

Enhancement by Cyclopropane and Halothane of Heart Rate Responses to Sympathetic Stimulation

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In sympathectomized mongrel dogs anesthetized with pentobarbital the administration of either cyclopropane or halothane was found to increase the chronotropic response to electrical stimulation of postganglionic sympathetic fibers supplying the heart. Neither anesthetic affected the rate at which norepinephrine was liberated from the coronary sinus during nerve stimulation. There was no evidence that cyclopropane affected the rate of metabolism of infused norepinephrine. It was concluded that the increased chronotropic response to cyclopropane and halothane during nerve stimulation depends upon an action exerted beyond the point of transmitter release. Earlier findings have been discussed in the light of the present results.

We have noted already that cyclopropane increases the chronotropic response of the heart to postganglionic sympathetic nervous activity.¹ In this report our previous assertion is documented and a similar effect of halothane is described. Most drugs which act in this way do so by interfering with uptake and restoration of norepinephrine which is liberated from sympathetic nerve terminations on arrival of the nerve action potential.² If cyclopropane and halothane were to behave similarly they would increase the amount of norepinephrine liberated from innervated tissues at any given level of sympathetic nervous activity. Conse-

quently, estimates of this activity which depend upon measurements of plasma concentrations of norepinephrine could be seriously in error. Because of this possibility and since knowledge of the modes of action of anesthetic agents is important in its own right, we elected to determine whether cyclopropane or halothane did affect the sympathetic postganglionic mechanisms.

Methods

Sixteen mongrel dogs weighing from 10 to 12 kg. were studied. All were lightly anesthetized with pentobarbital (25 mg./kg. intravenously), the trachea intubated with a No. 9 cuffed Magill tube and artificial respiration instituted (70 per cent oxygen and 30 per cent nitrogen, or 100 per cent oxygen, 4 LPM, Bird Mark No. 4 respirator) using a to-and-fro system with a Waters soda-lime canister. With the animal in the prone position, an 18-gauge needle was inserted through the L₁-S₁ interspace into the epidural space. Needle position was checked by loss of resistance to air injection and inability to aspirate cerebrospinal fluid. A 30-cm. polyethylene catheter then was inserted through the needle, advanced to its full extent, and the needle withdrawn. A total of 40 ml. mepivacaine (carbo-caine (0.5 per cent) was injected. Invariably a hypertensive response as described by Brewster and his associates³ was noted, followed by a moderate degree of arterial hypotension. Subsequent 20-ml. injections of mepivacaine given at hourly intervals failed to elicit either response, suggesting that sympathetic blockade was still present. On six occasions 20 ml. of Evans blue dye was injected through the catheter at the end of the study; at autopsy dye was found from the midcervical to the lower lumbar regions.

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Following the establishment of epidural anesthesia, the animal was turned to the supine position. Superficial tissues in the midline were dissected, the sternum split, and the thorax entered. At this time a load of 5 cm. H₂O was put on the expiratory valve. The left caudal sympathetic ganglion was identified and the principal efferent branch affecting cardiac rate (middle cardiosympathetic) was mounted on silver bipolar electrodes which were insulated except at the tip. Stimuli of 5 msec. duration and frequencies of either 5 or 8 cps. subsequently were applied via these electrodes from a Tektronix type 161 pulse generator. The e.m.f. employed approximated 6 to 8 volts and was supramaximal. The amperage delivered was monitored by measuring the voltage drop across a 100-ohm resistance, as displayed on the screen of a Tektronix 502 oscilloscope. Changes in amperage indicated either displacement of or "drowning" of the nerve in the tissue fluids or blood. When such changes occurred, their underlying causes were corrected. Once the surgical preparation had been completed, the thorax was filled with mineral oil which had been warmed to 37° C. and the temperature thereafter was maintained constant ($\pm 0.5^\circ$ C.) by external heating.

The arterial blood pressure was transduced by a Statham strain gauge which was connected by polyethylene tubing to the lumen of a femoral artery; the level of arterial pressure was recorded on a Grass polygraph and the arterial pulsations were made to inscribe the cardiac rate by means of a Grass tachograph. Samples of arterial blood were withdrawn at intervals and analyzed for pH, P_{CO₂}, and P_{O₂}, using an Instrumentation Laboratories model 101 electrode assembly. Bicarbonate solution was given intravenously to correct metabolic acidosis when it occurred. The amounts given ranged from none to 88 mEq. in various animals. Pa_{CO₂} was maintained below 40 mm. Hg and Pa_{O₂} was maintained at levels producing full arterial saturation. One hundred to 200 ml. of freshly-collected blood from a conscious donor dog was transfused during the study as required to replace blood lost during operation and sampling. Atropine (0.5 mg./30 min.) was given intravenously in all animals to block the effect of

stimulating vagal fibers which accompany the efferent supply to the canine heart.⁴ All preparations were organized to permit an estimation of the rate of release of catecholamines from the heart. For this purpose, the coronary sinus was catheterized blindly via the right atrial appendage, the position of the catheter subsequently being verified at autopsy. The catheter employed fitted tightly; all of the blood entering the sinus could be collected, and none other. Because of this, the coronary sinus outflow could be measured directly. In addition, the production of catecholamines by the heart could be estimated by analyzing blood * samples drawn from the coronary sinus.⁵ When not being sampled, the effluent blood was returned to the animal via a femoral vein by means of a pump.

In all but three animals the left accelerator nerve was stimulated before, during, and after administration of cyclopropane or halothane. The stimulus was employed for a constant period varying from 60 to 120 seconds in different animals. Withdrawal of samples from the coronary sinus occurred during the last part of the period and was commenced in each experiment at an equal interval (either 45 or 80 seconds) after the stimulus was begun. Eight dogs were given cyclopropane and five were given halothane. The control used for comparison with results during cyclopropane or halothane administration was an average of controls before and after administration of the agents.

In 11 experiments cyclopropane was administered at some point by inhalation, the inspired concentrations ranging from 25 to 30 per cent. Flow rates of 1.5 to 2.0 LPM were employed; 20 minutes elapsed before the new level of anesthesia was regarded as reasonably stable. Cyclopropane concentrations in the end-expired air were analyzed by the method of Linde and Price.⁶ In five experiments 1.0 to 1.5 per cent halothane was given for 20 minutes using a Fluotec vaporizer which had been calibrated by gas chromatography. In all cases the washout time following discon-

* Actually only plasma was analyzed. Preliminary experiments showed that norepinephrine did not enter erythrocytes when added to blood and equilibrated for 5 to 10 minutes at 37° C.

tinuation of the anesthetic and preceding the final control period was 25 to 30 minutes.

In three animals norepinephrine was infused intravenously at a rate ranging from 3 to 9 $\mu\text{g./min.}$ for one hour. After the establishment of a steady level of arterial blood pressure and heart rate, samples of coronary sinus and femoral arterial blood were simultaneously drawn and the plasma analyzed for norepinephrine. Cyclopropane was then administered for 20 minutes and the same procedure repeated. The concentration of cyclopropane in end-expired gas at this time averaged 23 per cent (range 20 to 30). Following this the anesthetic was discontinued for 20 minutes and a third pair of blood samples was withdrawn. The data were analyzed for statistical significance using paired t tests and rank-difference (Spearman) correlation methods.

Results

EFFECTS OF CYCLOPROPANE AND HALOTHANE UPON THE RESPONSE TO POST-GANGLIONIC NERVE STIMULATION

The principal findings are shown in table 1. In brief, the administration of either anesthetic decreased mean arterial blood pressure and cardiac rate. During nerve stimulation, the peak heart rate was increased above the level of the control response by both agents. The pressor response to nerve stimulation was unaltered by cyclopropane but significantly increased by halothane ($P < 0.05$). The coronary sinus blood flow usually was elevated when cyclopropane was given; reduced or unchanged during halothane administration. The increase in blood flow during stimulation

TABLE 1. Responses to High-frequency (5-8cps.) Stimulation of Cardiac Accelerator Nerve in Dogs

	Heart Rate (beats per minute)		Mean Arterial Pressure (mm. Hg)		Coronary Sinus Blood (ml. per minute)		Coronary Sinus Norepinephrine Output ($\mu\text{g.}$ per minute)
	Resting	During Stimulation	Resting	During Stimulation	Resting	During Stimulation	During Stimulation
Cyclopropane (8 Dogs)							
Control	130.1 (8.5)	191.6 (6.0)	74.1 (5.1)	81.1 (6.6)	40.7 (3.8)	60.4 (6.3)	0.298 (0.183)
During cyclopropane administration	124.4 (7.9)	215.4 (7.3)	63.1 (3.4)	68.9 (4.5)	50.8 (12.2)	80.0 (19.1)	0.316 (0.190)
Percentage change	-4.4%	+12.4%	-14.8%	-15.0%	+24.8%	+32.4%	+6.0%
Significance	$P < 0.05$	$P < 0.001$	N.S.	$P < 0.05$	$P < 0.05$	N.S.	N.S.
Halothane (5 Dogs)							
Control	120.6 (7.9)	183.0 (7.1)	83.8 (9.0)	88.3 (10.7)	32.4 (4.7)	67.0 (8.4)	0.270 (0.075)
During halothane administration	110.8 (7.5)	197.8 (8.0)	53.2 (4.4)	70.0 (6.7)	25.6 (3.7)	61.0 (5.8)	0.211 (0.054)
Percentage change	-8.1%	+8.1%	-36.5%	-20.7%	-20.9%	-8.9%	-21.8%
Significance	$P < 0.01$	$P < 0.05$	$P < 0.01$	$P < 0.05$	$P < 0.05$	N.S.	N.S.

() = S.E. of mean; N.S. = not significant.

TABLE 2. Effect of Cyclopropane on Myocardial Uptake of Infused Norepinephrine (3 Dogs)

	NE Infusion Rate ($\mu\text{g./minute}$)	Coronary Sinus Flow (ml./minute)	Arterial NE Concentration ($\mu\text{g./l.}$)	Coronary Sinus NE Concentration ($\mu\text{g./l.}$)	A-V Difference ($\mu\text{g./l.}$)	% Clearance of NE by Heart
Dog CS 20						
Before cyclopropane	9.2	14.8	6.57	0.60	5.97	90.9%
During cyclopropane	9.2	28.1	6.38	0.46	5.92	92.9%
After cyclopropane washout	9.2	20.7	10.33	1.30	9.03	87.2%
Dog CS 25						
Before cyclopropane	3.0	42.9	3.52	1.04	2.48	70.4%
During cyclopropane	3.0	23.8	4.52	0.70	3.82	84.5%
After cyclopropane washout	3.0	29.8	2.82	0.56	2.26	80.1%
Dog CS 29						
Before cyclopropane	3.8	40.4	2.32	0.49	1.83	79.0%
During cyclopropane	3.8	33.3	1.97	0.30	1.67	84.8%
After cyclopropane washout	3.8	24.3	2.32	0.33	1.99	85.9%

NE = norepinephrine (base).

appeared not to be modified by either anesthetic. The output of norepinephrine from the coronary sinus during stimulation was not significantly affected by either cyclopropane or halothane although it usually was increased by the former and reduced by the latter.

The tendency for an increase in norepinephrine output during cyclopropane anesthesia and for a reduction during halothane inhalation was accompanied by a similar directional change in coronary sinus flow. A Spearman rank-difference correlation test was significant at the 5 per cent level. In the absence of stimulation, the rate of norepinephrine liberation was negligible.

In five preparations, arterial plasma concentrations of norepinephrine during stimulation were measured in order to insure that these were insignificant, which they were, averaging 0.1 $\mu\text{g./l.}$ Concentrations in coronary sinus plasma ranged from 1.4 to 45.4 $\mu\text{g./l.}$ in various animals during nerve stimulation.

EFFECTS OF CYCLOPROPANE ON THE UPTAKE OF INFUSED NOREPINEPHRINE BY THE HEART

Before the administration of cyclopropane, roughly 20 per cent of the norepinephrine present in arterial plasma was found in the coronary venous plasma, and the extraction following the discontinuation of cyclopropane

was similar. During cyclopropane administration, the uptake was slightly, but not significantly, increased (table 2). The concentration of norepinephrine in arterial blood also was unchanged by the administration of cyclopropane.

EFFECTS OF TIME ON THE MEASURED VARIABLES

All measurements (Pa_{CO_2} , Pa_{O_2} , pH, norepinephrine output during nerve stimulation, coronary sinus flow, mean arterial blood pressure, and heart rate) made during the first control period were compared with those made after discontinuation of cyclopropane or halothane. No significant difference could be established. Therefore, the actions attributed to cyclopropane and halothane are unlikely to have been the result of deterioration of the preparation.

Discussion

Most drugs which potentiate biological responses to sympathetic nervous activity are believed to act like cocaine, that is, by interfering with the re-uptake and re-storage within sympathetic nerves of norepinephrine which has been released upon the arrival of the nerve action potential.² This re-uptake process is an active one requiring energy supplied via high-energy phosphate bonds (ATP), and it is the

principal means by which sympathetic actions are terminated after sympathetic activity has abated. Metabolism of the released norepinephrine is of little consequence and the administration of monamine oxidase or catechol-O-methyltransferase inhibitors does essentially nothing to modify responses to sympathetic stimulation.⁷ All drugs which have a cocaine-like action increase the amount of norepinephrine leaving a tissue during stimulation of its sympathetic nerves and also inhibit the uptake of infused norepinephrine.⁸ Therefore, the present data show that at least two anesthetics, cyclopropane and halothane, can potentiate sympathetic responses in some manner which does not involve the terminal sympathetic mechanisms. In every animal studied the chronotropic response to accelerator nerve stimulation was enhanced but there was no significant change in norepinephrine output from the coronary circulation.

The present study offered us the opportunity to learn whether the increased plasma concentration previously observed⁹ during cyclopropane inhalation and norepinephrine infusion was caused by some interference with catecholamine metabolism and uptake, or whether it represented an artifact caused by the experimental technique. One possibility is that uptake and storage of infused norepinephrine are inhibited by cyclopropane as a direct consequence of an increased level of sympathetic nervous activity which is normally induced by the anesthetic. As a second possibility, the administration of norepinephrine may, despite its positive inotropic effects, cause cardiac output to diminish by means of reflex suppression of the sinus node.¹⁰ If norepinephrine has this action, it reduces the rate at which it becomes available for storage or metabolism in the tissues, with the result that the concentration in plasma will be higher than in the absence of reflex activity. In the present study, cyclopropane had no effect upon the plasma concentration increment produced by infusion, suggesting that in the presence of sympathetic blockade it failed to affect either metabolism or tissue uptake in the absence of autonomic nervous activity. In a recent report¹¹ Gardier has shown that one atmosphere of cyclopropane can inhibit the

activity of catechol-O-methyltransferase by 29 per cent. Our experiments suggest that this effect is not detectable at clinically useful concentrations of cyclopropane.

Our previous studies have shown that the administration of cyclopropane causes an increase in circulating catecholamine levels in both man and dog.^{9,12} In the present series, the administration of cyclopropane had no effect on plasma catecholamine levels, indicating that the concentration increments in normal animals depend upon the presence of efferent sympathetic nervous activity. Since the degree of augmentation of catecholamine output during nerve stimulation in the present series was small (6 per cent) in relation to the increased plasma concentrations accompanying cyclopropane inhalation in intact men and dogs (about 300 per cent), it is probable that the activity of the sympathetic nervous system is increased by this anesthetic in both species. Observations in the rabbit supporting this conclusion, recently have been secured by means of recordings of nerve action potentials.¹³ We conclude that one pharmacologic effect of cyclopropane is to increase sympathetic nervous activity in proportion to the depth of anesthesia.

In the case of halothane, there is agreement that plasma levels of norepinephrine are not increased in man or in the dog.^{9,14} The present study shows that since halothane does not inhibit the release of norepinephrine by sympathetic nerve impulses, the previous studies of plasma norepinephrine concentration indicate that halothane does not stimulate sympathetic nervous activity, either in dogs or in man. On the other hand, recent evidence of such stimulation has been obtained in the rabbit by means of nerve impulse recording.¹³ In view of the foregoing argument, we must view those findings as representing a species difference.

Summary and Conclusions

In sympathectomized dogs cyclopropane and halothane increased the chronotropic response of the heart to stimulation of postganglionic sympathetic nerves. This effect was not accompanied by an increase in the rate of liberation of norepinephrine from the

coronary sinus. In the case of cyclopropane there was no evidence of any interference with the tissue uptake of infused norepinephrine. We conclude that cyclopropane and halothane, unlike cocaine, can enhance the chronotropic response to accelerator nerve stimulation without modifying the mechanism of norepinephrine release and re-storage. We found no evidence that cyclopropane interferes with the metabolism of norepinephrine.

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References

1. Price, H. L., and Price, M. L.: Relative ganglion-blocking potencies of cyclopropane, halothane and nitrous oxide, and the interaction of nitrous oxide with halothane, *ANESTHESIOLOGY* 28: 349, 1967.
2. Koelle, G. B.: Neurohumoral transmission and the autonomic nervous system. In *The Pharmacological Basis of Therapeutics*. Goodman, L. S., and Gilman, A., Editors. New York, MacMillan Co., 1965, p. 433.
3. Brewster, W. R., Isaacs, J. P., Osgood, P. F., and King, T. L.: The hemodynamic and metabolic inter-relationships in the activity of epinephrine, norepinephrine and the thyroid hormones, *Circulation* 13: 1, 1956.
4. Mizeres, N. J.: The origin and course of the cardioaccelerator fibers in the dog, *Anat. Rec.* 132: 261, 1958.
5. Price, H. L., and Price, M. L.: The chemical estimation of epinephrine and norepinephrine in humans and canine plasma. II. A critique of the trihydroxyindole methods, *J. Lab. Clin. Med.* 50: 769, 1957.
6. Linde, H. W., and Price, H. L.: Gas analyzer for rapid estimation of cyclopropane, *ANESTHESIOLOGY* 19: 757, 1958.
7. Crout, J. R.: Effect of inhibiting both catechol-o-methyl transferase and monoamine oxidase on cardiovascular responses to norepinephrine, *Proc. Soc. Exp. Biol. Med.* 108: 482, 1961.
8. Muscholl, E.: Pharmacology of cholinergic and adrenergic transmission. Koelle, G. B., Douglas, W. W., and Carlson, A., Editors. New York, MacMillan Company, 1965, p. 291.
9. Price, H. L., Linde, H. W., Jones, R. E., Black, G. W., and Price, M. L.: Sympathoadrenal responses to general anesthesia in man and their relation to hemodynamics, *ANESTHESIOLOGY* 20: 563, 1959.
10. Barcroft, H., and Starr, L.: Comparison of the actions of adrenaline and noradrenaline on the cardiac output in man, *Clin. Sci.* 10: 295, 1951.
11. Gardier, R. W., Endahl, G. L., and Hamelberg, W.: Cyclopropane effect on catecholamine bio transformation, *ANESTHESIOLOGY* 28: 677, 1967.
12. Deutsch, S., Linde, H. W., and Price, H. L.: Circulatory and sympatho-adrenal responses to cyclopropane in the dog, *J. of Pharmacol. Exper. Ther.* 135: 354, 1962.
13. Millar, R. A., and Biscoe, T. J.: Post-ganglionic sympathetic discharge and the effect of inhalation anaesthetics, *Brit. J. Anaesth.* 38: 92, 1966.
14. Millar, R. A., and Morris, M. E.: Induced sympathetic stimulation during halothane anaesthesia, *Canad. Anaesth. Soc. J.* 7: 423, 1960.

Anesthesia

UMBILICAL-CORD VENOUS PRESSURE Oxytocics have been used during the latter part of the second stage of labor to reduce postpartum blood loss. In a series of normal deliveries, venous pressure in the umbilical cord immediately after delivery was measured. Some of the mothers had received 0.2 mg. methylethergonine intravenously when the fetal head crowned. The mean cord venous pressure in the control group was 269.7 mm. of blood, but when the oxytocic had been used, the mean pressure was significantly higher, 429.6 mm. of blood. Multiparity and anesthesia did not seem to alter the cord venous pressure. Although all infants were normal on follow-up study, it is suggested that the abrupt and marked increase in cord venous pressure produced by intrapartum oxytocic drugs might be injurious to an infant with a cardiovascular abnormality. (*LeDonnen, A. T., and McGowan, L.: Effect of an Oxytocic on Umbilical Cord Venous Pressure, Obstet. Gynec.* 30: 103 (July) 1967.)