Augmented Responses to Vasoconstrictor Stimuli in Hypercholesterolemic and Atherosclerotic Monkeys

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SUMMARY. We examined effects of hypercholesterolemia and atherosclerosis on vasoconstrictor responses to norepinephrine and serotonin. Responses were compared in normal, atherosclerotic, and hypercholesterolemic but non-atherosclerotic cynomolgus monkeys. The hindlimb was perfused at constant flow so that changes in perfusion pressure indicated changes in vascular resistance. We measured the pressure gradient from the iliac to the dorsal pedal artery so that responses of the large artery segment could be determined. Serotonin decreased total hindlimb resistance in normal and hypercholesterolemic monkeys, but increased total resistance in atherosclerotic monkeys. There was a greater than 10-fold increase in constrictor responses of large arteries to serotonin in atherosclerotic monkeys, compared with normal and hypercholesterolemic monkeys. In contrast, we found that vasoconstrictor responses to norepinephrine are normal in atherosclerotic monkeys and increased in hypercholesterolemic monkeys prior to development of atherosclerosis. Hypercholesterolemia augmented responses of small vessels to norepinephrine. We conclude that, during early stages of hypercholesterolemia in cynomolgus monkeys, vasoconstrictor responses to norepinephrine are increased in small vessels. At a later stage, as atherosclerosis develops, responses to norepinephrine return to normal, but vasoconstrictor effects of large arteries to serotonin are greatly potentiated. (Circ Res 54: 711-718, 1984)

RECENT clinical evidence suggests that atherosclerotic vessels have an unusual propensity to develop spasm (Schroeder et al., 1977; Cipriano et al., 1979; Waters et al., 1983). Studies of vessels in vitro suggest several mechanisms by which hypercholesterolemia and atherosclerosis may augment contractile responses. First, increased cholesterol content of membranes augments cation permeability (Wiley and Cooper, 1975) and alters vascular responses to Ca++ and K+ (Yokoyama and Henry, 1979). Second, the number of serotonergic and adrenergic receptors is increased in atherosclerotic aortas (Nanda and Henry, 1982). Contractile responses to serotonin and adrenergic agonists are increased in atherosclerotic vessels (Henry and Yokoyama, 1980; Yokoyama et al., 1983). Third, several vasoactive substances release a potent vasodilator from endothelium (Furchgott and Zawadski, 1980; DeMey and Vanhoutte, 1982). One might speculate that dysfunction or loss of the endothelium in atherosclerosis may impair release of vasodilator substances from endothelium and thereby augment vasoconstrictor responses (Vanhoutte, 1983).

Four lines of evidence suggest that responses to vasoconstrictor stimuli in vivo may be augmented by hypercholesterolemia or atherosclerosis. First, coronary vasoconstrictor responses to norepinephrine have been reported to be increased modestly in hypercholesterolemic dogs (Rosendorff et al., 1981). Second, reflex coronary vasoconstriction has been reported to be greater in patients with atherosclerotic coronary disease than in those with normal coronary arteries (Mudge et al., 1976). Third, intravenous or intracoronary administration of histamine has been reported to produce coronary artery spasm in atherosclerotic swine (Shimokawa et al., 1983). Fourth, spasm of coronary arteries is common in patients with coronary atherosclerosis (Schroeder et al., 1977) and may occur in limb vessels of patients with peripheral vascular disease (Juergens et al., 1980). It has been suggested that the incidence of spasm of limb vessels, with Raynaud's phenomenon, is increased in patients with coronary vasospasm (Robertson and Oates, 1978; Miller et al., 1981).

The purpose of this study was to examine effects of hypercholesterolemia and atherosclerosis on vasoconstrictor responses in vivo. Several aspects of the study deserve emphasis. First, based on the finding that serotonergic receptors are increased more than α-adrenergic receptors in the aorta of atherosclerotic rabbits (Nanda and Henry, 1982), our hypothesis was that responses to serotonin would be potentiated more than responses to norepinephrine in atherosclerotic animals. Second, we
used a preparation that allowed quantitative determination of large artery resistance, as well as total limb resistance. Third, we studied a primate model of atherosclerosis.

Methods

Three groups of adult cynomolgus monkeys, without signs of senescence, were studied. Twenty control monkeys that weighed 5.7 ± 0.2 kg (mean ± se) were fed commercial laboratory chow (Purina monkey chow, Ralston Purina Co.). In 23 monkeys that weighed 5.1 ± 0.2 kg, atherosclerosis was produced by feeding them a semi-purified atherogenic diet containing 41% fat and 0.8% cholesterol for 3.2 to 5 years. In six monkeys that weighed 5.0 ± 0.2 kg, we produced hypercholesterolemia, but not atherosclerosis, by feeding the monkeys atherogenic diet for 4–5 months. At the time of the terminal study, the monkeys were sedated with ketamine HCl (12 mg/kg, im) and anesthetized with chloralose (75 mg/kg, iv).

All monkeys were caged individually in thermoregulated rooms. Venous blood samples were drawn after sedation with ketamine. Total cholesterol and triglycerides were determined by the method of Abell et al. (1952), as modified by the Lipid Research Clinics Protocol for the Autoanalyzer II (Technicon Instruments Inc.). Triglycerides were measured by the corresponding method in the same protocol.

After the monkeys were anesthetized, they were intubated and ventilated with room air and supplemental oxygen via a Harvard model 661 small animal respiratory pump (Harvard Apparatus Co.). Rectal temperature was maintained at 37–38°C with a heating pad. Catheters were placed in the right brachial artery and vein. Heparin (500 U/kg, iv) was used for anticoagulation and decamethonium bromide (0.5 mg/kg, iv) for skeletal muscle paralysis. Blood gases were monitored with an IL 113 Blood Gas Analyzer (Instrumentation Laboratories).

Hindlimb Perfusion

Through a laparotomy, the abdominal aorta and left iliac artery were exposed and isolated. The left dorsal pedal artery also was exposed. A calibrated Harvard model 1210 pulsatile perfusion pump was used to perfuse the left iliac artery at constant flow with blood from the abdominal aorta. When the pump was stopped, perfusion pressure decreased rapidly to 10–15 mm Hg, which indicates that vascular isolation was adequate. Pressure was measured continuously from a sidearm several centimeters upstream from the tip of the iliac cannula. The rate of the pump was adjusted so that perfusion pressure at the beginning of the experimental protocol was similar to the animal’s mean systemic arterial pressure. The left dorsal pedal artery was ligated, and a cannula was inserted upstream from the ligature to measure pressure. The difference between iliac perfusion pressure and dorsal pedal pressure reflects resistance of large arteries of the limb.

At the end of the experiment, the leg was severed through the hip joint. Volume of the leg was measured by displacement of water, so that limb blood flow and vascular resistance could be expressed per 100 ml of limb.

Experimental Protocol

We studied effects of norepinephrine and serotonin in all monkeys. Norepinephrine (0.2 and 1.2 µg) and serotonin (5 and 50 µg) were injected as a bolus into the iliac perfusion tubing.

Responses to serotonin and norepinephrine were observed before and after phentolamine and methysergide in four normal, four hypercholesterolemic, and five atherosclerotic monkeys. Responses to norepinephrine (0.2 and 1.2 µg) and serotonin (20 or 50 µg) were determined under control conditions, before the antagonists. Phentolamine (300 µg) was injected into the perfusion tubing in 15–30 seconds and, after a transient decrease in perfusion pressure, injections of norepinephrine were repeated. In all animals, vasoconstrictor responses to norepinephrine were reduced by phentolamine. Either 20 or 50 µg of serotonin then were injected ia. After recovery of α-receptor responses, as demonstrated by injections of norepinephrine, injections of 5 and 50 µg of serotonin were repeated. Methysergide, an antagonist primarily of 5-HT1 serotoninergic receptors (Peroutka and Snyder, 1983), was then infused at 25 µg/min, iv. Norepinephrine (0.2 and 1.2 µg) and serotonin (5 and 50 µg) were injected during infusion of methysergide. Because effects of methysergide are prolonged, these were the final responses observed in the hindlimb. Effects of ketanserin, an antagonist of 5-HT2 serotoninergic receptors (Peroutka and Snyder, 1983), were observed in four other atherosclerotic monkeys. Responses to norepinephrine (0.2 and 1.2 µg) and serotonin (20 or 50 µg) were observed before and after ketanserin (0.08 to 0.16 mg/kg, iv).

The gradient in pressure from the iliac to dorsal pedal artery is used to calculate resistance of large arteries, with the assumption that the interventions do not redistribute blood flow between the proximal and distal limb, and thereby alter the fraction of flow which traverses the large artery segment. Thus, although total flow to the perfused hindlimb was known, determination of the proportion of flow to proximal and distal segments of the hindlimb was also important. In five normal, five atherosclerotic, and five hypercholesterolemic monkeys, a catheter was placed in the left atrial appendage for injection of microspheres. The left brachial artery and right common carotid artery were isolated and cannulated in preparation for withdrawal of reference arterial blood samples. Microspheres labeled with 86Sr, 89Sr, 14C, and 125I were injected into the left atrium to obtain four measurements of blood flow in the perfused hindlimb. Microspheres 15 µm in diameter were injected during control conditions, during the plateau phase of the prolonged change in perfusion pressure which followed injections of 5 and 50 µg of serotonin ia, and during the recovery period. Shunting of microspheres was determined for each measurement of flow by withdrawing venous blood samples from a cannula placed in the right femoral vein and advanced to the bifurcation of the left iliac vein. Average shunting (ratio of counts in venous and arterial blood) was approximately 1% during control and after injection of serotonin, and ranged between 0 and 3%. After the monkey was killed, several muscle samples were taken from the thigh (rectis femoris, gracilis, and biceps femoris) and calf (tibialis anterior, medial gastrocnemius, and lateral gastrocnemius), and one sample was obtained from the foot (flexor digitorum brevis). Skin was sampled from the thigh and calf. Blood flow for each sample was calculated from the formula, flow = counts (per g tissue) · 100 · reference blood flow rate (rate of withdrawal of reference arterial blood samples) + counts (per ml arterial blood).
Morphological Study

The iliac and femoral arteries were removed, examined for gross lesions, and fixed in formalin in 12 of 20 normal, 6 of 6 hypercholesterolemic, and 13 of 20 atherosclerotic animals. Histological study was carried out on carbowax sections of preselected sites from the proximal iliac and femoral arteries.

Morphometric determination was performed with an image analyzer to evaluate the degree of stenosis, as described previously (Armstrong et al., 1983). Percent stenosis was calculated from the ratio of intimal area to cross-sectional area enclosed by the internal elastic lamina. The measurements were made in undistended arteries and corrected to the estimated values during distension (Armstrong et al., 1983).

Statistical Analysis

The paired t-test was used to compare two values: e.g., responses to an agonist before and after administration of an antagonist. Analysis of variance was used to compare three groups: e.g., responses to a vasoconstrictor stimulus in normal, atherosclerotic, and hypercholesterolemic cynomolgus monkeys. When significant intergroup differences were present (α level = P < 0.05), Duncan's test was used to determine which pairs of values were different.

Results

Plasma Lipids

Plasma cholesterol was 116 ± 4 mg/dl in control monkeys, 448 ± 35 mg/dl during the last month of atherogenic diet in hypercholesterolemic monkeys, and 530 ± 20 mg/dl during the last 3 months of atherogenic diet in atherosclerotic monkeys. Plasma triglycerides were 25-40 mg/dl in control, hypercholesterolemic, and atherosclerotic monkeys.

Morphological Changes

In atherosclerotic monkeys, morphological changes confirmed previous reports (Kramsch and Hollander, 1968; Armstrong, 1976; Wagner et al., 1978; Armstrong et al., 1983) that demonstrated dense fibrofatty intimal thickening with focal necrosis and calcification in monkeys fed atherogenic diet for sustained periods. In animals fed atherogenic diet for 4–5 months, the iliac and femoral arteries were normal in five monkeys; one monkey had a small intimal lesion in the iliac artery.

The percent stenosis in the iliac and femoral arteries was 54 ± 7 and 41 ± 11 in atherosclerotic monkeys, 3.9 ± 3.9 and 0 ± 0 in hypercholesterolemic monkeys, and 1.3 ± 1.1 and 0.2 ± 0.2 in normal monkeys.

Hemodynamic Studies

Baseline Changes

Total limb vascular resistance was not significantly different in normal, atherosclerotic, and hypercholesterolemic monkeys (P > 0.05) (Table 1). The pressure gradient from iliac to dorsal pedal arteries, and the resistance of the large artery segment, were significantly higher in atherosclerotic monkeys (P < 0.05) (Table 1).

Responses to Serotonin

In normal monkeys, serotonin produced vasodilation in the limb (reduction in iliac perfusion pressure) (Fig. 1). In five of six hypercholesterolemic monkeys, serotonin produced vasodilation in the limb; in the only hypercholesterolemic monkey that had a fatty streak in the limb, serotonin produced marked vasoconstriction. In atherosclerotic monkeys, serotonin produced vasoconstriction (increase in iliac perfusion pressure) (P < 0.05, atherosclerotic vs. normal and hypercholesterolemic monkeys).

Serotonin produced more constriction of large arteries in atherosclerotic monkeys than in normal or hypercholesterolemic monkeys (P < 0.05) (Fig. 1). In terms of relative potency, responses of large arteries were augmented more than 10-fold by atherosclerosis: 5 µg of serotonin produced more vasoconstriction in large arteries of atherosclerotic monkeys than 50 µg produced in normal monkeys.

Constriction of large arteries in the atherosclerotic limb after injection of serotonin produced a profound decrease in pressure in the dorsal pedal artery, from 59 ± 5 mm Hg during control to 17 ± 4 mm Hg after injection of 50 µg serotonin. Although the limb was perfused at constant flow, serotonin redistributed blood flow from the distal to proximal limb of atherosclerotic monkeys: serotonin produced a marked decrease in blood flow to the distal foot and calf muscle, and an increase in flow to skin of the thigh. Specifically, blood flow to distal (foot) muscle (ml/min per g) was 8.4 ± 2.8 during control, 1.1 ± 0.8 after 50 µg serotonin, and 8.2 ± 1.1 during recovery (P < 0.05 serotonin vs. control and recovery).
TABLE 1
Baseline Values in Normal, Atherosclerotic, and Hypercholesterolemic Monkeys

<table>
<thead>
<tr>
<th></th>
<th>Normal (n = 20)</th>
<th>Atherosclerotic (n = 20)</th>
<th>Hypercholesterolemic (n = 6)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean arterial pressure</td>
<td>70 ± 2.3</td>
<td>67 ± 2.6</td>
<td>76 ± 6.3</td>
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<tr>
<td>(mm Hg)</td>
<td></td>
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<tr>
<td>Hind limb blood flow</td>
<td>22 ± 1.8</td>
<td>21 ± 2.2</td>
<td>28 ± 3.4</td>
</tr>
<tr>
<td>(ml/min per 100 g)</td>
<td></td>
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<tr>
<td>Hind limb vascular</td>
<td>4.2 ± 0.4</td>
<td>4.9 ± 0.7</td>
<td>4.1 ± 0.9</td>
</tr>
<tr>
<td>resistance (mm Hg)</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>(mm Hg/ml per min per</td>
<td>29 ± 2.0</td>
<td>42 ± 6.2*</td>
<td>25 ± 7.3</td>
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<tr>
<td>100 g)</td>
<td></td>
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<tr>
<td>Large artery pressure</td>
<td>1.6 ± 0.2</td>
<td>2.6 ± 0.6*</td>
<td>1.1 ± 0.4</td>
</tr>
<tr>
<td>gradient (mm Hg)</td>
<td></td>
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<tr>
<td>Large artery resistance</td>
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<tr>
<td>(mm Hg/ml per min per 100 g)</td>
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Values are mean ± SE.

* Large artery pressure gradient and resistance were higher in atherosclerotic than in normal or hypercholesterolemic monkeys (P < 0.05).

Flow to calf muscle was 14 ± 2.6 during control, 4.8 ± 2.0 after serotonin, and 12 ± 2.9 during recovery (P < 0.05 serotonin vs. control and recovery). Flow to thigh skin was 6.2 ± 2.4 during control, 12 ± 3.6 after serotonin, and 5.8 ± 2.6 during recovery (P < 0.05 serotonin vs. control and recovery). Serotonin did not selectively alter distribution of flow to proximal and distal skin and muscle in normal or hypercholesterolemic monkeys, except serotonin decreased flow to calf muscle in normal monkeys: control = 16 ± 2.7, serotonin = 11 ± 1.8, and recovery = 17 ± 3.0 (P < 0.05 serotonin vs. control, NS vs. recovery).

Response to Norepinephrine

Norepinephrine produced more vasoconstriction in hypercholesterolemic monkeys than in normal or atherosclerotic monkeys (P < 0.05) (Fig. 2). Norepinephrine did not produce constriction of large arteries in any group (Fig. 2). Thus, augmented vasoconstrictor responses to norepinephrine in hypercholesterolemic monkeys resulted from increased responsiveness of small resistance vessels.

Effects of Antagonists

Methysergide, an antagonist of serotonergic receptors, blocked both the total limb and large artery vasoconstrictor response to serotonin in atherosclerotic monkeys (Fig. 3). Methysergide also attenuated the increase in total limb perfusion pressure after injection of 1.2 µg norepinephrine (from 38 ± 8 to 22 ± 5 mm Hg, P < 0.05), but this effect was substantially less than the blockade of responses to serotonin.

Methysergide tended to attenuate the vasodilator response to serotonin in normal monkeys: serotonin (20 µg in two monkeys, 50 µg in two monkeys) reduced total limb perfusion pressure 28 ± 8 mm Hg before and 16 ± 6 mm Hg after methysergide (P > 0.05). Methysergide blocked the constrictor re-
Perfusion
300 ng (P > 0.05, before vs. after phentolamine).

Monkeys) were not altered by intra-arterial injection of phentolamine in atherosclerotic cynomolgus monkeys. Responses to intra-arterial injection of serotonin (20 ng in two monkeys and 50 pg in two of atherosclerotic monkeys and no change, or a decrease. This potentiation of vasoconstrictor responses to serotonin in atherosclerotic monkeys is not mediated by hypercholesterolemia per se. This conclusion is based on the finding that hypercholesterolemia, before development of atherosclerosis, does not augment responses to serotonin. Third, vasoconstrictor responses to serotonin are blocked by methysergide, but not by phentolamine. Thus, as suggested by studies in vitro (Henry and Yokoyama, 1980), augmentation of responses to serotonin by atherosclerosis appears to be mediated by serotonergic receptors. Fourth, potentiation of vasoconstrictor responses by atherosclerosis is selective: responses to serotonin are augmented, but responses to norepinephrine are not altered.

In addition, we found that vasoconstrictor responses to norepinephrine are potentiated in hypercholesterolemic monkeys prior to development of atherosclerosis. At a later stage, as atherosclerosis develops, responses to norepinephrine return to normal, but vasoconstrictor effects of serotonin are greatly augmented.

Limb Perfusion and Large Artery Resistance

Our conclusions, that atherosclerosis potentiates responses to serotonin and hypercholesterolemia potentiates responses to norepinephrine, are based on changes in perfusion pressure. Calculation of large artery resistance, however, allows insight into the site of action of the altered responses.

Previous studies have demonstrated that norepinephrine produces constriction of arterioles and small arteries, but not large arteries (Haddy et al., 1957; Abboud, 1968). In our experiments, we also found that norepinephrine increases perfusion pressure, but not large artery resistance, which indicates that effects of norepinephrine are confined to small vessels. Thus, we conclude that potentiation of norepinephrine by hypercholesterolemia occurs at the level of small vessels.

In contrast to the site of action of norepinephrine, previous studies indicate that serotonin constricts large arteries, but not arterioles (Haddy et al., 1957; Daughtery et al., 1968; Abboud, 1968). In these experiments, we found that serotonin constricts large arteries, but decreases perfusion pressure in normal monkeys. Atherosclerosis augments responses to serotonin more than 10-fold in large arteries of the limb. Second, potentiation of responses to serotonin in atherosclerotic monkeys is not mediated by hypercholesterolemia per se. This conclusion is based on the finding that hypercholesterolemia, before development of atherosclerosis, does not augment responses to serotonin. Third, vasoconstrictor responses to serotonin are blocked by methysergide, but not by phentolamine. Thus, as suggested by studies in vitro (Henry and Yokoyama, 1980), augmentation of responses to serotonin by atherosclerosis appears to be mediated by serotonergic receptors. Fourth, potentiation of vasoconstrictor responses by atherosclerosis is selective: responses to serotonin are augmented, but responses to norepinephrine are not altered.

When we use changes in perfusion pressure from the iliac to dorsal pedal arteries as a reflection of...
large artery resistance, we assume that the interventions do not produce a redistribution of blood flow within the limb and thereby alter the fraction of blood flow which traverses the large artery segment. We found, using microspheres, that serotonin produces some redistribution of flow in the limb of atherosclerotic monkeys, as blood flow to the distal limb is reduced and flow to the proximal limb increases. Thus, a smaller fraction of limb blood flow traverses the large artery segment during serotonin than during control conditions. These data indicate that we have underestimated the effects of serotonin on large artery resistance in atherosclerotic monkeys; thus, potentiation of vasoconstrictor responses to serotonin by atherosclerosis is even greater than the 10-fold potentiation which we have observed.

Previous Studies in Vivo

We are aware of two previous studies which suggest that blood lipids affect vasoconstrictor responses in vivo. Infusion of a β-lipoprotein extract into the carotid artery of baboons has been reported to potentiate cerebral vasoconstrictor responses to serotonin (Eidelberg et al., 1978). Because the authors presented the response to serotonin only in terms of the fast clearance curve, and not total flow, it is difficult to interpret their findings (Marcus et al., 1981). In contrast to the findings of Eidelberg et al. (1978) in the cerebral circulation, in hypercholesterolemic monkeys before the development of atherosclerosis, we did not observe augmentation of responses to serotonin in the limb.

Coronary vasoconstrictor responses to norepinephrine have been reported to be augmented in unanesthetized dogs that were fed a high-cholesterol diet for 1 month (Rosendorff et al., 1981). Norepinephrine (1 μg/min) produced a modest increase in coronary vascular resistance in cholesterol-fed dogs, which was greater than the increase in resistance in normal dogs. In a preliminary report (Johannsen et al., 1981) we found that coronary vasoconstrictor responses to sympathetic nerve stimulation and phenylephrine in anesthetized dogs were not altered by a high-cholesterol diet for 8–10 weeks. Thus, effects of hypercholesterolemia on vasoconstrictor responses to norepinephrine in the coronary bed are not consistent.

Clinical studies indicate that ergonovine produces modest constriction of conduit coronary arteries, and that, in occasional susceptible patients, coronary "spasm" is induced (Schroeder et al., 1977; Cipriano et al., 1979; Waters et al., 1983). Potentiation of responses to ergonovine by atherosclerosis appears to be mediated primarily by serotonergic mechanisms (Henry and Yokoyama, 1980; Yokoyama et al., 1983). Thus, it is likely that susceptibility to ergonovine-induced coronary spasm which has been observed clinically is mediated by serotonergic mechanisms. In our experiments in the limb, we found that responses to serotonin in large arteries of atherosclerotic monkeys were potentiated to such an extent that pressure in the dorsal pedal artery was less than 20 mm Hg. This response might be characterized as "spasm" of large arteries.

Non-serotonergic mechanisms also have been proposed as contributors to augmented vasoconstrictor responses in patients and experimental animals with atherosclerosis. First, it has been suggested that histamine may produce coronary artery spasms (Ginsburg et al., 1981; Shimokawa et al., 1983). Second, the cold pressor test, a reflex vasoconstrictor stimulus, has been observed to increase coronary vascular resistance in patients with atherosclerotic coronary artery disease but not in patients with normal coronary arteries (Mudge et al., 1976). The authors suggested that augmented responses to the cold pressor test are mediated by depletion of coronary vasodilator reserve in atherosclerosis, but an alternative interpretation is that response of large coronary arteries to adrenergic stimuli are augmented. An important limitation of the study is that a thermodilution method was used to measure coronary blood flow. The method has major sources of error (Mathey et al., 1978), and it has not been demonstrated to be valid in the presence of coronary artery disease.

Most clinical studies that have described spasm relate to coronary, not limb, vessels. In the limb, Raynaud's phenomenon occurs rarely in arteriosclerotic disease, and more commonly in thromboangiitis obliterans (Juergens et al., 1980). It is not clear whether our findings of augmented responses to serotonin in the atherosclerotic limb can be extrapolated to other vascular beds, but the findings by other investigators which suggest increased responses to ergonovine in atherosclerotic coronary arteries and aorta (Schroeder et al., 1977; Henry and Yokoyama, 1980) support the concept that augmented responsiveness to serotonin may be a general characteristic of atherosclerotic vessels.

Possible Mechanisms of Altered Vasoconstrictor Responses

Studies by Henry and Yokoyama (1980) and Yokoyama et al. (1983) suggest that experimental atherosclerosis alters vascular contractile responses to several agonists. In hyperlipidemic and atherosclerotic rabbits, contractile responses of aortic strips and coronary strips to ergonovine and serotonin were augmented, and responses to KCl and phenylephrine were unchanged. Augmented responses to ergonovine were blocked by cyproheptadine, a serotonergic receptor antagonist, but not by α-receptor antagonists. In a preliminary report, Nanda and Henry (1982) found an increase in the number of serotonin receptors, and to a lesser extent α-adrenergic receptors, in the aorta of atherosclerotic rabbits. Our finding that vasoconstrictor responses to serotonin are potentiated in atherosclerotic monkeys confirms the studies of Henry and colleagues,
which were performed in vitro, and may be explained in part by an increase in number of vascular serotonergic receptors.

A second possible mechanism by which vascular responses are altered in hypercholesterolemia and atherosclerosis relates to changes in membrane cholesterol content. When membrane cholesterol content is increased, fluidity and permeability of cell membranes is increased (Poznansky et al., 1973; Jain and White, 1977). In coronary arteries in vitro, cholesterol can induce contraction and also potentiate vasococontractor responses to ionic calcium and potassium (Yokoyama and Henry, 1979). Potentiation of responses to calcium and potassium presumably occurs because cholesterol alters permeability of vascular muscle to the cations. Our findings of altered vascular responses may be explained in part by alterations in permeability of vascular membrane. One might expect, however, that membrane changes would alter responses to a variety of vasococontractor stimuli to a similar extent. Our findings of specific potentiation of responses to norepinephrine during hypercholesterolemia, and specific potentiation of responses to serotonin by atherosclerosis, suggest that membrane changes do not play a large role in these effects.

It is likely that dysfunction or loss of vascular endothelium in atherosclerosis may play an important role in potentiation of responses to serotonin. Monoamine oxidase in endothelial cells degrades serotonin (Gillis, 1980), so that endothelial dysfunction in atherosclerosis might be expected to impair the enzymatic degradation of serotonin and thereby augment responses to serotonin. Furthermore, Furchgott and Zawadzki (1980) and other investigators (DeMey and Vanhoutte, 1982) have demonstrated that removal of endothelium reduces vasodilator responses to several agonists in vitro, apparently because many vasoactive substances release an unidentified, potent vasodilator from the endothelium. A recent study (Cohen et al., 1983) indicates that aggregating platelets produce contraction in rings of coronary arteries; contractions were augmented by removal of endothelium and attenuated by serotonergic antagonists. The findings suggest an important role of endothelium in modulation of serotonin-induced vasocostriction.

Implications of Altered Responsiveness to Serotonin

We speculate that the finding that vasococontractor responses to serotonin are potentiated by atherosclerosis may have important implications. This speculation is based on the assumptions that findings in the primate model of atherosclerosis are applicable to atherosclerosis in humans, and that augmented responses to serotonin in the limb are applicable to other vascular beds. It has been suggested that platelets, which contain substantial amounts of serotonin, may aggregate at sites of endothelial dysfunction produced by atherosclerotic lesions, and that the platelets may release serotonin. We suggest that a pronounced increase in responsiveness of large arteries to serotonin in atherosclerotic vessels may play an important role in the pathogenesis of vascular spasm.

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