Disparate Effects of Substance P on Systemic and Coronary Beds in Conscious Dogs

Yukio Nakamura, MD; Robert Parent, PhD; and Michel Lavallée, PhD

Background. Previous studies in anesthetized animals indicated that substance P is a coronary and peripheral vasodilator. However, coronary vasodilation was only transient perhaps because of tachyphylaxis. In the present study, the steady-state effects of intravenous substance P on systemic and coronary beds were investigated in conscious, instrumented dogs.

Methods and Results. With intact autonomic reflexes, 5 ng/kg/min i.v. substance P resulted in increases \((p<0.01)\) in cardiac output by 22±5\%, in decreases \((p<0.01)\) in mean arterial pressure by 9±2\%, and in total peripheral resistance by 23±4\% 7–9 minutes after the beginning of substance P infusion. Heart rate increased \((p<0.01)\) by 35±7\% and left ventricular dP/dt \((p<0.05)\) by 13±4\%. In this situation, coronary blood flow decreased \((p<0.01)\) by 19±4\% and coronary vascular resistance increased \((p<0.05)\) by 13±5\%. Myocardial oxygen delivery was reduced \((p<0.05)\) by 13±5\% and the arteriovenous oxygen difference widened \((p<0.01)\). After ganglionic blockade, increases in cardiac output, heart rate, and left ventricular dP/dt with substance P administration were abolished, but total peripheral resistance and mean arterial pressure decreased \((p<0.01)\) by 12±3\% and 11±3\%, respectively. Under these conditions, coronary blood flow decreased \((p<0.01)\) by 37±5\% and coronary vascular resistance increased \((p<0.01)\) by 47±8\%, which were more \((p<0.01)\) than control responses. In this situation, myocardial oxygen delivery was reduced \((p<0.01)\) by 37±4\% and the arteriovenous oxygen difference widened \((p<0.01)\). Intracoronary infusion of substance P (0.4 ng/kg/min) resulted in significant and sustained decreases in coronary blood flow, which were similar before and after ganglionic blockade.

Conclusions. Thus, in conscious dogs, systemic vasodilation is the prevailing effect of substance P, but paradoxically, this peptide simultaneously elicits coronary vasoconstriction. (Circulation 1991;84:300–312)

Since the discovery of substance P by von Euler and Gaddum,1 this peptide has been demonstrated to be widely distributed in the walls of vessels subserving numerous organs,2,3 including human and canine coronary vessels.4–5 Whether substance P acts locally as a neurotransmitter and, thereby, intervenes in cardiovascular regulation remains to be determined. Substance P may also influence cardiovascular function as a hormone because circulating levels of substance P can reach physiological threshold.5–8

Prior studies in anesthetized animals indicated that substance P is a potent coronary and peripheral vasodilator.9 Coronary vasodilator responses are generally transient presumably because of tachyphylaxis.8–11 but other vascular beds may show sustained vasodepressor responses.12 In isolated vessels, substance P causes vascular relaxation primarily through an endothelium-dependent phenomenon.13–15 The possibility that endogenous peptides act as vasodilators has prompted clinical studies in which systemic and coronary dilations were reported with substance P.16,17 In a recent study18 conducted in conscious dogs with ganglionic blockade, we found that substance P paradoxically constricted resistance coronary vessels and dilated epicardial coronary arteries. In the present study, we considered that substance P may act as a peripheral vasodilator and simultaneously as a coronary vasoconstrictor under steady-state conditions. The mechanisms involved in peripheral and coronary responses to substance P were investigated. In addition, we investigated the effects of intracoronary administration of substance P before and after ganglionic blockade and considered that cardiac denervation may alter the coronary response to substance P. This study was conducted in conscious,
instrumented dogs to avoid the confounding influences of general anesthesia and recent surgery on cardiovascular responses.\textsuperscript{19}

**Methods**

**Instrumentation**

Nine mongrel dogs (weight, 25±1 kg) were anesthetized with sodium pentobarbital (30 mg/kg i.v.) and ventilated artificially. Under sterile conditions, a left thoracotomy was performed at the fifth intercostal space, and a medical grade Tygon catheter was implanted in the thoracic aorta to measure aortic pressure with a pressure transducer (model 800, Bentley Trantec, Irvine, Calif.). Mean aortic pressure (MAP) was obtained with an active filter with a time constant of 2 seconds. A catheter was implanted in the left atrium for the injection of radioactive microspheres. Another catheter that was made of Silastic was implanted in the coronary sinus to obtain blood samples for measuring hemoglobin concentration and oxygen saturation with a cooximeter (OSM-2, Hemoxymeter, Radiometer America, Inc., Westlake, Ohio). At necropsy, the position of the catheter within the coronary sinus was confirmed, and the tip was at least 10 mm from the ostium. A solid-state pressure gauge (P.6.5., Konigsberg Instruments, Inc., Pasadena, Calif.) and a catheter to cross-calibrate the pressure transducer were implanted in the left ventricular (LV) cavity through an apical stab wound. This transducer was used to measure LV systolic pressure (LVP), to measure LV end-diastolic pressure (LVEDP), and to obtain the first derivative of LVP, LV dp/dt. Electrodes were sutured to the right ventricular (RV) outflow tract to perform ventricular pacing. A cardiostachometer (Sensor Medics, Anaheim, Calif.) triggered by the LV pressure pulse was used to monitor heart rate (HR). An ultrasonic Doppler blood flow transducer was implanted around the left circumflex coronary artery, 2–3 cm from the bifurcation of the left main coronary artery. Coronary blood flow (CBF) was monitored with a 10-MHz pulsed Doppler flowmeter.\textsuperscript{20} Mean CBF was obtained with an active filter with a time constant of 2 seconds. The linearity of the relation between flow velocity and Doppler shift was previously demonstrated for this instrument and was confirmed in our laboratory by in vitro testing. At necropsy, the internal circumference of the vessel under the probe was measured to obtain the vessel cross-sectional area and to calculate a calibration factor in milliliters per minute per kilohertz. An ultrasonic flow probe (model 4s, Transonic System, Inc., Ithaca, N.Y.) was implanted around the ascending aorta to measure phasic aortic blood flow with a flowmeter (model 120, Transonic). This instrument monitors the phase shift of a wide beam of ultrasound between upstream and downstream transducers. The linearity and accuracy of this instrument were confirmed in our laboratory by performing bench calibrations with saline before and after implantation. Mean aortic blood flow was obtained with an active filter with a time constant of 8 seconds. All hemodynamic signals were recorded on an eight-channel tape recorder (model 3968A, Hewlett-Packard, Palo Alto, Calif.) and were played back on a direct ink-writing stripchart recorder (model 2800s, Gould-Statham, Cleveland, Ohio).

In six additional dogs (weight, 30±1 kg) that were instrumented with solid-state pressure gauges, aortic and LV catheters, and Doppler flow probes around the circumflex coronary artery, a Silastic catheter was implanted proximal to the flow probe in the lumen of the circumflex coronary artery (1–2 cm from the bifurcation of the left main coronary artery) by the approach of Gwirtz.\textsuperscript{21} The portion of the catheter within the coronary vessel had an external diameter of 0.6 mm.

Five additional dogs (weight, 24±1 kg) were instrumented after a cardiac denervation was performed with the technique of Randall et al. The intrapericardial cardiac denervation consists of 1) sectioning of the ventrolateral cardiac nerve, 2) advenitial stripping of the left pulmonary veins and the right and common pulmonary arteries, and 3) sectioning of the pericardial reflections in the transverse sinus and around the superior vena cava and ligating and sectioning of the azygos vein. In addition, anterior and posterior ansae subclaviae were bilaterally transected in the extrapericardium. Adequacy of cardiac denervation was confirmed in conscious animals by demonstrating the failure of HR and LV dp/dt to change after boluses of pressor doses of nitroglycerin (15 μg/kg i.v.) and phenylephrine (3 μg/kg i.v.). Absence of decreases in HR and MAP after administration of veratrine (8 μg/kg i.a.) confirmed elimination of afferent cardiac nerves.

**Experimental Protocols**

Experiments were initiated 2–4 weeks after surgery in conscious dogs that were trained to lie quietly on a table in a dimly illuminated laboratory. While continuously monitoring HR, LVP, LVEDP, LV dp/dt, MAP, mean CBF, and cardiac output (CO), we administered intravenous infusions of substance P (1-11, Institut Armand Frappier, Laval, Canada) in nine dogs. Substance P was freshly dissolved in saline before each experiment, and doses of 5 and 10 ng/kg/min i.v. were administered during a 9-minute period. At least 15 minutes was allowed between two infusions for the return to a steady-state baseline. The basic protocol performed in nine dogs consisted of the administration of 5 ng/kg/min substance P under baseline conditions, during RV pacing, and after ganglionic blockade with 35 mg/kg hexamethonium bromide (Sigma Chemical Co., St. Louis, Mo.) and 0.1 mg/kg atropine methyl bromide (Sigma). Adequacy of autonomic blockade was confirmed by the absence of reflex changes in HR and LV dp/dt to boluses of 10.0 μg/kg i.v. nitroglycerin and 3.0 μg/kg i.v. phenylephrine. Two to 3 days later, 10 ng/kg/min substance P was administered in six dogs before and after ganglionic blockade. The above protocols in-
volved the sampling of coronary sinus and aortic blood for the determination of hemoglobin content and oxygen saturation before the infusion of substance P and 7–9 minutes after the beginning of the infusion, when a steady state was reached. Blood samples were processed within 30 seconds after their collection.

In six of these dogs, 2–3 million radioactive microspheres labeled with one of the four available isotopes niobium-95, strontium-85, cerium-141, and scandium-46 were injected after ganglionic blockade, under baseline conditions, and during steady state after the administration of substance P (5 ng/kg/min) for the determination of regional blood flow distribution. Microspheres suspended in 10% dextran in saline containing 0.01% Tween 80 were ultrasonically and mechanically agitated before injection to ensure adequate dispersion that was confirmed by microscopic examination. While continuously monitoring hemodynamic variables, we injected 1 ml of the suspending medium (without microspheres) to confirm absence of any adverse effect on cardiovascular function. Starting 15 seconds before an injection of microspheres, a reference sample was drawn continuously from the aorta at a rate of 7.76 ml/min for 150 seconds. Microspheres were injected into the left atrial catheter and flushed with saline. The same procedure was repeated during the administration of substance P. After the animals were killed with an overdose of barbiturates followed by potassium chloride, the heart and samples from liver, spleen, ileum, adrenal gland, kidney, skeletal muscle, descending colon, stomach, and abdominal skin were obtained and placed in 2% formaldehyde for 48–72 hours. A ventricular ring 1–1.5 cm in thickness was cut at mid-distance between the base and apex, and transmural samples were obtained from the RV and the anterior and posterior walls of the LV. Each LV sample was divided into four layers from the endocardium to the epicardium. Only transmural samples were obtained from the RV. Kidney samples were divided into three layers corresponding to cortical, medullary, and papillary layers. All tissue samples were trimmed from adipose tissue, weighed, and placed in counting vials for the determination of radioactivity with a gamma counter (Kontron Instruments, Milan, Italy). Raw counts were corrected for cross-over, and regional blood flow in milliliters per minute was obtained for each sample with the following equation: 

\[ Q_m = \frac{(Q_r \times C_m)}{C_r} \]

where \( Q_m \) is regional blood flow, \( Q_r \) is withdrawal rate of the reference arterial blood (ml/min), \( C_m \) is counts per minute in the tissue sample, and \( C_r \) is counts per minute in the reference arterial blood. Regional blood flow was corrected for the weight of the samples and was expressed as milliliters per minute per gram of tissue.

In the six additional dogs instrumented with intracoronary catheters, substance P was directly administered into the circumflex coronary artery. Boluses of substance P (0.01 and 0.1 ng/kg in 0.2 ml saline at 38.9°C) were injected into the coronary artery catheter and then flushed with 1.5 ml warm saline delivered with an infusion pump during a 12-second period. Before any substance P administration, boluses of saline without the drug were injected to verify absence of changes in CBF. Infusions of 0.2 and 0.4 ng/kg/min i.c. substance P were also made during a 9-minute period. While monitoring hemodynamic variables under a continuous infusion of warm saline at a constant rate of 0.38 ml/min and when a steady-state baseline was obtained, we then substituted the substance P solution for the saline. An infusion of 2 ng/kg/min i.v. substance P was also administered in the same animals. The sequence of administration of boluses and infusions of substance P was randomly determined, and at least 15 minutes was allowed between two injections for the return to a steady-state baseline. The intracoronary and intravenous infusions of substance P were repeated after ganglionic blockade with 35 mg/kg hexamethonium bromide and 0.1 mg/kg methyl atropine. Adequacy of autonomic blockade was verified at the completion of each experiment.

In five dogs with cardiac denervation, 2 and 5 ng/kg/min i.v. substance P was infused. Arterial blood samples were obtained before and 7–9 minutes after the beginning of substance P administration for the determination of hemoglobin concentration and hematocrit level.

**Data Analysis**

Data were read from the stripcharts under baseline conditions, during peak early responses to substance P, and under steady-state hemodynamic conditions for experiments involving intravenous administration of substance P. Responses to intracoronary boluses of substance P are reported under baseline conditions and at peak increases in CBF. Hemodynamic responses to intracoronary infusions of substance P are reported under baseline conditions, at peak increases in CBF, and every minute throughout the infusion period. Coronary vascular resistance (CVR) was calculated with the ratio of MAP to CBF measured with the Doppler flowmeter. Total peripheral resistance (TPR) was obtained by dividing MAP by CO. Blood oxygen content was calculated with the following equation: hemoglobin concentration multiplied by percent \( O_2 \) saturation multiplied by hemoglobin binding coefficient (1.34). Myocardial oxygen delivery to the circumflex bed in milliliters per minute was determined as follows: aortic blood oxygen content multiplied by circumflex CBF.

The myocardial arteriovenous oxygen difference was obtained by subtracting coronary sinus blood oxygen content from aortic blood oxygen content. We assumed in these calculations that changes in CBF in the circumflex bed were representative of overall LV responses. The pressure-work index was used to estimate cardiac metabolic demand and was calculated with the following equation:

\[ P\cdot W = K_1(SBP\times HR) + K_2 \]
Clarity in briefly described, within peaking variance and the conditions thoroughly described.

Figure 1. Recording of left ventricular pressure (LVP), the first derivative of left ventricular pressure over time (LV dP/dt), arterial blood pressure (AP), phasic and mean cardiac output (CO), phasic and mean coronary blood flow (CBF), and heart rate (HR) before and during administration of 5 ng/kg/min i.v. substance P beginning at the arrow. Steady-state effects of substance P were characterized by a decrease in mean arterial pressure and a rise in cardiac output, heart rate, and LV dP/dt concomitant with a decrease in coronary blood flow.

\[
[(0.8SBP+0.2 DBP) \times HR \times SV]/BW + 1.43, \text{ where P-W is pressure-work index (in ml O}_2/\text{min/100 g), SBP is systolic blood pressure (in mm Hg), DBP is diastolic blood pressure (in mm Hg), HR is heart rate (beats/}
\min, SV is stroke volume (ml), BW is body weight (kg), K1 is 4.08 \times 10^{-4}, \text{ and K2 is 3.25} \times 10^{-4}.
\]

Values are reported as mean \pm SEM. Multiple comparisons within groups were made with analysis of variance and the Bonferroni’s correction. \text{Statistical significance was considered at a } p \text{ value less than 0.05.}

All experimental procedures were performed in accordance with the guidelines of the Canadian Council on Animal Care.

**Results**

Infusion of 5 and 10 ng/kg/min i.v. substance P for 9 minutes resulted in transient hemodynamic effects peaking within 60 seconds after the beginning of the infusion (Figure 1). These initial changes in hemodynamic variables are termed “early responses.” Stable hemodynamic conditions were reached 7–9 minutes after the beginning of substance P administration and are termed “steady-state responses.” For reasons of clarity in the data presentation and because steady-state responses are the focus of the present study, early responses to 5 ng/kg/min i.v. substance P are briefly described, and the data are reported in Table 1. Responses to 5 ng/kg/min i.v. substance P are thoroughly described under baseline conditions, during RV pacing, and after ganglionic blockade. Only steady-state coronary responses to 10 ng/kg/min i.v. substance P are reported (Table 3).

**Early Responses**

Peak early responses to 5 ng/kg/min i.v. substance P before and after ganglionic blockade are reported in Table 1. Early responses to substance P were characterized by increases in HR, LV dP/dt, CBF, and CO and by significant decreases in MAP, LVP, LVEDP, CVR, and TPR.

After ganglionic blockade, HR did not change from baseline, and decreases in MAP were greater than those before autonomic blockade. There was a tendency for greater decreases in LVP, but statistical significance was not reached. After ganglionic blockade, decreases in LVEDP were smaller than those before ganglionic blockade. Increases in LV dP/dt were blunted by ganglionic blockade. After ganglionic blockade, increases in CO were smaller than those before ganglionic blockade, but decreases in TPR did not differ. The amplitude of increases in CBF and of decreases in CVR was similar before and after ganglionic blockade.

**Steady-State Responses**

**Hemodynamic effects.** Under steady-state conditions, 7–9 minutes after beginning the infusion of 5 ng/kg/min i.v. substance P, MAP decreased; CO increased (22\%5\%), and TPR decreased (23\%4\%) from baseline values (Table 2, Figure 1). LVP did not change from baseline values; LVEDP decreased, and HR and LV dP/dt increased.

RV pacing prevented the increase in CO with intravenous administration of substance P, but decreases in
TABLE 1. Early Responses to Intravenous Administration of Substance P (5 ng/kg/min) Before and After Ganglionic Blockade

<table>
<thead>
<tr>
<th>Substance P (5 ng/kg/min i.v.)</th>
<th>Baseline</th>
<th>Δ From baseline</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>LV systolic pressure (mm Hg)</strong></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
| Control | 117±4 | −13±3*
| Ganglionic blockade | 92±3† | −20±1*
| **LV end-diastolic pressure (mm Hg)** | | |
| Control | 8.9±1.0 | −6.5±0.8*
| Ganglionic blockade | 2.5±0.8† | −2.2±0.6†‡
| **LV dP/dt (mm Hg/sec)** | | |
| Control | 2,823±201 | +858±150*
| Ganglionic blockade | 1,977±179† | −227±37*†
| **Mean arterial pressure (mm Hg)** | | |
| Control | 92±3 | −21±3*
| Ganglionic blockade | 73±2† | −33±2*§
| **Coronary blood flow (ml/min)** | | |
| Control | 29.2±1.8 | +12.3±2.1*
| Ganglionic blockade | 29.3±2.4 | +11.7±2.2*
| **Coronary vascular resistance (mm Hg/ml/min)** | | |
| Control | 3.30±0.28 | −1.44±0.23*
| Ganglionic blockade | 2.58±0.20† | −1.08±0.11*|
| **Cardiac output (l/min)** | | |
| Control | 2.3±0.3 | +1.2±0.1*
| Ganglionic blockade | 2.4±0.3 | +0.3±0.1†‡
| **Total peripheral resistance (mm Hg/l/min)** | | |
| Control | 44.7±4.8 | −20.6±2.5*
| Ganglionic blockade | 33.3±4.3† | −15.2±2.6*
| **Heart rate (beats/min)** | | |
| Control | 90±4 | +75±6*
| Ganglionic blockade | 128±6† | +1±1†

Values are mean±SEM.
LV, left ventricular.
*p<0.01 vs. baseline; †p<0.01 vs. control; ‡p<0.05 vs. baseline; §p<0.05 vs. control.

TPR persisted (17±3%), similar to control responses on a percent basis. MAP, LVP, and LVEDP all decreased, and LV dP/dt increased.

Ganglionic blockade prevented the rise in CO with intravenous infusion of substance P and attenuated (p<0.05) the decrease in TPR (12±3%). MAP, LVP, and LVEDP decreased, but increases in HR and LV dP/dt were abolished.

Coronary effects. With administration of 5 ng/kg/min i.v. substance P before ganglionic blockade and under steady-state conditions, CBF decreased (19±4%), and CVR increased (13±5%). In this situation, myocardial oxygen delivery to the circumflex bed decreased (p<0.05) by 13±5% from 4.2±0.2 ml/min in the face of an increase by 22±6% in the pressure-work index (Table 3, Figure 2). Aortic and coronary sinus blood oxygen content increased as a result of a rise in hemoglobin concentration, but the myocardial arteriovenous oxygen difference widened.

Under RV pacing, CBF decreased (30±5%), and CVR increased (30±9%) with intravenous administration of substance P. In this situation, myocardial oxygen delivery to the circumflex bed decreased (p<0.01) disproportionately more (24±6% from 4.8±0.3 ml/min) than did the pressure-work index (7±3%). Aortic and coronary sinus blood oxygen
TABLE 2. Steady-State Effects of Intravenous Administration of Substance P (5 ng/kg/min) on Left Ventricular Function and Systemic Hemodynamics

<table>
<thead>
<tr>
<th>Substance (P 5 ng/kg/min i.v.)</th>
<th>Baseline</th>
<th>Δ From baseline</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>LV systolic pressure (mm Hg)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>117±4</td>
<td>-3±2</td>
</tr>
<tr>
<td>Pacing</td>
<td>112±2</td>
<td>-7±2*</td>
</tr>
<tr>
<td>Ganglionic blockade</td>
<td>92±3†</td>
<td>-6±2‡</td>
</tr>
<tr>
<td><strong>LV end-diastolic pressure (mm Hg)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>8.9±1.0</td>
<td>-4.2±0.6*</td>
</tr>
<tr>
<td>Pacing</td>
<td>3.9±0.9†</td>
<td>-0.9±0.1†</td>
</tr>
<tr>
<td>Ganglionic blockade</td>
<td>2.5±0.8†</td>
<td>-1.0±0.3*†</td>
</tr>
<tr>
<td><strong>LV dP/dt (mm Hg/sec)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>2,823±201</td>
<td>+342±90*</td>
</tr>
<tr>
<td>Pacing</td>
<td>1,929±90†</td>
<td>+184±51*</td>
</tr>
<tr>
<td>Ganglionic blockade</td>
<td>1,977±179†</td>
<td>-76±29†‡</td>
</tr>
<tr>
<td><strong>Mean arterial pressure (mm Hg)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>92±3</td>
<td>-8±2*</td>
</tr>
<tr>
<td>Pacing</td>
<td>97±3</td>
<td>-12±2*</td>
</tr>
<tr>
<td>Ganglionic blockade</td>
<td>73±2†</td>
<td>-9±2*</td>
</tr>
<tr>
<td><strong>Cardiac output (l/min)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>2.3±0.3</td>
<td>+0.5±0.1*</td>
</tr>
<tr>
<td>Pacing</td>
<td>2.5±0.4</td>
<td>+0.2±0.1§</td>
</tr>
<tr>
<td>Ganglionic blockade</td>
<td>2.4±0.3</td>
<td>0.0±0.0†</td>
</tr>
<tr>
<td><strong>Total peripheral resistance (mm Hg/l/min)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>44.7±4.8</td>
<td>-10.7±2.1*</td>
</tr>
<tr>
<td>Pacing</td>
<td>42.7±4.5</td>
<td>-6.6±1.0*§</td>
</tr>
<tr>
<td>Ganglionic blockade</td>
<td>33.3±4.3†</td>
<td>-3.9±1.0*†</td>
</tr>
<tr>
<td><strong>Heart rate (beats/min)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>90±4</td>
<td>+32±7*</td>
</tr>
<tr>
<td>Pacing</td>
<td>157±3†</td>
<td>0±0†</td>
</tr>
<tr>
<td>Ganglionic blockade</td>
<td>128±6†</td>
<td>+3±3†</td>
</tr>
</tbody>
</table>

Values are mean±SEM.
LV, left ventricular.
*p<0.01 vs. baseline; †p<0.01 vs. control; §p<0.05 vs. baseline; ¶p<0.05 vs. control.

content increased as a result of the rise in hemoglobin concentration, but the myocardial arteriovenous oxygen difference widened.

After ganglionic blockade, CBF decreased (37±5%), and CVR increased (47±8%) during substance P infusion. The pressure-work index decreased slightly by 4±1%, but myocardial oxygen delivery to the circumflex bed declined disproportionately more, by 37±4% from 4.0±0.3 ml/min. Hemoglobin concentration and aortic blood oxygen content did not change from baseline values during intravenous infusion of substance P. In this situation, coronary sinus oxygen content decreased, and the myocardial arteriovenous oxygen difference widened. Regional blood flow. Peripheral blood flow distribution was measured with microspheres injected under baseline conditions and during intravenous infusions of substance P after ganglionic blockade. With administration of 5 ng/kg/min i.v. of substance P, MAP decreased by 11±3 from 85±3 mm Hg, and TPR decreased by 16±3% from 32.7±2.4 mm Hg/l/min. Transmural myocardial blood flow of the LV decreased by 23±2%, and endocardial to epicardial blood flow ratio decreased slightly to 1.28±0.05 from 1.35±0.06 (Figure 3). Regional ventricular perfusion under baseline conditions and changes with substance P were similar at the various sites of measurements within the LV.
RV transmural blood flow decreased by 17±2% from 0.79±0.09 ml/min/g during steady-state effects of substance P.

Except for the skin and spleen where regional perfusion decreased during steady-state responses to intravenous infusion of substance P, all other vascular beds showed either no change or increases in regional blood flow (Table 4).

Intracoronary Substance P

In six additional dogs, boluses of 0.01 and 0.1 ng/kg i.c. substance P resulted in peak increases of CBF by 96±8% and 156±8%, respectively, without any other hemodynamic effect (Table 5). The effects of infusions of 0.2 and 0.4 ng/kg/min i.c. substance P are reported in Figure 4. With administration of 0.4 ng/kg/min i.c. substance P for 9 minutes, CBF transiently increased by 69±12% from 30.8±3.6 ml/min within the first minute of the infusion. CBF decreased to 20±8% under baseline values at 3 minutes after the beginning of the intracoronary infusion of substance P. At 9 minutes, CBF remained reduced and was 24±8% under baseline levels. Baseline MAP (90±3 mm Hg), LV dP/dt (2.973±140 mm Hg/sec), and HR (72±2 beats/min) did not change throughout the intracoronary infusion of substance P. Ganglionic blockade did not prevent the early increase in CBF (63±10%, from 35.2±4.2 ml/min), and it did not prevent the sustained decrease in CBF under baseline levels at 3 (29±7%) and 9 (28±7%) minutes after the beginning of the intracoronary infusion of substance P. Under these conditions, baseline LV dP/dt (2.263±222 mm Hg/sec) and HR (120±5 beats/min) remained stable throughout the infusion period. Except for a transient decrease by 10±2 from 86±3 mm Hg at 2 minutes within the infusion period, MAP did not differ from baseline values. Administration of 0.2 ng/kg/min i.c. substance P resulted in a similar pattern of CBF responses without any significant change in HR, MAP, and LV dP/dt.

In the same animals, administration of 2 ng/kg/min i.v. substance P for 9 minutes resulted in steady-state decreases (p<0.01) in CBF by 25±6% from 27.8±2.2 ml/min and increases (p<0.05) in CVR by 26±8% from 3.37±0.28 mm Hg/ml/min. At that time, MAP and LVEDP had decreased (p<0.05) by 7±2 from 91±2 mm Hg and by 2.6±0.7 from 11.1±2.3 mm Hg, respectively. LVP and LV dP/dt did not significantly differ from baseline (112±2 mm Hg and 3.096±113 mm Hg/sec, respectively). HR increased (p<0.01) by 15±3 from 80±4 beats/min. With ganglionic blockade in five of these animals, CBF decreased (p<0.01) by 36±8% from 27.9±2.0 ml/min, and CVR increased (p<0.05) by 48±17% from 3.03±0.28 mm Hg/ml/min. MAP and LVP decreased (p<0.05) by 8±2 from 82±4 mm Hg and by 5±2 from 98±4 mm Hg, respectively. Baseline LVEDP (3.4±0.8 mm Hg), LV dP/dt (2.24±154 mm Hg/sec), and HR (117±7 beats/min) did not change during intravenous administration of substance P.

Intravenous Substance P in Cardiac-Denervated Dogs

Hemodynamic and coronary responses to intravenous administration of substance P in dogs with cardiac denervation are reported in Table 6. Infusion of 2 ng/kg/min i.v. substance P resulted in a steady-state decrease in CBF by 20±4%. CVR increased by 22±4%, 7–9 minutes after the beginning of the infusion. This was the only statistically significant effect of 2 ng/kg/min i.v. substance P on hemodynamic variables in cardiac-denervated dogs. With infusion of 5 ng/kg/min i.v. substance P, CBF decreased by 32±3%, and CVR increased by 31±7% under steady-state conditions. At that time, LVP and MAP decreased, and LVEDP, LV dP/dt, and HR did not significantly differ from baseline values. Hematocrit and hemoglobin levels increased significantly with infusion of 5 ng/kg/min i.v. substance P.

Discussion

The present study indicates that substance P has disparate effects on systemic and coronary hemodynamics. With and without autonomic blockade and under steady-state conditions, intravenous adminis-
Prior studies indicated that substance P dilates both peripheral and coronary beds. In the study by Maxwell,11 boluses of substance P resulted in transient increases in CBF and decreases in peripheral resistance as we noted early within the infusion period in the present study. However, the study by Maxwell did not examine steady-state effects of substance P, which differed in coronary and systemic beds. In a recent study conducted in humans, intracoronary administration of substance P induced coronary vasodilation as suggested by increases in coronary sinus oxygen saturation.16 This observation is consistent with studies conducted in anesthetized dogs with intracoronary administration of substance P.4,15 However a steady state may not have been reached in these studies, which could explain why
coronary vasoconstrictor responses were not found. Yeo et al.8 examined the effects of prolonged infusions of substance P in conscious dogs. With respect to changes in peripheral resistance, our data are consistent with the finding of systemic vasodilation with substance P. Coronary responses in the present study differed from those reported by Yeo et al. Substance P failed to alter coronary perfusion in the study by Yeo et al, whereas sustained decreases in CBF followed transient vasodilator responses in the present study. Differences in the site of microsphere injection, in the withdrawal time of reference blood samples, in the duration of substance P infusions, and in the period of recovery after surgery may explain why the conclusions reached in the study by Yeo et al. differed from ours regarding CBF.

Our selection of doses and route of administration of substance P was guided by earlier reports showing that infusions of 7 ng/kg/min i.v. substance P result in elevations of circulating substance P levels similar to those achieved in the postprandial state.6,9 Therefore, we administered substance P at 2, 5, and 10 ng/kg/min i.v., which should approximate the physiological range. We also examined the effects of intracoronary administration of substance P in doses that lacked significant peripheral hemodynamic influence, in a dose range close to the levels used in an earlier study in humans.10

Decreases in TPR under control conditions could be the combined result of direct and reflex effects of substance P on peripheral vessels. However, decreases in TPR with substance P were smaller after ganglionic blockade than before. This suggests that reflex mechanisms did not antagonize the vasodilator effects of substance P. A lower baseline TPR after ganglionic blockade may explain why substance P was apparently less potent in this situation. Similar absolute levels of peripheral resistance were reached before and after ganglionic blockade during substance P administration. Consequently, reflex influences did not alter to a significant extent the level of peripheral resistance achieved during substance P administration.

In the present study, the mechanism of changes in CO with substance P was considered. Under control conditions, the rise in CO was concomitant with an increase in HR, a decrease in LVP, and a modest rise in LV dP/dt. Increases in CO were abolished with ventricular pacing, whereas changes in LVP and in LV dP/dt did not differ from control responses. Thus, increases in HR were primarily responsible for the rise in CO.

Consistent with earlier studies, substance P lacked direct chronotropic effects on the heart.9 Increases in HR under baseline conditions involved a reflex mechanism that was suppressed with ganglionic blockade or with cardiac denervation. Increases in LV dP/dt under control conditions were also of reflex origin because ganglionic blockade or cardiac denervation abolished these responses. Thus, under the present experimental conditions, substance P lacked significant direct chronotrophic and inotrophic effects.

In contrast to the peripheral vasodilation caused by substance P, the coronary bed showed biphasic responses; decreases in CBF and increases in CVR followed a transient coronary vasodilation during prolonged administration of substance P.

The steady-state decrease in CBF and the increase in CVR could be secondary to a sympathetically mediated reflex triggered by the decrease in arterial

---

**TABLE 4. Regional Blood Flow Distribution During Intravenous Administration of Substance P (5 ng/kg/min) After Ganglionic Blockade**

<table>
<thead>
<tr>
<th>REGION</th>
<th>Baseline P (5 ng/kg/min i.v.)</th>
<th>Steady state P (5 ng/kg/min i.v.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Liver</td>
<td>0.20±0.05</td>
<td>0.43±0.06*</td>
</tr>
<tr>
<td>Spleen</td>
<td>1.99±0.39</td>
<td>1.27±0.19†</td>
</tr>
<tr>
<td>Kidney</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cortex</td>
<td>5.02±0.42</td>
<td>6.18±0.57</td>
</tr>
<tr>
<td>Medulla</td>
<td>2.78±0.23</td>
<td>2.98±0.25</td>
</tr>
<tr>
<td>Papilla</td>
<td>0.34±0.11</td>
<td>0.44±0.12†</td>
</tr>
<tr>
<td>Adrenal</td>
<td>1.31±0.19</td>
<td>3.45±0.47*</td>
</tr>
<tr>
<td>Stomach</td>
<td>0.37±0.05</td>
<td>0.33±0.06</td>
</tr>
<tr>
<td>Ileum</td>
<td>0.71±0.10</td>
<td>0.58±0.08</td>
</tr>
<tr>
<td>Colon</td>
<td>0.62±0.09</td>
<td>0.65±0.11</td>
</tr>
<tr>
<td>Skeletal muscle</td>
<td>0.08±0.02</td>
<td>0.07±0.01</td>
</tr>
<tr>
<td>Skin</td>
<td>0.10±0.03</td>
<td>0.04±0.02†</td>
</tr>
</tbody>
</table>

Values are mean±SEM in ml/min/g tissue.

*p<0.01 vs. baseline; †p<0.05 vs. baseline.
was found with intravenously administered substance P in conscious dogs.18

The decreases in CBF and the increases in CVR with intravenous administration of substance P may be related to changes in cardiac metabolic demand. Decreases in CBF and increases in CVR were associated with a widening of the myocardial arteriovenous oxygen difference. In this situation, decreases in myocardial oxygen delivery were disproportionately greater and even opposite to changes in cardiac metabolic demand estimated with the pressure-work index. Thus, coronary vasoconstrictor responses cannot be accounted for by changes in cardiac metabolic demand. This conclusion is also supported by the observation that intracoronary infusions of substance P induced sustained reductions of CBF. This approach excludes the complicating influences of changes in coronary perfusion pressure and in the determinants of cardiac metabolic demand.

Increases in hemoglobin concentration with intravenous infusion of substance P could have influenced coronary hemodynamics and myocardial oxygen metabolism. In earlier studies27–29 conducted in anesthetized dogs, increases in arterial blood oxygen content secondary to large increases in hematocrit levels resulted in decreases in CBF, increases in CVR, and augmented myocardial oxygen extraction, but myocardial oxygen delivery was not altered. In the present study, the decrease in CBF resulted in a reduced myocardial oxygen delivery to the circumflex coronary bed in the face of an increase in cardiac metabolic demand. Consequently, the widening of the myocardial arteriovenous oxygen difference was the result of an imbalance between coronary perfusion and myocardial oxygen demand; a coronary vasoconstrictor effect of intravenous infusion of substance P was involved. Consistent with the hypothesis that increases in hemoglobin levels were not primarily responsible for the coronary vasoconstriction with intravenous administration of substance P, decreases in CBF and increases in CVR persisted after ganglionic blockade in the absence of changes in hemoglobin levels. In this situation, the widening of the myocardial arteriovenous oxygen difference indicates that the reduction of myocardial blood flow and oxygen delivery were secondary to a coronary vasoconstrictor effect.

A rise in hematocrit level has been reported after intravenous administration of substance P in other studies.30,31 Changes in vascular permeability have been considered as the mechanism responsible for this phenomenon.30,31 In the present study, this possibility was not directly investigated, but the observation that ganglionic blockade prevented the rise in hemoglobin after intravenous substance P suggests that a reflex mechanism could intervene as well.

Differential rates of tachyphylaxis could explain the variability of the time-dependent attenuation of substance P effects in various peripheral vascular beds.8,10–12,32 In the present study, this mechanism could have intervened because decreases in TPR
were always greater early during the infusion than at 7–9 minutes into the infusion. With respect to coronary hemodynamics, a higher rate of tachyphylaxis could explain a more-rapid return of CBF toward baseline levels than in other peripheral beds. However, tachyphylaxis alone cannot explain the sustained decrease in CBF under baseline levels that we report in the present study. On a speculative basis, vasodilator influences of substance P on the coronary bed may be attenuated in a time-dependent manner, thereby progressively revealing a concealed coronary vasoconstrictor effect.

Because our data indicate that substance P acts simultaneously as a peripheral vasodilator and as a coronary vasoconstrictor, we considered that regional blood flow responses may differ in various peripheral beds. Except for the heart, the spleen, and the skin, none of the vascular beds that we examined demonstrated a reduction in regional perfusion when substance P was administered after ganglionic blockade. Therefore, the overall vasodilator effect of substance P on the systemic circulation was not a feature shared by all regional vascular beds.

Substance P resulted in slightly greater reductions of LV perfusion in endocardial than in epicardial layers. This effect on regional blood flow distribution may be explained by a greater effect of substance P on the endocardium. However, other important factors such as decreases in coronary perfusion pressure and changes in intramyocardial tissue pressure associated with LV preload and afterload reductions may influence the transmural...
distribution of myocardial perfusion. Thus, the apparently greater effect of substance P on the endocardium could be secondary to changes in hemodynamic variables.

In conclusion, substance P exerted disparate effects on systemic and coronary beds. Vasodilation prevailed in the systemic bed. In contrast, under steady-state conditions, substance P elicited significant coronary vasoconstriction. This reduction of CBF could limit and even antagonize the influence of an increase in cardiac metabolic demand on myocardial perfusion. Therefore, in conscious dogs, substance P acts simultaneously as a...
vasodilator of the systemic circulation and as a powerful vasoconstrictor of the coronary circulation.

References


**Key Words**: substance P • coronary blood flow • peripheral resistance • myocardial oxygen metabolism • cardiac denervation
Disparate effects of substance P on systemic and coronary beds in conscious dogs.
Y Nakamura, R Parent and M Lavallée

Circulation. 1991;84:300-312
doi: 10.1161/01.CIR.84.1.300
Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1991 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7322. Online ISSN: 1524-4539

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circ.ahajournals.org/content/84/1/300

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Circulation is online at:
http://circ.ahajournals.org//subscriptions/