two, and usually three, series of responses to stimuli were obtained before administration of halothane. Arterial pressure was allowed to return to baseline before repetition of stimuli. Halothane was randomly administered at 0.5, 1, and 2% end-tidal concentrations, as measured by a mass spectrometer (Marquette Electronics, Milwaukee, WI). Studies in our laboratory have shown that the MAC of halothane in the cat is 1.22 ± 0.05. A minimum of 30 min was allowed to elapse between each change in anesthetic concentration and stimulus presentation. Stimuli sequences were randomized and graded pressor responses determined at each anesthetic concentration. On completion of the stimulus sequences during halothane administration, a 30–90-min "washout" period without halothane was allowed for the acute animals and an overnight "washout" was allowed for the chronically instrumented animals, and electrode sites were again stimulated. Core body temperature was monitored and maintained within normal limits during all experiments.

Midcollicular Transection Technique

Anesthesia was induced in five animals *via* mask administration of halothane. After arterial and venous cannulation and tracheostomy were accomplished, animals were placed in a stereotaxic restraint. A midline incision exposed the skull over the occipital and parietal lobes. A craniotomy exposed the cerebrum from the tentorial plate to the squamous suture laterally and to the sagittal suture dorsally. The dura was reflected and, using a blunt spatula, the occipital lobe was retracted and removed, thus exposing the bilateral inferior and superior colliculi. A midcollicular transection was accomplished using a wire loop. Figure 1 demonstrates the location of the transection. Multiple passes rostrally and medially with a small needle electrode with continuous lesioning current destroyed the rostral brain. Anesthesia was then discontinued, positive pressure ventilation was initiated, and pressor responses were elicited by reticular formation and medullary stimulation.

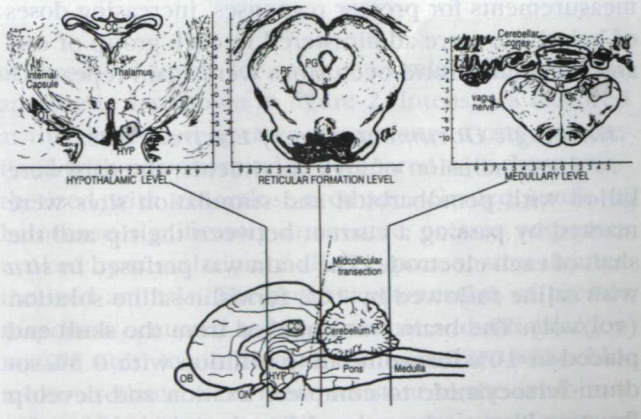

Fig. 1. Schematic representation of CNS sites for stimulating electrodes in sagittal and coronal planes. Diagrams correspond to Horsely-Clark coordinates in frontal plane. See text for specific coordinates of the right ventrolateral hypothalamus (HYP) (N = 22), left reticular formation (RF) (N = 27), and the right medulla (N = 17). Hatched line represents plane for midcollicular transection. CC = corpus callosum; OB = olfactory bulb; ON = optic nerve; OT = optic tract; PG = periaquaductal gray; 5 = trigeminal nerve; SC = superior colliculus; V3 and V4 = 3rd and 4th ventricles respectively.

Bleier R: The Hypothalamus of the Cat. Baltimore, The Johns Hopkins Press, 1961, pp 58–62.

** Berman A: The Brain Stem of the Cat. Madison, University of Wisconsin Press, 1961, pp 14–16, 48–50.

Chronic Implantation Technique

Methodology for chronic implantation technique has been described in detail previously.[23] Cats were anesthetized with halothane and tracheally intubated, and their lungs mechanically ventilated. All surgical sites were prepared and draped in a sterile fashion. The right external jugular vein and right carotid artery were cannulated for fluid and drug administration and pressure measurements, respectively. Catheters were tunneled subdermally to a lateral thoracic exit site. The techniques for electrode placement were similar to those described above, with stimulating electrodes advanced into the right ventrolateral hypothalamus and left reticular formation. The electrodes were anchored to the skull within a miniature headplate and tissue layers were closed. Animals were allowed to recover for a minimum of 7–10 days and were trained to rest quietly in a restraining sling.

Bilateral Carotid Occlusions

In two separate groups of cats, after administration of the baseline anesthetic (either sodium pentobarbital [n = 6] or chloralose-urethane [n = 6], which were assigned in random fashion), the animals underwent arterial and venous cannulation and ventilation of their lungs, as described above. The carotid arteries were isolated and bilateral carotid occlusions were performed for 10 s and then released using a specially constructed hydraulic occluder. After baseline control measurements for pressor responses, increasing doses of halothane were administered in each group of cats and bilateral carotid occlusions were again repeated.

Histologic Documentation of Electrode Sites

At the conclusion of all experiments, the cats were killed with pentobarbital and stimulation sites were marked by passing a current between the tip and the shaft of each electrode. The brain was perfused *in situ* with saline followed by 10% formalin-saline solution (vol/vol). The brain was removed from the skull and placed in 10% formalin-saline solution with 0.5% sodium ferrocyanide to complete fixation and develop Prussian Blue marks at the electrode tip sites. After fixation for 48 h, the brains were cut into blocks, frozen, and sectioned for histologic determination of electrode sites.

Statistical Analysis

Pressor responses were calculated as change in systolic arterial pressure from baseline measurements taken immediately before stimulation of each CNS site. Pressor responses before and during anesthesia were analyzed with a repeated-measures ANOVA with one between factor (treatment groups) and two within factors (dose of halothane and current stimulation) for each CNS stimulation site. All statistical analyses were performed using SAS (Statistical Analysis System) procedure GLM software on a VAX mainframe computer. Multivariate tests (Roy's greatest root) for overall significance (main effects and two-way and three-way interactions) were performed. When the interaction tests were significant, individual comparisons were made to identify which means were different. A Bonferroni multiple comparison procedure was used to make comparisons between treatment groups for a single site, dose and stimulation current intensity, and comparing halothane level back to control at a single site and voltage level. All changes were considered significant when $P < 0.05$. All data are expressed as mean ± SEM.

Results

Histologic Documentation of Electrode Sites

Histologic examination showed that hypothalamic, reticular formation, and medullary sites were located within 1–1.5 mm of the target coordinates in all three axes (fig. 1). Hypothalamic sites were located in the ventrolateral hypothalamus adjacent to the third ventricle, extending caudally to the mamillotegmental tract and rostrally toward the mamillothalamic tract. Reticular formation sites were located slightly lateral to the periaqueductal gray area and included the central tegmental field. Medullary sites were located dorsolaterally, at the level of the fourth ventricle, vagus nerve, and lateral tegmental field, and extended posteriorly toward the nucleus tractus solitarius. Thus, electrode sites for electrical stimulation were within generally accepted boundaries of the target pressor regions.

Alterations in Hemodynamics Produced by Halothane

The effects of graded levels of halothane on systemic hemodynamics in cats anesthetized with pentobarbital or chloralose-urethane, in cats after midcollicular transection, or in chronically instrumented cats are shown in table 1. There were no differences between groups in heart rate at baseline control. Halothane significantly decreased heart rate of animals in all groups and, after its discontinuation, there were no differences in heart

Table 1. Change in Systemic Hemodynamics after Halothane Administration in Cats Anesthetized with Pentobarbital, Chloralose–Urethane, Midcollicularly Transected, or Chronically Instrumented

			Halothane			
	Group	Control	0.5%	1.0%	2.0%	Post
HR (beats/min)	PB	245 ± 8	205 ± 15	189 ± 14*	169 ± 5*	218 ± 14
	CU	240 ± 13	230 ± 14	224 ± 18	205 ± 21*‡	235 ± 14
	MC	197 ± 12	178 ± 14	174 ± 9	164 ± 5*	202 ± 17
	C	207 ± 15	192 ± 21	163 ± 22	117 ± 11*§	204 ± 14
SBP (mmHg)	PB	142 ± 4	110 ± 6*§	93 ± 6*§	71 ± 4*	140 ± 7
	CU	157 ± 11‡	151 ± 11†‡	142 ± 14†‡	96 ± 8*‡¶	152 ± 12
	MC	149 ± 11	120 ± 2*	100 ± 3*	62 ± 1*§	147 ± 9
	C	123 ± 4§	109 ± 9§	92 ± 10*§	58 ± 6*§	134 ± 9
DBP (mmHg)	PB	103 ± 5	77 ± 7*	70 ± 6*	45 ± 4*‡	98 ± 8
	CU	100 ± 13	91 ± 10	84 ± 10	52 ± 5*‡	105 ± 14
	MC	92 ± 4	83 ± 3	68 ± 3*	37 ± 2*	101 ± 5
	C	80 ± 4	72 ± 9	56 ± 9*	27 ± 3*†§	84 ± 6
MAP (mmHg)	PB	117 ± 4	88 ± 6*	78 ± 5*	54 ± 4*	112 ± 7
	CU	119 ± 12	112 ± 7	103 ± 10	67 ± 6*	121 ± 11
	MC	111 ± 5	95 ± 3*	79 ± 3*	45 ± 1*	116 ±5
	C	94 ± 4	85 ± 9	68 ± 9*	37 ± 3*	101 ± 6

PB = pentobarbital (n = 7); CU = chloralose–urethane (n = 7); MC = midcollicular transection (n = 5); C = chronically instrumented (n = 8); HR = heart rate; SBP = systolic blood pressure; DBP = diastolic blood pressure; MAP = mean arterial pressure.

Data are mean ± SEM. Post values represent measurements 24 h after discontinuation of halothane for C animals and 30–90 min for all other groups. Note the similar decreases in heart rate during halothane in all groups. Halothane decreased arterial pressure of animals in all groups with the greatest changes in the MC and C groups. There were significant halothane main effects for each variable and group main effects for SBP.

* Significantly different ($P < 0.05$) from control (conscious for chronically implanted or baseline for anesthetized and transected).

† Significantly different ($P < 0.05$) from pentobarbital (PB).

‡ Significantly different ($P < 0.05$) from chronically instrumented (C).

§ Significantly different ($P < 0.05$) from chloralose–urethane (CU).

¶ Significantly different ($P < 0.05$) from midcollicularly transected (MC).

rates between groups or within any group as compared to prehalothane control values.

Systolic arterial pressures were significantly different at baseline control between chloralose-urethane and chronic groups (157 ± 11, chloralose-urethane group *versus* 123 ± 4 mmHg, chronic group). Diastolic and mean arterial pressures were not different between any groups during prehalothane controls or after discontinuation of halothane. Halothane significantly decreased arterial pressure in all groups.

Central Nervous System Pressor Site Responses in Anesthetized Cats

The results of the pressor responses before, during, and after halothane administration in pentobarbital- and chloralose-urethane-anesthetized animals is shown in figures 2–4. Stimulation of all CNS vasomotor sites significantly increased arterial pressure in a current-dependent fashion in anesthetized cats. Current intensities at threshold levels for each site under each baseline anesthetic are shown in figure 2. Intensities required at threshold were greater at each site in cats anesthetized with pentobarbital as compared with those anesthetized with chloralose-urethane. Graded levels of halothane significantly attenuated pressor responses evoked by stimulation of all CNS pressor sites in anesthetized animals. The greatest attenuation of pressor responses occurred at hypothalamic and reticular formation sites. In addition, cats anesthetized with chloralose-urethane were more resistant to the inhibitory effects of halothane on CNS pressor site stimulation, as compared with cats anesthetized with pentobarbital (at 2% halothane, maximal reticular formation stimulation; 5 ± 2 mmHg pentobarbital group *versus* 33 ± 19 mmHg chloralose-urethane group). Indeed, after medullary stimulation in chloralose-urethane anesthetized cats, the attenuation of pressor responses by halothane are only significant at the threshold stimulation level,

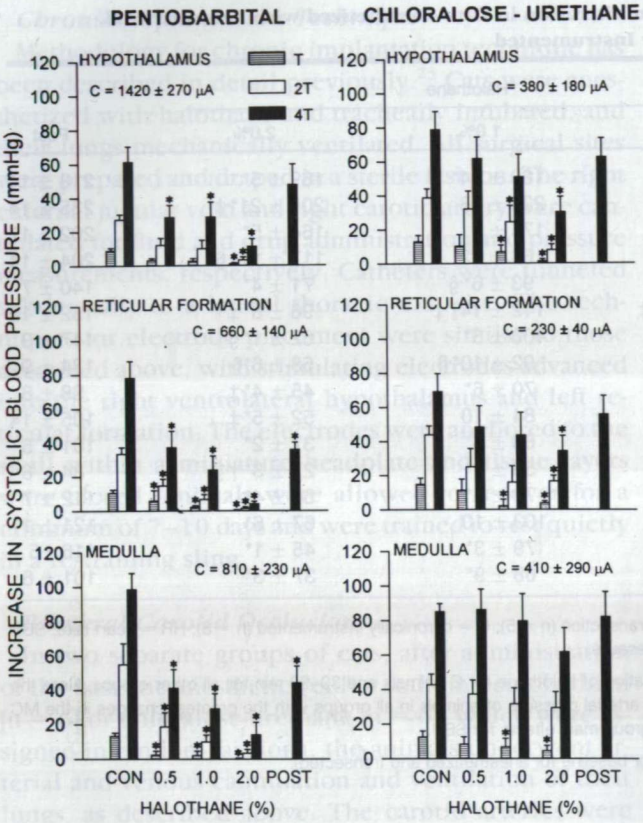

Fig. 2. Effects of graded concentrations of halothane on systolic arterial pressure responses to threshold (T) and two and four times threshold (2T and 4T) stimulation at hypothalamic, reticular formation, and medullary sites. C = Threshold current intensity; CON = control conditions with either pentobarbital (N = 7) or chloralose-urethane (N = 7) as baseline anesthetic. Data are expressed as mean ± SEM. *Significantly different ($P < 0.05$) from control response at the same current intensity. Note the greatest attenuation of pressor responses during halothane administration at hypothalamic and reticular formation sites. Also, cats given chloralose-urethane were more resistant to halothane effects as compared with pentobarbital-anesthetized cats.

even at concentrations of 2% halothane. In contrast, 0.5% halothane significantly attenuates medullary pressor responses in pentobarbital-anesthetized cats at all stimulation intensities. Figure 3 illustrates typical effects of halothane on arterial pressure responses after CNS stimulation in cats anesthetized with pentobarbital.

Central Nervous System Pressor Site Responses in Nonanesthetized Cats

Midcollicular transections resulted in stable hemodynamics that were not significantly different than those of anesthetized or chronic-conscious animals (table 1). Midcollicullar transections also produced significant and stereotypic decerebrate posturing in all animals. Medullary and reticular formation stimulation in midcollicullarly transected cats resulted in significant increases in arterial pressure that were greater than those elicited in cats that were chronically implanted or in anesthetized cats (figs. 4 and 5). Administration of halothane after midcollicular transection resulted in a significant dose-dependent attenuation of pressor responses. The medulla was not as resistant to the effects of halothane depression in the midcollicular animals as in the anesthetized animals. Figure 6 depicts a representative tracing of pressor responses to stimulation of central pressor sites in midcollicularly transected cats after administration of halothane.

Hemodynamic baseline measurements of chronically instrumented cats are shown in table 1. There were no significant differences in the heart rate and diastolic or mean arterial pressure of conscious-intact cats as compared with those of cats in other groups. Systolic arterial pressure was significantly lower in conscious cats than that in chloralose-urethane-anesthetized cats. Stimulation of both the reticular formation and hypothalamus produced significant pressor responses in the conscious state (figs. 4 and 7) and produced what appeared to be an "alerting response," characterized by bilateral pupillary dilatation, mild tachypnea, movement of the head, and behavioral manifestations of increased atten-

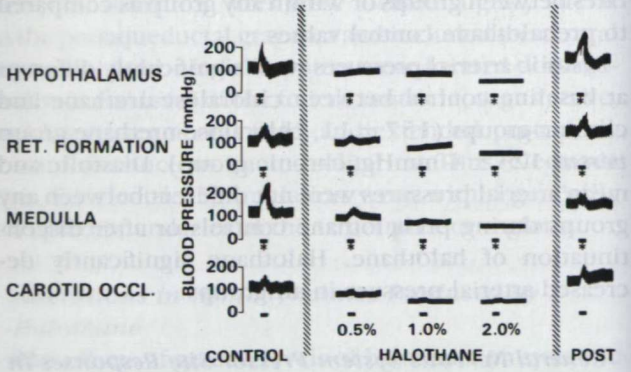

Fig. 3. Representative tracings of a pentobarbital-anesthetized cat showing the effects of graded doses of halothane on systolic arterial pressure responses to four times threshold CNS stimulation or after bilateral carotid occlusion. Note the marked attenuation of arterial pressor responses by halothane. Central nervous system sites were hypothalamus, reticular formation, or medulla.

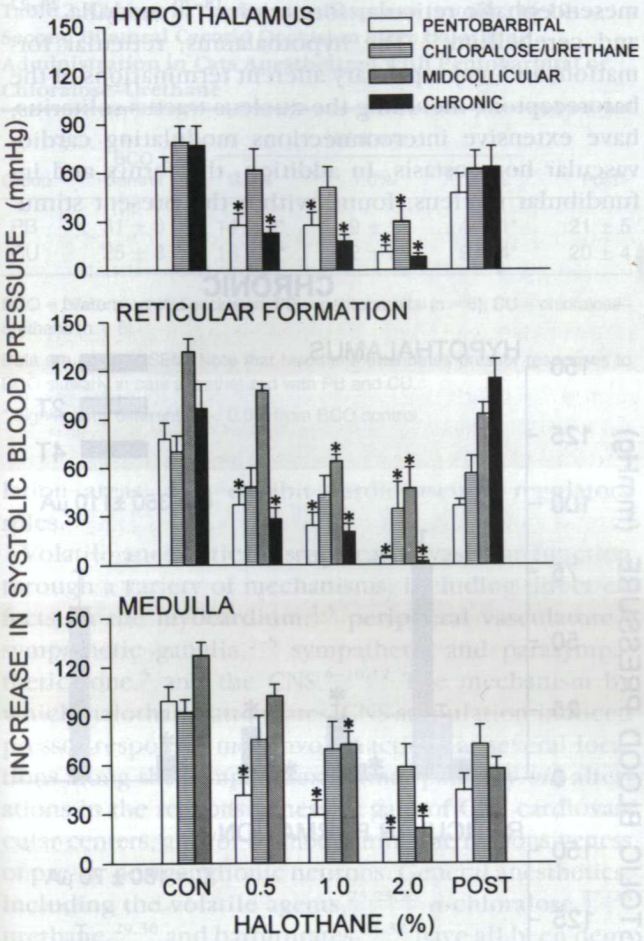

Fig. 4. Effects of graded concentrations of halothane on systolic arterial pressure responses to four times threshold stimulation at hypothalamic, reticular formation, and medullary sites. Treatment groups include: anesthetized with either pentobarbital (N = 7) or chloralose-urethane (N = 7), midcollicular transectioned nonanesthetized (N = 5), and conscious, chronically instrumented cats (N = 8). CON = control-baseline. Data are expressed as mean ± SEM. *Significantly different ($P < 0.05$) from control response at the same current intensity. Note the least attenuation of pressor responses at the medullary site, and that midcollicular transection decreased this medullary resistance. Also, note the similarity in responses between pentobarbital anesthetized and chronically instrumented cats.

tiveness. Administration of halothane attenuated the stimulation-induced pressor responses. Although 0.5% halothane appeared to produce a degree of anesthesia in this animal, stimulation of the reticular formation produced similar manifestations of behavioral attentiveness, as observed in the conscious state. These effects lasted 15–30 s after stimulus presentation. Higher levels (1 and 2%) of halothane diminished these behavioral effects in addition to the marked attenuation of the CNS-induced pressor response.

Pressor Responses to Carotid Occlusion

Bilateral carotid artery occlusion produced marked increases in blood pressure in anesthetized cats. There

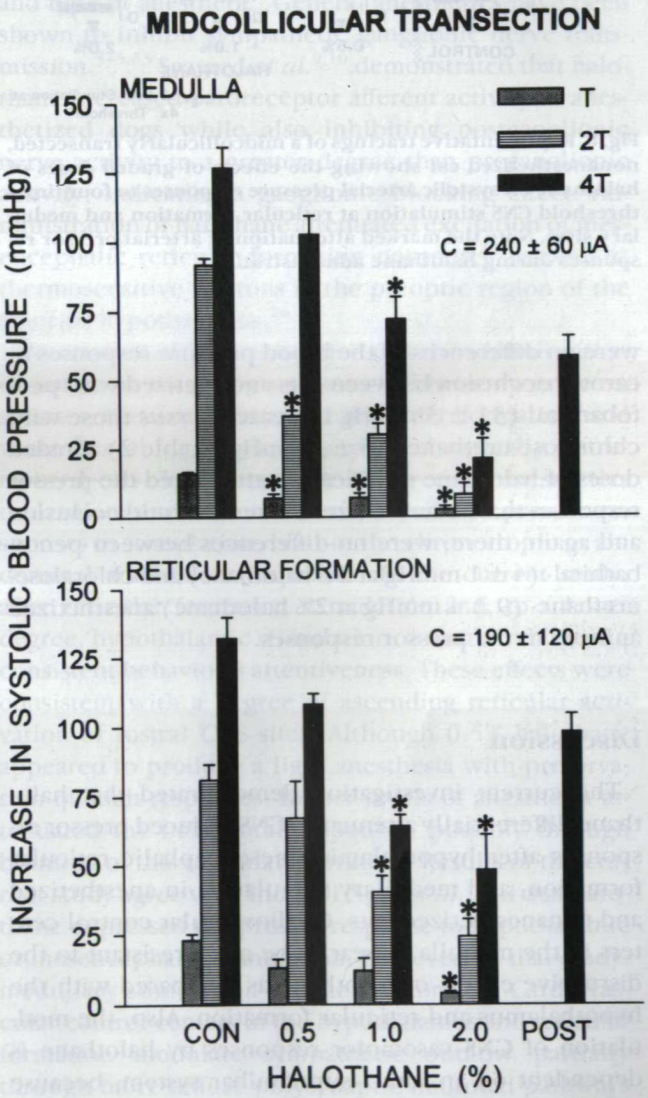

Fig. 5. Effects of graded concentrations of halothane on systolic arterial pressure responses to threshold (T) and two and four times threshold (2T and 4T) stimulation at reticular formation and medullary sites in nonanesthetized cats with midcollicular transections (N = 5). C = Threshold current intensity; CON = control-baseline. Note the increased sensitivity of the medulla to the effects of halothane as compared with the reticular formation. Data are expressed as mean ± SEM. *Significantly different ($P < 0.05$) from control response at the same current intensity.

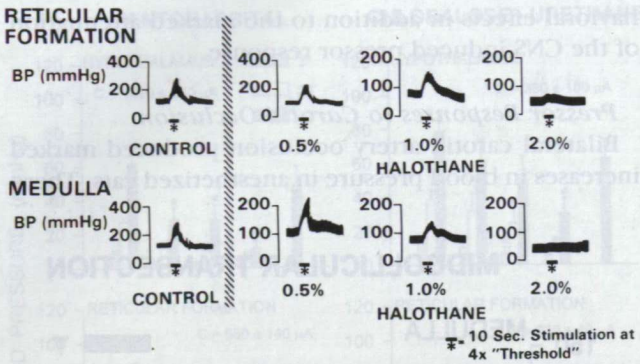

Fig. 6. Representative tracings of a midcollicularly transected, nonanesthetized cat showing the effects of graded doses of halothane on systolic arterial pressure responses to four times threshold CNS stimulation at reticular formation and medullary sites. Note the marked attenuation of arterial pressor responses during halothane administration.

were no differences in the blood pressure responses to carotid occlusion between cats anesthetized with pentobarbital (31 ± 3 mmHg increase) *versus* those with chloralose-urethane (25 ± 3 mmHg) (table 2). Graded doses of halothane significantly attenuated the pressor responses that occurred after bilateral carotid occlusion and again, there, were no differences between pentobarbital- (4 ± 1 mmHg at 2% halothane) and chloralose-urethane- (9 ± 4 mmHg at 2% halothane) anesthetized animals in the pressor responses.

Discussion

The current investigation demonstrated that halothane differentially attenuated CNS-induced pressor responses after hypothalamic, mesencephalic reticular formation, and medullary stimulation in anesthetized and nonanesthetized cats. Cardiovascular control centers in the medulla appear to be more resistant to the disruptive effects of halothane as compared with the hypothalamus and reticular formation. Also, the modulation of CNS vasomotor responses by halothane is dependent on an intact suprabulbar system, because midcollicular transection resulted in greater sensitivity to the effects of halothane at the medullary level. Halothane effects on CNS pressor responses are also influenced by the presence and type of baseline anesthetic.

Control of cardiovascular responses involves neuronal integration/modulation at all levels of the neuraxis. Several vasomotor "control centers" have been described, including sites within the hypothalamus,[14–17] mesencephalic reticular formation,[18–20] medulla,[20–22] and cerebellum.[24] The hypothalamus, reticular formation, and major primary afferent terminations of the baroreceptors, including the nucleus tractus solitarius, have extensive interconnections modulating cardiovascular homeostasis. In addition, the fornix and infundibular nucleus, found within the present stimu-

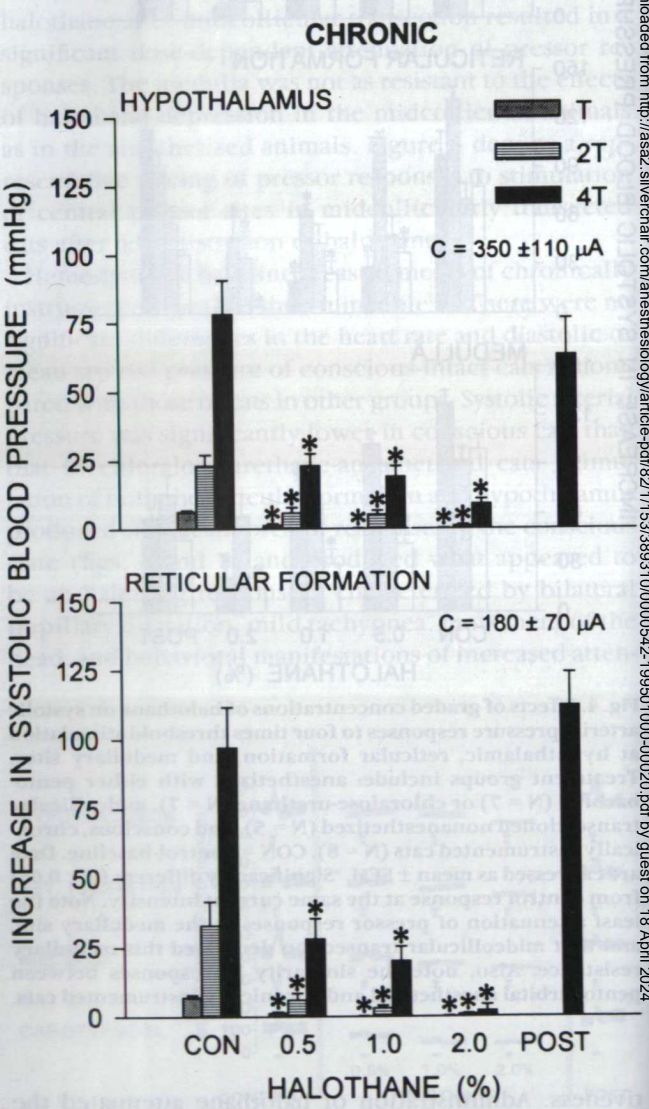

Fig. 7. Effects of graded concentrations of halothane on systolic arterial pressure responses to threshold (T) and two and four times threshold (2T and 4T) stimulation at hypothalamic and reticular formation sites in nonanesthetized, chronically instrumented cats (N = 8). C = Threshold current intensity; CON = control-baseline. Note the marked blunting of pressor responses by halothane. Data are expressed as mean ± SEM. *Significantly different ($P < 0.05$) from control response at same current intensity.

Table 2. Change in the Pressor Response (mmHg) to 10-Second Bilateral Carotid Occlusion after Halothane Administration in Cats Anesthetized with Pentobarbital or Chloralose–Urethane

Group	BCO Control	Halothane 0.5%	Halothane 1.0%	Halothane 2.0%	Post
PB	31 ± 3	14 ± 2*	9 ± 1*	4 ± 1*	21 ± 5
CU	25 ± 3	15 ± 2*	12 ± 2*	9 ± 4*	20 ± 4

BCO = bilateral carotid occlusion; PB = pentobarbital (n = 6); CU = chloralose–urethane (n = 6).

Data are mean ± SEM. Note that halothane attenuates pressor responses to BCO similarly in cats anesthetized with PB and CU.

* Significantly different ($P < 0.05$) from BCO control.

lation areas, also exhibit cardiovascular regulatory roles.

Volatile anesthetics disrupt cardiovascular function through a variety of mechanisms, including direct effects on the myocardium,[1,5] peripheral vasculature,[2] sympathetic ganglia,[3–5] sympathetic and parasympathetic tone,[5] and the CNS.[6–10,12] The mechanism by which halothane attenuates CNS-stimulation-induced pressor responses may involve actions at several locations along the sympathoexcitatory pathway *via* alterations in the responsiveness or gain of CNS cardiovascular centers, and/or by modulating the responsiveness of pre- or postganglionic neurons. General anesthetics, including the volatile agents,[23,25,26] α-chloralose,[27–28] urethane,[29,30] and barbiturates,[31,32] have all been demonstrated to differentially alter CNS cardiovascular control centers. However, the relative importance of central *versus* peripheral mechanisms of general anesthetics in terms of modulation of cardiovascular responses remains unclear. Halothane, delivered to an isolated cephalic circulation in pentobarbital-anesthetized dogs, decreased arterial pressure, heart rate, contractile force, and hemodynamic responses to carotid sinus occlusion.[6] Also, in vagotomized, decerebrate dogs anesthetized with chloralose, local microinjections of halothane, cyclopropane, or procaine decreased medullary stimulated pressor and depressor responses,[7] indicating a halothane-mediated depression of CNS-mediated cardiovascular responses. However, these studies used only one stimulus intensity and local microinjection of solvent equilibrated with anesthetics. In contrast, Wang *et al.*[8] found little or no reduction of reflex vascular responses to medullary stimulation when halothane was delivered to a cephalic perfusion, yet marked changes in responses when halothane was delivered caudally to the body only, indicating that halothane may exert its hypotensive effect, in part, *via* peripheral circulatory depression.

Excitatory synapses in the CNS are typically depressed by general anesthetics, while synapses mediating inhibitory neurotransmission may be depressed or excited by general anesthetics, depending on the brain region and type of anesthetic. General anesthetics have been shown to inhibit sympathetic ganglionic nerve transmission.[3–5,33] Seagard *et al.*[9,10] demonstrated that halothane increased baroreceptor afferent activity in anesthetized dogs while also inhibiting postganglionic nerve activity to a greater degree than preganglionic activity, indicating a ganglionic blocking effect. Administration of halothane attenuated excitation of mesencephalic reticular formation neurons,[25] as well as thermosensitive neurons in the preoptic region of the anterior hypothalamus.[26]

Poterack *et al.*[23] demonstrated that the administration of isoflurane markedly attenuates all hemodynamic responses after stimulation of reticular formation and ventrolateral hypothalamic sites in chronically instrumented cats. In contrast, the intravenous anesthetics midazolam and etomidate attenuated the pressor responses to stimulation, but not the chronotropic or abdominal aortic flow responses. In the current study, after reticular formation stimulation and, to a lesser degree, hypothalamic stimulation, animals exhibited a consistent behavioral attentiveness. These effects were consistent with a degree of ascending reticular activation of rostral CNS sites. Although 0.5% halothane appeared to produce a light anesthesia with preservation of such responses, higher levels of anesthesia attenuated the behavioral responses, possibly through blunting of this reticular activation. Results of the current study agree with those of Ngai *et al.*[34] in that halothane depressed the pressor response to hypothalamic or mesencephalic stimulation; however, in that study, medullary stimulation was not performed. Cardiovascular control centers in the hypothalamus and reticular formation modulate sympathetic outflow partially through more diffuse polysynaptic neuronal pathways than sites in the medulla and, thus, may be more susceptible to effects induced by halothane.

In the current study, we have demonstrated that the type of baseline anesthetic used significantly modulates the effects of halothane on CNS cardiovascular control. Chloralose or the combination of chloralose and urethane generally remain the most popular baseline anesthetics used in cardiovascular or neuroscience lab-

oratory studies. Classically, the intravenous anesthetic agent chloralose has been employed to preserve reflex behaviors and hemodynamics and cause minimal depression of the baroreceptor reflex as compared with barbiturates.[25,28,33] However, chloralose also depresses resting minute volume and the ventilatory response to CO_2.[34] Urethane is an intravenous agent that is used for inducing profound anesthesia with minimal effects on circulatory dynamics. However, urethane also appears to inhibit α-2 mediated cardiovascular responses, both peripherally and in the CNS.[27] Administration of pentobarbital has been demonstrated to reduce sympathetic nervous activity in brainstem-intact and decerebrated cats.[30] Barbiturates have no effect on cervical preganglionic sympathetic activity in C1 spinal cord-transected cats, indicating an inhibition of medullary pressor neurons.[30]

Several previous investigations have demonstrated that anesthesia interferes with autonomic responses and central mechanisms regulating the cardiovascular system.[17,27,29,31,34] The administration of urethane or pentobarbital has been shown to significantly attenuate or even reverse the pressor effect caused by local microinjection of norepinephrine into the nucleus tractus solitarius.[27] Intracerebroventricular injection of dopamine has been demonstrated to increase blood pressure in anesthetized cats,[35] but to decrease blood pressure in conscious dogs.[36] Similarly, stimulation of the locus coeruleus,[37] or paraventricular nucleus,[17] which causes increases in blood pressure in conscious rats results in opposite effects in rats anesthetized with chloralose.[28,38] The current study shows that significant pressor responses may be elicited by stimulation at hypothalamic, reticular formation, and medullary sites in cats anesthetized with pentobarbital or chloralose-urethane, and that these responses were similar to those observed in conscious, chronically instrumented cats. However, the stimulation current intensities required to elicit a threshold response tended to be greater in pentobarbital-anesthetized cats than either chloralose-urethane-anesthetized cats or nonanesthetized, micollicularly transected or intact animals. This finding indicates that pentobarbital more profoundly suppressed these vasomotor areas, rendering them less susceptible to electrical stimulation-induced perturbations. Although the administration of graded doses of halothane attenuated the evoked pressor responses in anesthetized cats as it did in conscious animals, cats anesthetized with the two baseline anesthetics differed significantly in their sensitivities to the effects of halothane. Pressor responses after CNS stimulation in pentobarbital-anesthetized cats were significantly more attenuated by halothane than were responses in chloralose-urethane-anesthetized cats. Indeed, the blunting of pressor responses by halothane in pentobarbital-anesthetized cats was very similar to that in chronically instrumented, nonanesthetized cats. The conclusions regarding differences between baseline anesthetics is limited by the use of a single dose of each agent and, thus, results may be related to dose rather than type of agent. This does not appear likely based on previous findings regarding differential cardiovascular modulation by these agents. In addition, we have shown that pressor responses that occur after bilateral carotid occlusions are attenuated by halothane to the same extent whether the baseline anesthetic is pentobarbital or chloralose-urethane. In other words, the attenuation, by halothane, of pressor responses evoked by CNS stimulation varied depending on the baseline anesthetic, while those evoked by carotid occlusion were independent of the type of baseline anesthetic. These differential effects of halothane have not been previously described, and their reason(s) are unclear.

The nucleus tractus solitarius and paramedian reticular nucleus receive the primary afferent fibers of the baroreceptors.[20,38,39] From the nucleus tractus solitarius, secondary neurons project to the spinal cord intermediolateral cell column, the site of origin of many sympathetic preganglionic neurons,[40] and to brainstem areas, the hypothalamus, the rostral and ventral lateral medulla, and through the medulla oblongata centrally to the nucleus ambiguous.[21] Based on the current findings, we postulate that halothane is acting in the CNS to modulate pressor responses. More specifically, our results indicate that halothane may be modulating CNS-induced pressor responses partially at the level of the tractus solitarius, nucleus tractus solitarius, or medullary tegmental fields, or at secondary projections from this level, including those to the caudal and ventrolateral medulla (A1 and C1 areas, respectively), which are involved in baroreceptor modulation of the cardiovascular system. An action at modulatory cardiovascular centers within the mesencephalic reticular formation may also be involved. Because the actions of both intravenous anesthetics involve, in part, the mesencephalic reticular formation, it may be predicted that, during carotid occlusion, halothane would disrupt afferent information to cardiovascular control centers in the reticular formation and hypothalamus and at other, more rostral CNS sites, and would, therefore, be in-

dependent of the baseline anesthetic. Additionally, if halothane is acting in the CNS, in part, by modulating the activity of the nucleus tractus solitarius, stimulation at other CNS loci would result in pressor responses that are differentially attenuated depending on the baseline anesthetic. In contrast, if halothane was acting predominantly in the periphery to effect sympathetic efferent activity, the effects of halothane would be independent of baseline anesthetic and independent of the etiology of the pressor response, which was not demonstrated in the current study.

Although isoflurane may preserve baroreflex control mechanisms and maintain sympathetic efferent outflow to a greater extent than halothane,[5,12] the attenuation of hypothalamic and reticular formation pressor responses by isoflurane[23] and halothane were very similar. However, one difference that is apparent between the central modulation of isoflurane and halothane is that, during emergence from isoflurane, conversions of pressor to depressor responses were observed.[23] In the current study, these paradoxic responses did not occur during or after the administration of halothane. The etiology for these conversion responses is unclear, but may be attributable to a selective blunting of predominantly excitatory transmission at low residual levels of isoflurane, resulting in neuronally mediated vasodepression.[23]

To eliminate the confounding influence of a baseline anesthetic, as well as the influence of rostral CNS sites, cats underwent midcollicular transections. Not only did the administration of halothane still attenuate CNS-mediated pressor responses after midcollicular transection, but medullary stimulation-induced pressor responses, typically the most resistant to the effects of halothane, showed increased sensitivity. These findings are in agreement with multiple studies demonstrating that cardiovascular control centers in the medulla are influenced by, and regulated in association with, more rostral CNS locations. Transections of the neuraxis at the midcollicular level differentially spare neuronal pools[41,42] and release lower cardiovascular CNS centers from tonic rostral control, thereby allowing these lower centers to function independently. Indeed, separating the hypothalamus from the caudal brainstem is important in that the mesenchalic reticular formation is known to influence the hypothalamus.[43] Studies in cats with brainstem transections at various levels along the neuraxis allow differentiation of CNS function. For example, various levels of brainstem transections indicated that the primary site of action for cannabinoid-induced hypothermia is in the caudal brainstem, probably ponto-medullary.[42] The current results indicate that the CNS modulation of cardiovascular function by halothane may be partially independent of forebrain structures. Similarly, Rampil *et al.*[44] recently demonstrated that anesthetic potency of isoflurane in terms of somatomotor responses is unchanged after precollicular decerebration, *i.e.,* independent of forebrain structures in the rat.

Limitations in the methodology of the current study, as in any study involving electrical stimulation, include the possibility of current spread and lesions produced by the electrode. The importance of these factors has been lessened by using threshold current levels and minimizing the electrode manipulation. In addition, stimulation of areas adjacent to the target regions does not produce similar hemodynamic responses (unpublished observations). A limitation also includes the possibility that carotid cannulation compromised cerebral blood flow. However, there is no evidence of cerebral ischemia in behavior, hemodynamics, or regional EEG (unpublished observations).

In summary, pressor responses elicited from three distinct CNS sites in the hypothalamus, reticular formation, and medulla were significantly attenuated by halothane. The degree of attenuation was dependent on the presence or absence and type of a baseline anesthetic, the presence of an intact brainstem, and the CNS loci stimulated. These findings imply that an important effect of halothane on the regulation of cardiovascular hemodynamics involves an inhibition of various CNS vasomotor control sites. In addition, this study begins to delineate the role of halothane in the homeostatic modulation of cardiovascular functioning. Based on the results of this investigation, previous and future studies that investigate the autonomic actions of halothane should be critically evaluated in terms of the presence and type of baseline anesthetic used. Future directions for further delineating the role of anesthetics in the central control of cardiovascular modulation should include single neuronal studies in vasomotor regions and *in vitro* studies, such as brain and spinal cord slices, in which neuronal recordings without confounding afferent input may be obtained.

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