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The Influence of Vagal Afferents on the Left Ventricular Contractile Response to Intracoronary Administration of Catecholamines in the Conscious Dog

KIRK W. BARRON AND VERNON S. BISHOP

SUMMARY We studied the effect of vagal afferents on the left ventricular contractile response (LdP/dtmU) to administration of sympathomimetic amines into the left circumflex coronary artery in conscious dogs. The positive inotropic effects of bolus administration of epinephrine, norepinephrine, and isoproterenol were greater during bilateral vagal cold block (BVB) than in the control state with the vagi intact (P < 0.001). The inotropic potentiation observed with BVB was not due to vagal efferent interruption, since parasympathetic efferent block with atropine did not potentiate the inotropic effects of intracoronary epinephrine (P > 0.05). BVB also potentiated the inotropic effects of constant intracoronary infusion of epinephrine (P < 0.01). BVB alone had no significant effect on LdP/dtmU; however, BVB during constant intracoronary infusion of epinephrine did result in an increase in LdP/dtmU (P < 0.05). We propose that the potentiation of the inotropic effects during BVB could result from either interruption of a vagal afferent-mediated negative feedback reflex and/or an unmasking of a sympathetic afferent-mediated positive feedback effect induced by BVB. However, the observations that ganglionic blockade also potentiated the contractile effects of epinephrine supports the concept that a negative feedback reflex does attenuate the inotropic effect of catecholamines in the control state. In conclusion, these data suggest that vagal afferents attenuate the positive inotropic effects of intracoronary administration of catecholamines via a cardiocardiac negative feedback reflex which occurs through a vagal afferent inhibition of sympathetic efferent activity back to the left ventricle.


NUMEROUS studies have shown that unmyelinated vagal afferents originating from mechanoreceptors in the lungs, atria, and ventricles exert a tonic inhibitory influence on sympathetic outflow from the vasomotor center (Thorén et al., 1976). Atrial receptors respond to deformation (Linden, 1976) and similarly, receptors in the lungs are sensitive to deformation through either stretch or deflation (Paintal, 1977). However, the stimulus for ventricular receptors is less certain.

Afferent vagal C fiber activity originating from ventricular receptors has been shown to rise with increases in intraventricular pressure (Sleight and Widdicombe, 1965; Öberg and Thoren, 1972). Also, it has been reported that elevations in intraventricular pressure produce vagal afferent-mediated reflex vasodilation and bradycardia (Aviado and Schmidt, 1959; Salisbury et al., 1960; Rose et al., 1961; Mark et al., 1977; Chevalier et al., 1974; Zelis et al., 1977). The stimulus for these vagal afferent receptors is less certain.

A recent study from this laboratory, in anesthe-
tized cats by Shimizu et al. (1979), has demonstrated that the increase in mean arterial pressure during vagal cold block was related to the existing cardiac sympathetic efferent activity. Furthermore, vagal cold block was also found to produce an increase in left ventricular contractility. Shimizu et al. (1979) postulated that cardiac sympathetic activity may modulate vagal afferent activity from cardiopulmonary receptors through changes in ventricular contractility. They suggested that this could serve as a negative feedback system to regulate myocardial contractility, such that increases in cardiac sympathetic activity could increase cardiac vagal afferent activity which, in turn, could act to restrain sympathetic outflow to the heart and thus buffer increases in contractility. A similar feedback system was suggested by Fox et al. (1977).

This study was designed to test the hypothesis that increases in the inotropic state of the left ventricle in conscious dogs activate ventricular receptors with vagal afferents which, in turn, reflexly alter the inotropic state of the left ventricle through changes in sympathetic outflow from the vasomotor center. Results suggest that positive inotropic stimulation of the left ventricle can initiate a vagal afferent-sympathetic efferent negative feedback reflex that reduces sympathetic outflow to the heart, thereby attenuating the inotropic effects of sympathomimetic amines.

**Methods**

Nineteen mongrel dogs weighing from 14 to 27 kg were used in this study. Anesthesia was induced with intravenous sodium pentobarbital (20 mg/kg). The animals were then intubated and ventilated artificially with a Bird respirator during the course of the thoracic instrumentation. Anesthesia was maintained with halothane, and under sterile procedures a left thoracotomy was performed through the 4th intercostal space. To measure systemic arterial pressure, an 18-gauge polyvinyl catheter was placed in the thoracic aorta proximal to the aortic arch. Another 18-gauge catheter was placed in the left atrial appendage to measure left atrial pressure. A solid state pressure transducer (Konigsberg P-18) was implanted within the left ventricle through a stab wound near the apex.

The left circumflex coronary artery was exposed near its origin by blunt dissection. A polyethylene catheter was placed in the proximal left circumflex coronary artery for administration of drugs into the coronary circulation. Approximately 1 cm of PE 10 tubing was implanted within the lumen of the vessel. The PE 10 segment was joined to a 50-cm length of PE 50 tubing. For support and protection of the PE 50 tubing, an 18-gauge polyvinyl catheter was placed over the length of the PE 50. All catheters and leads were exteriorized from the dorsal midcervical region of the neck.

**Denervation of Aortic Arch Baroreceptors**

To eliminate aortic baroreceptor reflexes which may travel with the vagus nerves, the aortic arch was denervated at the time of instrumentation by surgically stripping the nerves and connective tissue from the aortic arch and applying isopropyl alcohol to the region. Verification of denervation was determined during subsequent denervation of the carotid sinus by the absence of a heart rate response due to bilateral carotid occlusion prior to closure of the neck. Postoperatively, the completeness of the sinoaortic denervation was verified when the reflex heart rate responses to nitroglycerin (6 μg/kg, iv) or phenylephrine (5 μg/kg, iv) were eliminated or when heart rate changed by less than 6 beats/min.

**Implantation of Vagal Cooling Coils**

Two weeks after the thoracic surgery, each animal was anesthetized with intravenous sodium thiopental (30 mg/kg). A ventral midline incision was made in the mid-cervical region, and a 6-cm length of both the right and left vagosympathetic nerve trunks was dissected from the common carotid artery and surrounding tissues. The cervical sympathetic nerves were carefully dissected and removed from the vagal nerves to prevent cold block of the cervical sympathetics. A coil made of 10-gauge stainless steel tubing was placed around each vagal nerve trunk, and a Silastic tubing connected to each coil was exteriorized through the skin. After placement around the vagus, each coil then was insulated from the surrounding tissue with a jacket fashioned from medical grade Silastic rubber. The carotid arteries were isolated from the coil by suturing a layer of muscle over the arteries. To monitor temperature of the region surrounding the nerves, thermocouple wires were placed inside each coil near the nerve. The leads from the thermocouples were exteriorized at the back of the neck so the temperature within the coils could be monitored continuously on either the pen recorder or an oscilloscope. The nerve activity in the vagi was blocked by circulating cooled antifreeze through each coil while the temperature between the coil and the vagus was monitored continuously. Bilateral vagal cold block (BVB) was assumed to be complete when the temperature reached 2° to 0°C. At this temperature, heart rate was elevated and respiration became slower and deeper. The block was reversed by circulating warm fluid through the coils. The details of this technique have been published previously (Bishop and Peterson, 1978).

The effectiveness of BVB was assessed by two methods. The first method tested the completeness of vagal efferent blockade and required that the heart rate responses were similar with BVB and atropine. The second method tested vagal afferent blockade and was determined when the bradycardia and hypotension observed with intracoronary injec-
tion of veratridine (0.1 µg/kg) were abolished. Bishop and Peterson (1976) also have reported that evoked responses due to electrical stimulation of the vagus nerves are eliminated by BVB. Gross and microscopic examinations of the vagus nerve trunk verified that no pathological changes in the nerve trunk had occurred during the course of instrumentation (Bishop and Peterson, 1978). After implantation of the cooling coils, an interval of 3 days was allowed for the dogs to recover before experiments were initiated.

**Recording Procedures**

Arterial and left atrial pressures were measured with Statham P23Dc and P23BB strain gauge manometers, respectively. Aortic and left atrial pressures were zeroed to the midline of the sternum with the dog lying on its right side. The solid state pressure transducers were precalibrated at 38°C before implantation. To correct for zero drift of the pressure cells, the left ventricular end-diastolic pressure was set equal to mean left atrial pressure (Horwitz and Bishop, 1972). To obtain left ventricular dP/dtmax, left ventricular pressure was differentiated with an operational amplifier; signal amplitude decreased by 3 dB at 100 Hz. All signals were inscribed on a Beckman type R dynograph recorder.

**Experimental Protocol**

All experiments were performed while the dogs were conscious and lying unrestrained on a hammock. The doses of epinephrine used in these experiments met two criteria: (1) left ventricular dP/dtmax increased with intracoronary drug administration and (2) neither intracoronary nor intravenous administration of the same dose had any effect on mean arterial pressure or heart rate. Unless otherwise stated, all administrations of sympathomimetic agents can be assumed to be into the left circumflex coronary catheter. In vagal block studies, bolus doses of epinephrine (0.01, 0.02, 0.03, 0.04, and 0.05 µg/kg) were administered randomly with intact vagi or during bilateral vagal cold block. Bolus doses of norepinephrine (2.5 and 5.0 mg/kg) and isoproterenol (4.0 mg/kg) also were administered into the coronary circulation. The effects of constant infusion of epinephrine (0.1 µg/kg per min) were examined with the vagi intact and during vagal cold block. An infusion withdrawal pump, Harvard model 903, was used in all constant infusion experiments. The effects of administering fluid into the coronary circulation were determined by injection of volumes of lactated Ringer’s solution (2.0 ml) into the circumflex catheter. This volume was equal to or greater than the volumes of drugs which were administered into the circumflex catheter.

Since vagal cold block interrupts both afferent and efferent nerve traffic, it was not possible to determine whether differences in cardiovascular responses observed during vagal cold block were due to blockade of afferent fibers, efferent fibers, or both. Atropine sulfate (0.1–0.2 mg/kg) was used to determine the effects of parasympathetic efferent blockade on the myocardial contractile response to the inotropic agents. To assess further the involvement of vagal afferent nerves, the inotropic effects of epinephrine were compared before atropine, after administration of atropine, and then during vagal cold block following atropine. The effectiveness of atropine was determined by elimination of the tachycardia and hypotension due to intravenous acetylcholine (10 µg/kg).

To assess the inotropic effects of epinephrine without cardiovascular reflex involvement, ganglionic blockade was used. Hexamethonium (7–12 mg/kg) plus atropine (0.05 mg/kg) was administered to five dogs and trimethaphan (iv drip) to one other. Ganglionic blockade was judged effective when the following effects were eliminated: bradycardia with intravenous phenylephrine (50 µg), tachycardia with intravenous nitroglycerin (12 µg/kg), or the bradycardia and hypotension with intracoronary administration of veratridine (0.1 µg/kg).

**Drugs**

The following compounds were used: epinephrine bitartrate (Sigma), norepinephrine HCl (Sigma), atropine sulfate injection (Eli Lilly), trimethaphan camysylate (Arfonad, Roche Laboratories), hexamethonium HCl (National Biochemicals Company), and isoproterenol HCl (Sterling-Winthrop Research Institute). Drugs were dissolved in lactated Ringer’s solution immediately prior to each experiment. Doses of drugs are expressed in the conjugate salt form.

**Statistical Analysis**

Comparison of means were analyzed by Student’s t-test for paired data. A within-groups, two-way analysis of variance was employed in comparison of groups of more than two. Significant main effects were determined by individual group comparisons with the Newman-Keuls range test (Dayton, 1970). Linear regression analyses were used to test for correlations. To determine whether the augmentation of the inotropic effects of epinephrine by vagal cold block represented a true potentiation, the two curves illustrated in Figure 2 were tested for parallelism (Goldstein, 1964). The criterion for significance was set at the 95% confidence level, P < 0.05.

**Results**

**The Effects of Bilateral Vagal Cold Block and Atropine**

The effects of bilateral vagal cold block and muscarinic blockade are shown in Table 1. Cooling
the vagi (0°–2°C) resulted in significant increases in mean arterial pressure, left ventricular systolic pressure, and heart rate, while mean left atrial pressure was decreased and left ventricular dP/dtmax was not altered. In this study, vagal cold block did not alter blood pH, PCO₂ or PO₂. Muscarinic receptor blockade with atropine sulfate (0.1–0.2 mg/kg) produced a tachycardia similar to that observed with bilateral vagal cold block and a decrease in mean left atrial pressure. Atropine did not significantly alter mean arterial blood pressure, left ventricular systolic pressure, or dP/dtmax.

### Intracoronary Bolus Administration of Epinephrine

Intracoronary injection of Ringer’s solution produced no significant changes in any of the parameters measured. The average increase in LVdP/dt max in eight dogs was 1.2% with the vagi intact and 1.0% during vagal cold block. As shown in Table 2, intravenous administration of epinephrine, 0.05 μg/kg, produced no significant change in any of the measured parameters, but when this same dose was administered into the left circumflex circulation, an increase in heart rate was found before (14 ± 6 beats/min, P < 0.05) and during (13 ± 5 beats/min, P < 0.01) vagal cold block. To avoid possible heart rate effects with intracoronary epinephrine administration, two lower doses of epinephrine (0.02 and 0.04 μg/kg) were selected and administered in subsequent protocols. Responses to intracoronary injections of epinephrine (0.04 μg/kg) before and during vagal cold block are illustrated in Figure 1. Note that whereas the inotropic effects of epinephrine

### Table 1 Effects of Bilateral Vagal Cold Block and Atropine

<table>
<thead>
<tr>
<th></th>
<th>Mean arterial pressure (mm Hg)</th>
<th>Heart rate (beats/min)</th>
<th>Mean left atrial systolic pressure (mm Hg)</th>
<th>Left ventricular dP/dtmax (mm Hg/sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CON</td>
<td>Δ BVB</td>
<td>Δ CON</td>
<td>Δ BVB</td>
</tr>
<tr>
<td></td>
<td>93 ± 2</td>
<td>116 ± 4</td>
<td>15 ± 3</td>
<td>15 ± 2</td>
</tr>
<tr>
<td></td>
<td>±2</td>
<td>±10</td>
<td>±10</td>
<td>±10</td>
</tr>
<tr>
<td></td>
<td>4.9 ± 0.8</td>
<td>2.2 ± 0.9</td>
<td>118 ± 4</td>
<td>136 ± 4</td>
</tr>
<tr>
<td></td>
<td>3 ± 3</td>
<td>4 ± 4</td>
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<td></td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

### Table 2 Changes Produced by Bolus Administration of Epinephrine in the Control State and during Vagal Cold Block

<table>
<thead>
<tr>
<th>Epinephrine</th>
<th>Mean arterial pressure (mm Hg)</th>
<th>Heart rate (beats/min)</th>
<th>Mean left atrial systolic pressure (mm Hg)</th>
<th>Left ventricular dP/dtmax (mm Hg/sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Δ CON</td>
<td>Δ BVB</td>
<td>Δ CON</td>
<td>Δ BVB</td>
</tr>
<tr>
<td>0.01 μg/kg ic</td>
<td>2 (10§)</td>
<td>1</td>
<td>-0.3</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>±2</td>
<td>±1</td>
<td>±2</td>
<td>±1</td>
</tr>
<tr>
<td>0.02 μg/kg ic</td>
<td>2</td>
<td>6</td>
<td>±0.2</td>
<td>±1</td>
</tr>
<tr>
<td></td>
<td>±2</td>
<td>±4</td>
<td>±0.6</td>
<td>±1</td>
</tr>
<tr>
<td>0.03 μg/kg ic</td>
<td>2</td>
<td>6</td>
<td>±0.2</td>
<td>±1</td>
</tr>
<tr>
<td></td>
<td>±2</td>
<td>±6</td>
<td>±0.3</td>
<td>±1</td>
</tr>
<tr>
<td>0.04 μg/kg ic</td>
<td>1</td>
<td>6</td>
<td>±0.4</td>
<td>±1</td>
</tr>
<tr>
<td></td>
<td>±2</td>
<td>±6</td>
<td>±0.9</td>
<td>±1</td>
</tr>
<tr>
<td>0.05 μg/kg ic</td>
<td>1</td>
<td>5*</td>
<td>-0.8</td>
<td>±2</td>
</tr>
<tr>
<td></td>
<td>±1</td>
<td>±4</td>
<td>-0.1</td>
<td>±2</td>
</tr>
<tr>
<td>0.06 μg/kg ic iv</td>
<td>2 (7§)</td>
<td>6</td>
<td>0.9 (5§)</td>
<td>±1</td>
</tr>
<tr>
<td></td>
<td>±3</td>
<td>±5</td>
<td>±0.7</td>
<td>±1</td>
</tr>
<tr>
<td>0.07 μg/kg ic iv</td>
<td>2 (7§)</td>
<td>6</td>
<td>0.9 (7§)</td>
<td>±1</td>
</tr>
<tr>
<td></td>
<td>±3</td>
<td>±5</td>
<td>±0.7</td>
<td>±1</td>
</tr>
</tbody>
</table>

Results are expressed as mean ± SE produced by administration of epinephrine in the control state (Δ CON) and during bilateral vagal cold block (Δ BVB); ic = intracoronary administration; iv = intravenous administration.

* P < 0.001; † P < 0.01; ‡ P < 0.001.
§ Number of dogs with intracoronary administrations.
| Number of dogs with intravenous administrations. |
VAGAL AFFERENTS AND LEFT VENTRICULAR CONTRACTILITY/Barron & Bishop

MAP (mm Hg)
0 150
0
240
HR (beats/min)
0
MLAP (mm Hg)
0 10
LVP (mm Hg)
0 270
LVdP/dt_{max} (mm Hg/sec)
0 5000

**FIGURE 1** Representative recording of the effects of intracoronary administration of epinephrine (0.04 μg/kg) in the conscious dog with the vagi intact (control) and during bilateral vagal cold block (BVB). MAP = mean arterial pressure, HR = heart rate, MLAP = mean left atrial pressure, LVP = left ventricular pressure, LVdP/dt_{max} = maximum rate of rise of left ventricular pressure, ↑ EPI = beginning of epinephrine injection.

were potentiated during vagal cold block, there were no concomitant changes in either mean arterial pressure or heart rate associated with the contractile response to epinephrine.

Intracoronary bolus administration of epinephrine resulted in dose-dependent increases in dP/dt_{max} with the vagi intact and during vagal cold block (Table 2); however, the inotropic effects of intracoronary epinephrine were greater during vagal cold block than when the vagi were intact (P < 0.001, Fig. 2). For example, intracoronary administration of epinephrine (0.04 μg/kg) resulted in an increase in dP/dt_{max} of 1150 ± 174 mm Hg/sec during vagal cold block and this was greater than the increase of 829 ± 171 mm Hg/sec produced by epinephrine when the vagi were intact (P < 0.001). The control effects of epinephrine on peak left ventricular systolic pressure were variable, with significant increases at 0.02 and 0.04 μg/kg. During vagal cold block, intracoronary epinephrine produced increases in left ventricular systolic pressure at each dose tested (Table 2).

**FIGURE 2** Change in LVdP/dt_{max} due to epinephrine in the control state (○) and during bilateral vagal cold block, BVB (△). *** P < 0.001. Analysis of variance indicated that the two lines were significantly different (P < 0.001). A test for parallelism demonstrated that there was true potentiation. Mean ± SE, n = 10.

**FIGURE 3** Change in LVdP/dt_{max} due to intracoronary epinephrine in the control state, during atropine alone, and during BVB following atropine. Atropine alone did not alter the inotropic effects of epinephrine from the control effects (P > 0.05). The increases in LVdP/dt_{max} produced by epinephrine (0.02 and 0.04) during atropine + BVB were greater than the responses during atropine (P < 0.05). Mean ± SE, n = 6.
Individual group comparisons with the Newman-Keuls range test demonstrated that the inotropic effects of epinephrine were not altered by atropine ($P > 0.05$); however, the contractile changes during BVB were greater than either the control inotropic effects ($P < 0.05$) or the inotropic effects after atropine ($P < 0.05$). Furthermore, the amount of potentiation (change during vagal cold block minus change during control) produced by epinephrine during vagal cold block was similar in both control and atropinized states. Vagal cold block potentiated the inotropic responses to 0.02 and 0.04 $\mu$g/kg epinephrine by 406 ± 148 and 321 ± 126 mm Hg/sec, respectively, prior to atropine, which were similar to the potentiations following atropine (353 ± 84 and 483 ± 131 mm Hg, respectively).

Linear regression analyses were applied to determine if the magnitude of the potentiation of the inotropic effects of epinephrine was related to the increase in mean arterial pressure that occurs during vagal cold block. Two analyses were performed, one for the inotropic effects of 0.02 $\mu$g/kg epinephrine vs. the pressor effects of vagal cold block and a similar analysis for 0.04 $\mu$g/kg epinephrine. At these two doses there were no significant correlations between the inotropic potentiations during vagal cold block and the increases in pressure during vagal cold block. The correlation coefficients for 0.02 and 0.04 $\mu$g/kg epinephrine were: $r = 0.09$, df = 8 and $r = 0.05$, df = 8, respectively.

**Intracoronary Bolus Administration of Norepinephrine and Isoproterenol**

In five dogs we examined the inotropic effect of intracoronary injection of two other sympathomimetic amines, norepinephrine and isoproterenol, before and during bilateral vagal cold block. As shown in Table 3, intracoronary administration of both of these agents increased the inotropic state of the left ventricle without altering heart rate or mean arterial pressure. The $dP/dt_{\max}$ responses to doses of norepinephrine (2.5 and 5.0 ng/kg) were increased during vagal cold block (Table 3). Similarly, injection of isoproterenol (4.0 ng/kg) produced an increase in $dP/dt_{\max}$ during bilateral vagal cold block, which was greater ($P < 0.01$) than the control effect with the vagi intact. These data indicate that the inotropic responses to sympathomimetic amines are augmented when vagal afferents are interrupted by vagal cold block.

**Constant Infusion of Epinephrine**

Constant intracoronary infusion of epinephrine produced a positive inotropic response that was greater during vagal cold block (855 ± 177 mm Hg/sec) than when the vagi were intact (455 ± 62 mm Hg/sec, $P < 0.001$; Table 4). Left ventricular systolic pressure was decreased during epinephrine infusion (5 ± 1.0 mm Hg, $P < 0.05$) when the vagi were intact but not during vagal cold block (3 ± 1.5 mm Hg, $P > 0.05$). Constant infusion of epinephrine had no significant effect on any other parameters, either with the vagi intact or during vagal cold block (Table 4). In agreement with previous results of this study, vagal cold block in this group of dogs produced no effect on $dP/dt_{\max}$. However, when vagal cold block was performed during epinephrine infusion, vagal cold block increased $dP/dt_{\max}$ by 420 ± 152 mm Hg/sec ($P < 0.05$, Fig. 4). Vagal cold block during epinephrine infusion did not produce any other cardiovascular effect than that which normally occurred during vagal cold block without epinephrine infusion. Figure 4 also demonstrates that the net inotropic effect of epinephrine with the vagi intact plus the effect of subsequent vagal cold block (855 ± 176 mm Hg/sec) results in contractile changes similar to those observed when the order of these maneuvers is reversed (855 ± 177 mm Hg/sec). The effects of atropine on constant intracoronary infusion of epinephrine (0.1 $\mu$g/kg per min) were examined in three dogs. In these animals, epinephrine infusion increased $dP/dt_{\max}$ from 2493 to 2874 mm Hg/sec before atropine and 2573 to 2879 mm Hg/sec after atropine.

**Ganglionic Blockade**

Ganglionic blockade was employed to examine the effects of intracoronary epinephrine without

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**Table 3 Changes Produced by Intracoronary Bolus Administration of Norepinephrine and Isoproterenol**

<table>
<thead>
<tr>
<th></th>
<th>Mean arterial pressure (mm Hg)</th>
<th>Heart rate (beats/min)</th>
<th>Mean left atrial pressure (mm Hg)</th>
<th>Left ventricular systolic pressure (mm Hg)</th>
<th>Left ventricular $dP/dt_{\max}$ (mm Hg/sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CON</td>
<td>BVB</td>
<td>CON</td>
<td>BVB</td>
<td>CON</td>
</tr>
<tr>
<td>Norepinephrine</td>
<td>0</td>
<td>0</td>
<td>±1</td>
<td>±1</td>
<td>±0.4</td>
</tr>
<tr>
<td>2.5 ng/kg</td>
<td>±1</td>
<td>±1</td>
<td>±2</td>
<td>±2</td>
<td>±0.3</td>
</tr>
<tr>
<td>Norepinephrine</td>
<td>0</td>
<td>0</td>
<td>±1</td>
<td>±1</td>
<td>±0.4</td>
</tr>
<tr>
<td>5.0 ng/kg</td>
<td>±1</td>
<td>±1</td>
<td>±2</td>
<td>±2</td>
<td>±0.3</td>
</tr>
<tr>
<td>Isoproterenol</td>
<td>0</td>
<td>0</td>
<td>±1</td>
<td>±1</td>
<td>±0.5</td>
</tr>
<tr>
<td>4.0 ng/kg</td>
<td>±1</td>
<td>±1</td>
<td>±0.5</td>
<td>±0.4</td>
<td>±3</td>
</tr>
</tbody>
</table>

Results are expressed as mean changes ± s.e. ($n = 5$ dogs) produced by intracoronary administration of norepinephrine and isoproterenol; B - C = differences in drug effects on changes in $dP/dt_{\max}$ between BVB and CON.

* $P < 0.05$; † $P < 0.01$. 

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TABLE 4 Constant Infusion of Epinephrine and Vagal Cold Block

<table>
<thead>
<tr>
<th></th>
<th>MAP (mm Hg)</th>
<th>HR (beats/min)</th>
<th>MLAP (mm Hg)</th>
<th>LVSP (mm Hg)</th>
<th>LVdP/dtmax (mm Hg/sec)</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>X ± AC ± AE ± V</td>
<td>X ± AC ± AE ± V</td>
<td>X ± AC ± AE + V</td>
<td>X ± AC ± AE + V</td>
<td>X ± AC ± AE + V</td>
</tr>
<tr>
<td>CON</td>
<td>97 ± 6</td>
<td>87 ± 6</td>
<td>50 ± 1.3</td>
<td>117 ± 6</td>
<td>3093 ± 260</td>
</tr>
<tr>
<td>EPI</td>
<td>98 ± 6</td>
<td>94 ± 6</td>
<td>56 ± 0.6</td>
<td>122 ± 6</td>
<td>3547 ± 454</td>
</tr>
<tr>
<td>EPI + BVB</td>
<td>115 ± 6*</td>
<td>158 ± 6*</td>
<td>71* ± 6*</td>
<td>136 ± 5*</td>
<td>3967 ± 420*</td>
</tr>
<tr>
<td></td>
<td>116 ± 6*</td>
<td>150 ± 6*</td>
<td>66* ± 6*</td>
<td>131 ± 5*</td>
<td>3921 ± 312*</td>
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<td></td>
<td>116 ± 6*</td>
<td>150 ± 6*</td>
<td>66* ± 6*</td>
<td>131 ± 5*</td>
<td>3944 ± 312*</td>
</tr>
</tbody>
</table>

Mean values (X) ± SE (n = 6 dogs) are shown during control states (CON); intra coronary epinephrine infusion (EPI), 0.1 μg/kg per min; epinephrine infusion and subsequent bilateral vagal cold block (EPI + BVB); and bilateral vagal cold block and subsequent epinephrine (BVB + EPI). AC = mean difference between control values and those observed during EPI, EPI + BVB, BVB, BVB + EPI. AE + V = mean difference between EPI and (EPI + BVB). AV + E = mean difference between BVB and (BVB + EPI).

• *P < 0.05; †P < 0.01.

FIGURE 4 Comparison of changes in LVdP/dtmax under five conditions: Epi = constant intracoronary infusion of epinephrine 0.1 μg/kg per min; BVB during Epi = bilateral vagal cold block initiated after the onset of Epi infusion; Epi + BVB during Epi = total inotropic change due to Epi infusion plus subsequent effects of BVB; BVB = bilateral vagal cold block; BVB + Epi = effects of Epi initiated during BVB. Mean ± se, n = 6.

Discussion

These experiments demonstrate that bilateral vagal cold block potentiates the inotropic effects of sympathomimetic amines when these agents are administered into the left circumflex coronary circulation of the conscious dog. The mechanism of cardiovascular reflex involvement. Ganglionic blockade was produced, using either trimethaphan or hexamethonium and atropine. Table 5 shows that ganglionic block produced a decrease in mean arterial pressure and an increase in heart rate while reducing dP/dt max. The reduction in dP/dt max by ganglionic blockade indicates that cardiac sympathetic nerves contribute to the contractile state of the heart under resting conditions. The contractile effects of epinephrine (0.02 μg/kg, 0.04 μg/kg, and 0.1 μg/kg per min) were potentiated significantly during ganglionic blockade. Epinephrine (0.04 μg/kg) significantly increased left ventricular systolic pressure, but the responses before and during ganglionic blockade were not different. Table 5 illustrates that these doses of epinephrine produced no significant effects on heart rate or mean arterial pressure either before or during ganglionic block.

Comparison of the Inotropic Potentiations Produced by Vagal Cold Block and Ganglionic Blockade

Because of the initial lower value of dP/dt max after ganglionic blockade, we compared the percent change in the inotropic potentiations produced by vagal cold block and ganglionic blockade. As shown in Figure 5, ganglionic blockade and vagal cold block produced similar degrees of inotropic potentiation to epinephrine.

These experiments demonstrate that bilateral vagal cold block potentiates the inotropic effects of sympathomimetic amines when these agents are administered into the left circumflex coronary circulation of the conscious dog. The mechanism of
TABLE 5  Changes Produced by Epinephrine before and during Ganglionic Blockade

<table>
<thead>
<tr>
<th></th>
<th>CON</th>
<th>GB</th>
<th>CON</th>
<th>GB</th>
<th>CON</th>
<th>GB</th>
<th>CON</th>
<th>GB</th>
<th>CON</th>
<th>GB</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean arterial pressure (mm Hg)</td>
<td>90 ±3</td>
<td>81 ±5</td>
<td>94 ±5</td>
<td>142 ±9</td>
<td>4 ±1.4</td>
<td>5 ±1.5</td>
<td>119 ±5</td>
<td>110 ±7</td>
<td>3025 ±203</td>
<td>2983 ±193</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>7 ±2</td>
<td>7 ±2</td>
<td>7 ±2</td>
<td>7 ±2</td>
<td>7 ±2</td>
<td>7 ±2</td>
<td>7 ±2</td>
<td>7 ±2</td>
<td>7 ±2</td>
<td>7 ±2</td>
</tr>
<tr>
<td>Mean left atrial pressure (mm Hg)</td>
<td>0 ±1</td>
<td>0 ±1</td>
<td>0 ±2</td>
<td>0 ±2</td>
<td>0 ±2</td>
<td>0 ±2</td>
<td>0 ±2</td>
<td>0 ±2</td>
<td>0 ±2</td>
<td>0 ±2</td>
</tr>
<tr>
<td>Left ventricular systolic pressure (mm Hg)</td>
<td>10 ±3</td>
<td>10 ±3</td>
<td>10 ±3</td>
<td>10 ±3</td>
<td>10 ±3</td>
<td>10 ±3</td>
<td>10 ±3</td>
<td>10 ±3</td>
<td>10 ±3</td>
<td>10 ±3</td>
</tr>
<tr>
<td>Left ventricular dP/dt max (mm Hg/sec)</td>
<td>512 ±50</td>
<td>512 ±50</td>
<td>688 ±68</td>
<td>688 ±68</td>
<td>664 ±68</td>
<td>664 ±68</td>
<td>907 ±213</td>
<td>907 ±213</td>
<td>123 ±43</td>
<td>123 ±43</td>
</tr>
</tbody>
</table>

Results are expressed as mean ± SE; GB = ganglionic blockade; GB changes produced by epinephrine during ganglionic blockade; Δ = differences in changes in dP/dt max produced by epinephrine between ganglionic blockade and control.

* P < 0.05; † P < 0.01; ‡ P < 0.001.

this potentiation apparently is associated with vagal afferent blockade, since parasympathetic efferent blockade with atropine did not potentiate the inotropic effects of epinephrine. Elimination of autonomic reflex effects with ganglionic blockade also augmented the inotropic actions of epinephrine similar to that observed with vagal cold block.

Interuption of the parasympathetic efferent component of the vagus conceivably could augment the contractile effects of intracoronary catecholamine administration through several actions. For instance, acetylcholine has been demonstrated to inhibit the inotropic actions of norepinephrine in vivo (Hollenberg et al., 1965; Ross, 1973). However, if vagal efferent block were responsible for the potentiation of the inotropic effects of catecholamines then atropine should have potentiated the contractile effects of the adrenergic stimulation. The fact that vagal cold block after atropine still potentiated the inotropic effects of intracoronary administration of epinephrine demonstrates that atropine does not inhibit the potentiation of epinephrine during BVB. Furthermore, the amount of potentiation observed with BVB without atropine and following atropine were similar, demonstrating that atropine did not alter the magnitude of the potentiation of the contractile response to intracor-
VAGAL AFFERENTS AND LEFT VENTRICULAR CONTRACTILITY/Barron & Bishop

Elevation in mean arterial pressure potentiates the inotropic effects of intracoronary injection of catecholamines, then the animals that had greater increases in mean blood pressure during vagal cold block would be expected to show greater inotropic potentiation than those animals that had minimal elevations in blood pressure during vagal cold block. However, we found no significant correlation between the increases in mean blood pressure due to vagal cold block and the amount of inotropic potentiation observed with vagal cold block. Furthermore, the independence of the dP/dt max response can be demonstrated by the fact that control vagal cold block produced almost identical changes in heart rate, mean left atrial pressure, and mean arterial pressure when compared to vagal cold block during constant intracoronary infusion of epinephrine; however, dP/dt max was increased significantly with vagal cold block only during constant intracoronary infusion of epinephrine.

The above discussion indicates that the potentiation of the positive inotropic effects of epinephrine during vagal cold block cannot be accounted for through alterations in the mechanical properties of the heart or through vagal efferent interruption. Elimination of these factors leaves only one alternative to account for the inotropic potentiations during vagal cold block, and that is interruption of vagal afferent nerve activity.

The inotropic effects of epinephrine could be potentiated during BVB by two reflex mechanisms. One possibility is through a vagal afferent-mediated negative feedback reflex in which the positive inotropic effects of epinephrine would activate cardiac vagal afferents, and this increase in vagal afferent activity would reduce sympathetic outflow to the heart. Previous studies have demonstrated that positive inotropic stimulation of the heart can increase vagal afferent nerve traffic (Sleight and Widdicombe, 1965; Muers and Sleight, 1972; Thorén, 1977), and that activation of vagal afferent nerves can elicit a decrease in cardiac sympathetic efferent nerve activity (Downing and Siegel, 1963; Muers and Sleight, 1972; Schwartz et al., 1973). The second possibility is that the positive inotropic effect of epinephrine activates cardiac sympathetic afferents, which via positive feedback effects could potentiate the contractile effects of epinephrine (Maliani et al., 1972). BVB could promote this reflex because tonic vagal afferent activity can mask sympathetic afferent reflexes (Shimizu et al., 1978). A recent study by Casati et al. (1979) has demonstrated that cardiac sympathetic afferent nerve activity can be increased with positive inotropic stimuli.

If a negative feedback mechanism reduces the inotropic effects of epinephrine, then interruption of the efferent arm of the reflex should result in potentiated contractile effects, whereas a sympathetic afferent reflex would require that sympathectic efferent activity to the heart be intact. To examine these possibilities we interrupted autonomic outflow with ganglionic blockade.

The results of this study demonstrate that the inotropic actions of epinephrine were augmented during ganglionic blockade. This observation is consistent with a negative feedback reflex, and the only cardiac reflex which could account for this negative feedback arc is a vagal afferent inhibition of sympathetic outflow to the heart. This concept is supported by the fact that the inotropic potentiation was similar with BVB and ganglionic block, which would be expected if the two interventions interrupted the same reflex arc. These data suggest that the inotropic effects of epinephrine can be attenuated by a negative feedback reflex and that the reflex is mediated afferently by the vagus and efferently by sympathetic withdrawal. However, the experiments with ganglionic block do not rule out that a sympathetic afferent mechanism might also participate in the potentiation of the inotropic effects of epinephrine during BVB.

Preliminary studies in our laboratory have shown that intracoronary veratridine elicits a reflex negative inotropic response which is reversed to a positive inotropic response during vagal cold block (Barron and Bishop, 1980). The positive inotropic response does not occur after either ganglionic blockade or β-adrenergic blockade. These results suggest that veratridine excites cardiac receptors with both vagal and sympathetic afferents but that the inhibitory influence of vagal afferents suppresses the excitatory reflex mediated by sympathetic afferents.

The increase in mean arterial pressure which occurred during vagal cold block is consistent with a previous study from this laboratory (Bishop and Peterson, 1978) and can be attributed to interruption of tonic vagal afferent inhibitory influences on sympathetic outflow (Oberg and White, 1970; Mancia et al., 1973; Thorén, 1977; Shimizu et al., 1979). Left ventricular dP/dt max was not altered during vagal cold block; however, a recent report from this laboratory has indicated that interruption of vagal afferents results in an increase in left ventricular contractility in anesthetized cats (Shimizu et al., 1979). Shimizu et al. (1979) proposed that the tonic restraining influence of vagal afferents was related to the contractile state of the myocardium such that an increase in cardiac sympathetic outflow would increase myocardial contractility and hence also increase vagal afferent restraining influences. In conscious dogs, in which the basal sympathetic tone is low, we did not observe an increase in LVP/dt max during vagal cold block. However, we found, when myocardial contractility was increased with constant infusion of epinephrine, that vagal cold block resulted in an increase in LVP/dt max. The tonic restraining influence of vagal cold block in the anesthetized open-chest cat can be attributed to a
greater level of sympathetic activation resulting from the anesthesia and the trauma of recent surgery (Vatner and Braunwald, 1975).

Intracoronary epinephrine did not produce a reflex depressor effect as might have been anticipated from the effects of veratridine. One possible explanation for this is that the vagal afferent stimulation by veratridine was much more intense than that resulting from the inotropic effects of the limited doses of catecholamines. Furthermore, the possibility that epinephrine produces reflex effects on myocardial contractility does not necessarily dictate that other reflex effects must occur. Previous studies have demonstrated that cardiac reflexes can be quite specific in regard to effector site (Furnival et al., 1971; Randall et al., 1972). It also is possible that nonuniform changes in vascular resistance could occur in different vascular beds in the absence of changes in mean arterial pressure (Abboud et al., 1976).

If vagal afferents originating from the heart are biased to affect sympathetic outflow to the heart, then this may partially explain why no depressor effects were observed with inotropic stimulation. Many previous reports have demonstrated that ventricular distention results in peripheral vasodilation due to reductions in sympathetic vasoconstrictor tone (Aviado and Schmidt, 1959; Salisbury et al., 1960; Rossi et al., 1961; Mark et al., 1973; Chevalier et al., 1974; Zelis et al., 1979). Fox et al. (1977), using anesthetized dogs, have reported that positive inotropic stimulation of the left ventricle produces vagal afferent-mediated falls in peripheral resistance. The inotropic stimulations employed in our study with epinephrine resulted in 9% to 32% increases in dP/dt\text{max}, whereas Fox et al. (1977) used doses that increased dP/dt\text{max} 9% to 700% (with a mean of 193%). If the degree of reflex vasodilation is roughly proportional to the degree of inotropic stimulation, as proposed by Fox et al. (1977), then the inotropic stimuli used in our study may have been too low to produce observable effects on mean arterial pressure.

In summary, elimination of vagal afferent but not vagal efferent nerve activity potentiates the inotropic effects of intracoronary catecholamines. The potentiation occurring during vagal cold block could conceivably result from the added contractile effects of a sympathetic afferent positive feedback reflex and/or disinhibition of a negative inotropic effect mediated by vagal afferent restraint on cardiac sympathetic outflow. The potentiation of the contractile effects of epinephrine during ganglionic block supports the concept that a negative feedback reflex attenuates the effects of epinephrine through withdrawal of cardiac sympathetic efferent activity. Thus, this study offers experimental evidence in the conscious dog for the existence of a cardiocardiac, vago-sympathetic, negative feedback reflex, which can be initiated by an increase in myocardial contractility. This reflex would offer a mechanism for the autonomic nervous system to sense and modulate the contractile state of the ventricular myocardium. This evidence supports previous work by Shimizu et al. (1979) and previous proposals by Fox et al. (1977) and Estrin et al. (1979).

Acknowledgments

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