Structural and Hemodynamic Responses of Peripheral Arteries of Macaque Monkeys to Atherogenic Diet

Mark L. Armstrong, Donald D. Heistad, Melvin L. Marcus, Marjorie B. Megan, and Donald J. Piegors

The arteries of monkeys given atherogenic diets develop marked intimal thickening and medial thinning, but luminal size apparently changes minimally. The hemodynamic significance of the atherosclerotic changes is therefore uncertain. To evaluate vascular function in atherosclerotic arteries, we studied the hind-limb vessels of adult male rhesus and cynomolgus monkeys to assess the structural and hemodynamic responses to an atherogenic diet given for about 1.5 years or for much longer periods (6.5 years for rhesus and 4.3 years for cynomolgus monkeys). The intimal cross-sectional area greatly increased after the atherogenic diet, but there was no significant luminal narrowing after either the 1.5-year diet or the longer diet periods. The media of atherosclerotic arteries showed focal atrophy and focal thinning after pressure fixation, but the total medial mass was not decreased even after the long diet periods. Hemodynamic studies indicated mild functional impairment in the atherosclerotic vessels; resting resistance increased and vasodilator responses decreased, but adrenergic responses were preserved. Thus, the marked changes that occur in the arterial wall in experimental primate atherosclerosis include adaptations to lesion formation that permit a long prestenotic phase of atherosclerosis in which vascular dysfunction is minimal. (Arteriosclerosis 5:336-346, July/August 1985)
Third, we have usually studied atherosclerosis in monkeys after an atherogenic diet of about 1.5 years.\textsuperscript{18,20} at which time there are prominent arterial lesions.\textsuperscript{25-27} Do longer periods of atherogenic diet cause greater structural changes with more marked hemodynamic abnormalities?

Methods

Animals

We studied adult male rhesus and cynomolgus monkeys and used diet regimens to prepare control and atherosclerotic monkeys. Duration of the diet regimens was either about 1.5 years, a time we designated "medium-term," or for much longer periods (about 6 years in rhesus and 4 years in cynomolgus monkeys) that we designated "long-term." The number of animals assigned to each regimen, the average and range of the diet intervals, and the initial and final body weights are shown in Table 1.

The control monkeys were fed commercial laboratory chow (Purina Monkey Chow, Ralston Purina Company, Richmond, Indiana.) Rhesus atherosclerotic monkeys were fed a semipurified atherogenic diet\textsuperscript{29} that contained 41% of the total calories as fat and 1.2% cholesterol. The cynomolgus atherosclerotic monkeys were fed the same atherogenic diet except that the cholesterol content was 0.8%.\textsuperscript{20,29} All monkeys were individually caged in thermoregulated rooms. Venous blood samples were drawn bimonthly after sedation with ketamine HCl or phencyclidine HCl. The total cholesterol was determined by the method of Abell et  al.,\textsuperscript{30} as modified by the Lipid Research Clinics Protocol for the Autoanalyzer II (Technicon Insruments Inc., Tarrytown, New York). Triglycerides were measured by the corresponding method in the same protocol.

Morphological Studies

The monkeys were killed by exanguination at the end of the hemodynamic studies (see following section). The iliac and femoral arteries in all rhesus monkeys and in most cynomolgus monkeys (12 control and 13 atherosclerotic animals) were removed and fixed by immersion in formalin. In six control and 13 atherosclerotic cynomolgus monkeys, the arteries were fixed in situ at 120 mm Hg with formalin-barium-gelatin by a technique described previously.\textsuperscript{24} Except for animals whose arteries were pressure-fixed for postmortem angiography, the intimal surface involvement was estimated visually in 5% units by two observers and was expressed as the mean and range of involvement.\textsuperscript{20} Tissue samples were taken at four standardized sites in the hindlimb arteries (the proximal and midsegment areas of the iliac and femoral arteries); these samples were prepared for microscopy, and paraaffin cross-sections were taken for histologic study.

Morphometric comparisons were made using methods and calculations previously described.\textsuperscript{20} In brief, the cross-sectional areas of the intima and media were projected and digitized. To determine the luminal area in immersion-fixed arteries, the cross-sectional area enclosed by the internal elastic lamina was corrected to a circle by applying the form factor P/477 to the measurement of the internal elastic lamina, where I = length of the lamina; the luminal area was calculated as the difference between the corrected area within the internal elastic lamina and the intimal area.

We used morphometric methods to estimate the extent of the focal medial atrophy and the extent of the eccentricity of the intimal lesions in atherosclerosis. To express these abnormalities quantitatively, we computed the average widths of the media and in atherosclerotic monkeys, the intima in each projected microscopic section. We then measured the minimal width of the media and in atherosclerotic monkeys, the maximal width of the intima. We used the ratio of minimal medial width to average medial width in each section as an index of focal medial thinning (Figure 1 A and B). Focal medial atrophy in atherosclerosis was defined as medial thickening greater than that in controls.

The eccentricity of the intimal lesions (i.e., the non-uniform thickening of the atherosclerotic intima) was expressed by a lesion shape ratio. The maximal inti-

Table 1. Body Weight and Duration of Diet

<table>
<thead>
<tr>
<th>Diet</th>
<th>Period</th>
<th>Regimen</th>
<th>No.</th>
<th>Initial (kg)</th>
<th>Final (kg)</th>
<th>Diet duration (mos)</th>
<th>Avg</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rhesus</td>
<td>Medium-term</td>
<td>Control</td>
<td>8</td>
<td>5.3 ± 0.15</td>
<td>7.0 ± 0.26</td>
<td>18 (17–22)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Atherogenic</td>
<td>18</td>
<td>5.1 ± 0.14</td>
<td>6.2 ± 0.22</td>
<td>18 (16–23)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Long-term</td>
<td>Control</td>
<td>8</td>
<td>5.9 ± 0.40</td>
<td>8.6 ± 0.58</td>
<td>69 (56–88)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Atherogenic</td>
<td>7</td>
<td>6.7 ± 0.37</td>
<td>8.9 ± 0.36</td>
<td>77 (72–82)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cynomolgus</td>
<td>Medium-term</td>
<td>Control</td>
<td>11</td>
<td>4.2 ± 0.16</td>
<td>4.8 ± 0.26</td>
<td>20 (19–22)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Atherogenic</td>
<td>11</td>
<td>5.3 ± 0.31</td>
<td>5.2 ± 0.35</td>
<td>20 (18–23)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Long-term</td>
<td>Control</td>
<td>18</td>
<td>5.0 ± 0.20</td>
<td>5.4 ± 0.22</td>
<td>53 (41–60)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Atherogenic</td>
<td>26</td>
<td>5.1 ± 0.15</td>
<td>5.4 ± 0.17</td>
<td>52 (43–58)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Body weight values are means ± SEM.
mal thickness was normalized to a computed average intimal thickness in each section (Figure 1C). Uniform thickening of the intima around the circumference of the lumen would form a concentric lesion with a shape ratio of 1.0; the shape ratio rose with the increasing eccentricity of the lesion.

Hemodynamic Studies

At the conclusion of the diet periods, we evaluated the pressure-flow curves, the responses to neural and humoral vasoconstrictor stimuli, and the vasodilator responses with previously described methods. In brief, the monkeys were anesthetized with chloralose (75 mg/kg, i.v.), and an iliac artery was perfused at constant flow with a pulsatile pump. The dorsal pedal artery was cannulated to measure the iliac-dorsal pedal pressure gradient and to estimate the resistance of the large arteries of the limb. The baseline perfusion pressure of the hindlimb bed was established by adjusting blood flow so that the perfusion pressure was similar to the animal’s mean systemic arterial pressure. The perfusion pressure was measured at the three levels of blood flow used in the resting state. The dose of papaverine was then increased to 5.0 mg/min. The higher dose usually did not produce a further decrease in perfusion pressure, which indicates that the vessels were dilated maximally to the stimulus. In those experiments in which further vasodilation was observed, pressure-flow determinations were repeated.

The vascular resistance was calculated by dividing the perfusion pressure by the flood flow through the perfused iliac artery. The allocation of monkeys to each of these hemodynamic studies is shown in Table 2.

Statistical Analysis

The pressure-flow slopes fulfilled the conditions of parallelism, and covariance analyses were used to test for significant differences between atherosclerotic and control groups. In parametric tests, the values are expressed as means ± SEM unless otherwise stated. In nonparametric comparisons (e.g., of normalized media and intima) the rank-sum test was used and the estimates of the average values are expressed as medians. A significance level of $\alpha = 0.05$ was used.

Results

Plasma Lipids

Rhesus Monkeys

The cholesterol levels in the control monkeys after 2 to 3 months of chow diet were 128 ± 6 mg/dl in the group whose diet period was of medium-term duration and 110 ± 4 mg/dl in the long-term group fed for about 6 years. In atherosclerotic monkeys, the cholesterol level rose rapidly during the first 5 months to plateau levels of 400 mg/dl to 800 mg/dl. In the mon-
Table 2. Allocation of Monkeys to Hemodynamic Studies

<table>
<thead>
<tr>
<th>Study</th>
<th>Rhesus monkey diet period</th>
<th>Cynomolgus monkey diet period</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Medium-term</td>
<td>Long-term</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>ATH</td>
</tr>
<tr>
<td>Vasoconstrictor responses</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Humoral agonists</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phenylephrine</td>
<td>8</td>
<td>18</td>
</tr>
<tr>
<td>Angiotensin</td>
<td>8</td>
<td>18</td>
</tr>
<tr>
<td>Norepinephrine</td>
<td>8</td>
<td>18</td>
</tr>
<tr>
<td>Neural stimuli</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Carotid occlusion</td>
<td>10</td>
<td>18</td>
</tr>
<tr>
<td>Direct sympathetic stimulation</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vasodilator responses</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Papaverine/adenosine</td>
<td>8</td>
<td>18</td>
</tr>
</tbody>
</table>

Numbers indicate the number of animals studied. C = control diet regimen; ATH = atherogenic diet regimen.

keys fed an atherogenic diet for a medium-term period (18 months), the cholesterol was 557 ± 23 mg/dl in the 4 months before autopsy. In the monkeys fed the atherogenic diet for a long-term period (about 6 years), the cholesterol levels were 10% higher and averaged 614 ± 27 mg/dl in the 4 months before autopsy. Plasma triglycerides were similar in all groups during the control and atherogenic diets, ranging from 15 mg/dl to 40 mg/dl.

Cynomolgus Monkeys

The cholesterol levels in the controls were 98 ± 2 mg/dl in the group whose diet duration was of medium-term duration and 106 ± 3 mg/dl in the long-term group fed for about 4 years. In atherosclerotic cynomolgus monkeys, the cholesterol rose rapidly to a plateau, as in the rhesus monkeys. In the monkeys fed the atherogenic diet for a medium-term period (20 months), cholesterol was 530 ± 26 mg/dl in the 4 months before autopsy. In the monkeys fed the atherogenic diet for a long-term period (about 4 years), cholesterol levels were 607 ± 30 mg/dl in the 4 months before autopsy. Plasma triglycerides ranged from 25 to 40 mg/dl in both control and atherosclerotic cynomolgus monkeys.

Morphologic Findings

Rhesus Monkeys

The controls had no gross lesions in the limb arteries. After 18 months of the atherogenic diet, the extent of intimal surface involvement was 20% in the iliac arteries (the mean of visual estimates ranging from 15% to 50%) and 13% (10% to 35%) in the femoral arteries. After about 6 years of the atherogenic diet, the intimal surface involvement was 72% (30% to 95%) in the iliac and 38% (10% to 60%) in the femoral arteries. The animals fed the atherogenic diet for 18 months showed a well-defined histologic picture of fibrofatty intimal thickening with areas of necrosis and calcification. These changes have been extensively characterized previously.25,33 Rhesus monkeys fed the atherogenic diet for about 6 years had marked intimal thickening and more extracellular lipid in the lesions than was found at 1.5 years.

Cynomolgus Monkeys

The control monkeys had no gross lesions in the limb arteries. After 20 months of the atherogenic diet, the intimal surface involvement was 32% (15% to 60%) and 12% (5% to 30%) in the iliac and femoral arteries, respectively. After about 4 years of the atherogenic diet, the involvement was 78% (25% to 95%) and 31% (15% to 65%) in the iliac and femoral arteries, respectively. In monkeys fed the atherogenic diet for 20 months, the vessels had marked intimal thickening. There often was calcification of intima in cynomolgus monkeys, as we have previously described after a medium-term diet interval.18 In monkeys given the atherogenic diet for about 4 years, there was slightly more intimal thickening, but little qualitative change in lesion characteristics from the findings at 20 months.

Morphometric Findings

The changes in the total wall mass in response to the atherogenic diet are shown in Table 3 for the rhesus and cynomolgus monkeys. After a medium-term atherogenic diet (about 1.5 years), there was marked intimal thickening in both species. After the long-term atherogenic diet (about 6 years in rhesus and about 4 years in cynomolgus monkeys), there was further intimal thickening. In contrast, the total medial mass showed little change in either species, even after the long-term atherogenic diet.

Because medial mass is stable, the ratio of the intimal mass to the medial mass may be used to compare the relative lesion size in the four arterial segments (Figure 2). The values for the relative le-
Lesion Size

The ratios of intimal mass to medial mass in atherosclerotic monkeys are shown after the medium-term diet period (stippled bars) and the long-term diet period (open bars). The values are medians at the four preselected sites: proximal and midsite iliac and femoral arteries, *p < 0.05, long-term period vs medium-term period, by the rank-sum test.

Figure 2. The ratios of intimal mass to medial mass in atherosclerotic monkeys are shown after the medium-term diet period (stippled bars) and the long-term diet period (open bars). The values are medians at the four preselected sites: proximal and midsite iliac and femoral arteries, *p < 0.05, long-term period vs medium-term period, by the rank-sum test.

Lesion Shape

An index of the shape of intimal lesions is summarized in Figure 3. "Shape" is the ratio of maximal lesion thickness to average thickness; an increased ratio indicates nonuniform thickness (eccentricity). After the medium-term diet (about 1.5 years), the intimal lesions were significantly eccentric in rhesus, but not in cynomolgus, monkeys at all arterial sites. Because the lesions in rhesus monkeys were smaller than those in cynomolgus monkeys after the medium-term diet (Table 3), some of the difference may be due to the fact that smaller lesions tend to have higher shape ratios. After the longer diet intervals, there were no significant differences in the shape ratios of intimal lesions of rhesus and cynomolgus monkeys. Thus, the more advanced lesions in limb arteries were concentric.

Focal Medial Atrophy

To obtain quantitative evidence of focal medial atrophy in the presence of normol overall medial mass (Table 3), we compared medial thinning in control and atherosclerotic vessels. After a medium-length atherogenic diet (about 1.5 years), there was an inconsistent increase in focal medial thinning (Figure 4). The average degree of pathologic thinning (thinning that exceeded the variation in controls) was about 15% for all sites after a medium-term atherogenic diet. The focal absence of media, which was never seen in controls, was noted in a few sections from atherosclerotic animals of both species. After an atherogenic diet of longer duration, both rhesus and cynomolgus monkeys showed more severe focal medial atrophy (Figure 4). The average pathologic thinning after the long-term atherogenic diet was about 40% to 50%, which is evidence of marked pathologic change.

Effect of Perfusion Fixation on Medial Thinning

A type of focal medial thinning seen only in pressure-fixed arteries has been described in human...
Table 3. Arterial Wall Mass (as Area) in Rhesus and Cynomolgus Monkeys

<table>
<thead>
<tr>
<th>Site</th>
<th>Medium-term</th>
<th>Long-term</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Atherogenic</td>
</tr>
<tr>
<td>Iliac artery</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proximal</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intima</td>
<td>0.13 ± 0.04</td>
<td>0.64 ± 0.14*</td>
</tr>
<tr>
<td>Media</td>
<td>1.60 ± 0.11</td>
<td>1.38 ± 0.06</td>
</tr>
<tr>
<td>Mid</td>
<td>0.91 ± 0.01</td>
<td>0.42 ± 0.11*</td>
</tr>
<tr>
<td>Media</td>
<td>1.14 ± 0.09</td>
<td>1.19 ± 0.06</td>
</tr>
<tr>
<td>Femoral artery</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proximal</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intima</td>
<td>0.01 ± 0.01</td>
<td>0.63 ± 0.12*</td>
</tr>
<tr>
<td>Media</td>
<td>1.03 ± 0.05</td>
<td>1.24 ± 0.06</td>
</tr>
<tr>
<td>Mid</td>
<td>0.01 ± 0.01</td>
<td>0.35 ± 0.09*</td>
</tr>
<tr>
<td>Media</td>
<td>0.97 ± 0.05</td>
<td>1.02 ± 0.04</td>
</tr>
<tr>
<td>Cynomolgus monkeys</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Iliac artery</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proximal</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intima</td>
<td>0</td>
<td>1.76 ± 0.16</td>
</tr>
<tr>
<td>Media</td>
<td>1.35 ± 0.09</td>
<td>1.10 ± 0.07</td>
</tr>
<tr>
<td>Mid</td>
<td>0</td>
<td>1.56 ± 0.20</td>
</tr>
<tr>
<td>Media</td>
<td>0.87 ± 0.04</td>
<td>0.99 ± 0.07</td>
</tr>
<tr>
<td>Femoral artery</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proximal</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intima</td>
<td>0</td>
<td>1.20 ± 0.11</td>
</tr>
<tr>
<td>Media</td>
<td>0.99 ± 0.07</td>
<td>1.08 ± 0.09</td>
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<tr>
<td>Mid</td>
<td>0</td>
<td>0.57 ± 0.07</td>
</tr>
<tr>
<td>Media</td>
<td>0.69 ± 0.03</td>
<td>0.89 ± 0.07</td>
</tr>
</tbody>
</table>

Values are means ± SEM of area expressed as mm²

* p < 0.05 compared to control values.
† p < 0.05 compared to values at 18 months of atherogenic diet.

We looked for this thinning in response to pressure by comparing the minimal medial widths in pressure-fixed and in immersion-fixed arteries of cynomolgus monkeys given long-term diets. We compared monkeys from matched groups. The minimal medial widths in pressure-fixed and immersion-fixed arteries are shown in Figure 5. In pressure-fixed control arteries the minimal widths were only 6% less than those measured after immersion fixation; however, the minimal widths were 41% less in atherosclerotic arteries fixed by pressure rather than those fixed by immersion (p < 0.05). Thus, the media of atherosclerotic arteries may differ markedly from the media of controls in the local thinning response to pressure.

Lumen Size
Changes in lumen size are shown in Figure 6. After the medium-term diets, there was no consistent reduction in lumen size despite increases in intimal thickness. After longer periods there was a tendency...


Figure 6. Lumen size (mm²) at the four standardized sites in the iliofemoral bed. The data points are means ± SEM in control (●) and atherosclerotic (○) monkeys after the medium-term diet period (left), the long-term period (center), and the long-term period in pressure-fixed vessels (right).

for the lumen to increase rather than decrease in size, agreeing with recently reported data. This was true in both immersion-fixed and in pressure-fixed arteries (Figure 6, lower right).

Dimensions of the Atherosclerotic Artery
The total cross-sectional area of atherosclerotic arteries was greater than that of control arteries. Three factors accounted for the increase in size: the lumen size was generally unchanged or even increased, the intimal mass was increased, and the overall medial mass did not decrease despite focal medial atrophy. The average dimensions of the control and atherosclerotic segments are illustrated in Figure 7.

Hemodynamic Studies
Pressure-Flow Relationships in Atherosclerosis
Iliac perfusion pressure, and thus limb vascular resistance, tended to be higher in atherosclerotic monkeys studied after both diet periods than in controls (Figure 8). Resistance did not increase further in animals given the long-term atherogenic diet compared to those given medium-term diets. The average increase in limb vascular resistance in atherosclerotic monkeys was 22%. The pressure gradient from iliac to dorsal pedal arteries and the corresponding resistance of the large artery segment was also higher in atherosclerotic vessels than in controls (Figure 9).

Vasoconstrictor Responses
These responses were not impaired in atherosclerotic monkeys. The responses to norepinephrine, phenylephrine, and angiotensin were similar (p > 0.05) in control and atherosclerotic rhesus monkeys (Figure 10, top). Studies of the norepinephrine responses in all other diet groups showed similar responses in control and atherosclerotic monkeys (data not shown). Responses to neural stimuli were also similar in control and atherosclerotic monkeys (Figure 10, bottom). Sympathetic stimulation, the chemoreceptor reflex, and bilateral carotid occlusion produced similar vasoconstrictor responses in the two groups. It should be noted, however, that the magnitude of the stimulus during carotid occlusion
was less in atherosclerotic than in normal monkeys. The decrease in carotid pressure during carotid occlusion was 38 ± 8 mm Hg in atherosclerotic monkeys and 59 ± 14 mm Hg in control monkeys at 15 seconds and 39 ± 5 in atherosclerotic monkeys and 50 ± 6 mm Hg in control monkeys at 30 seconds (p < 0.05).

Thus, adrenergic vasoconstrictor responses were preserved in atherosclerotic monkeys. This finding indicates that the vascular bed was functionally intact.

**Vasodilator Responses**

These responses of the limb arteries were impaired in the atherosclerotic monkeys (Figure 11). The pressure-flow curves were shifted upward on the pressure axis during maximal vasodilatation. Decreased dilator responses were seen after both medium-term and long-term periods of atherogenic diet. The average pressure during maximal vasodilatation was 14 mm Hg higher in atherosclerotic monkeys (p < 0.05).
Two hemodynamic changes were seen in atherosclerotic vessels in both species: 1) Total vascular resistance was increased and vasodilator responses were impaired, even though lumen size was not decreased. 2) Vasoconstrictor responses were intact. This suggests that the segments of the arterial bed necessary for these responses were functionally intact.

These findings suggest that primate atherogenesis includes a sequence of events in the arterial wall that postpones critical narrowing of the lumen and preserves vascular responses. We will discuss: 1) potential mechanisms by which lumen size is preserved in atherosclerosis; 2) the nature of hemodynamic changes in prestenotic atherosclerosis; 3) the significance of preservation of medial responsiveness, especially with reference to focal atrophy and evidence of viscoelastic medial changes; and 4) consideration of the concept that prestenotic atherosclerotic changes in artery size and shape may be interpreted as compensatory rather than degenerative.

Maintenance of Lumen Size

In primate atherosclerosis, lumen size is maintained despite pronounced intimal growth. Although the ratio of the intima to lumen increases the absolute lumen size does not as a rule decrease in experimental primate atherosclerosis. This fundamental observation, first described under controlled experimental conditions by Bond et al. has been confirmed and the present study shows the maintenance of lumen size after years of an atherogenic diet. This maintenance of lumen size is possible because both media and intima yield to hydraulic forces to form an artery of greater total circumference. Thus, a near-normal lumen is achieved as long as the intimal lesions enlarge in an outward direction with a subsequent reshaping of the media into a thinner tissue enclosing a thickened intima. Damage to the media may not be a necessary precondition, as has been suggested. The viscoelastic changes may contribute importantly to the medial thinning at the sites of increased lesions (Figure 5). The maintenance of lumen size in the atherosclerotic vessels also suggests that the thickened intima does not have a rigid inner boundary, despite considerable fibrosis.

Early Hemodynamic Changes in Atherosclerosis

After both the medium-term and the long-term atherogenic diets, vascular resistance was increased in atherosclerosis. Increases in vascular resistance will occur with decreases in lumen size, with loss of kinetic energy if the flow becomes nonlaminar, and with increases in viscosity. Increased resistance was found not only in the resting state but also during maximal vasodilation when neurohumoral influences are suppressed and only structural differences are thought to be reflected in resistance. Impaired vaso-

Figure 11. Pressure-flow curves of the hindlimb bed during maximal vasodilation in control (●) and atherosclerotic (○) monkeys. Values are means ± SEM. *p < 0.05, control vs atherosclerotic monkeys.

Discussion

We have evaluated the structural and hemodynamic changes in hindlimb arteries during induced atherosclerosis in two primate species, rhesus and cynomolgus monkeys, and found the structural and hemodynamic responses of the two species to be similar.

There were five major structural responses of the vessel walls to the atherogenic diet: 1) Intimal thickening, the hallmark of the atherogenic response, was readily induced. The amount of intimal thickening tended to vary with the duration of atherogenic diet; intimal thickening was greater in rhesus monkeys after the long-term diet, and tended to be greater in cynomolgus monkeys, than after medium-term diets of about 1.5 years. 2) The lumen size of the atherosclerotic arteries was not less than the lumen size of controls, despite the increase in intimal mass. Thus, intimal thickening during atherosclerosis did not encroach on the lumen. 3) Overall medial mass was maintained in atherosclerosis. Diffuse medial thinning occurred, however, because the media surrounded an enlarging intima. 4) There were marked focal decreases in medial thickness because of focal medial atrophy. 5) There was significant pressure-related focal thinning of the media which was studied only in cynomolgus monkeys. This finding suggests a viscoelastic medial change in atherosclerosis.
dilator responses caused by structural changes in large arteries that limit dilation of the distal bed are characteristic of atherosclerosis. Because lumen size was maintained in atherosclerosis (Figure 6), the increases in vascular resistance could not be attributed to luminal stenosis. The trend toward higher resistance in atherosclerotic large artery segments compared to controls (Figure 9) nonetheless suggests that part of the increased resistance is within diseased segments. Thus, although the resistive effect of a narrowed cross-sectional luminal area is not present, we speculate that kinetic energy losses from nonlaminar flow and the greater viscosity of hypercholesterolemic blood may contribute to the higher pressure required in atherosclerotic vessels for a given flow.

**Medial Preservation**

The medial width was reduced by outward displacement of an enlarging intima, but the overall medial mass of hindlimb atherosclerotic arteries was not decreased in cross-sectional measurements. Because atherosclerotic arteries elongate, it is possible that the total medial mass may, in fact, increase modestly in experimental atherosclerosis.

We have considered the functional implications of the medial changes. Marked increases in large artery constrictor responses to serotonin are found in cynomolgus monkeys given a long-term atherogenic diet. Thus, evaluation of vasoconstrictor responses indicates that medial function is not reduced by atherosclerosis.

We also found focal medial thinning in atherosclerosis. The focal changes in the media in atherosclerosis are of several types. True focal reduction of medial tissue as described initially by Thomas is the most common. There may also be an arteritic invasion of the media and replacement fibrosis. All these changes were observed in this study. Although these changes imply diminished function, the finding that large artery constrictor responses are actually enhanced is evidence that focal medial atrophy or loss does not restrict the hemodynamic function of large arteries in this stage of experimental atherosclerosis.

We suggest, however, that in atherosclerosis a change may occur in the viscoelastic properties of the media that permits a change in shape with continued medial responsiveness. Crawford and Levine described the focal medial thinning that appeared under plaques in human iliac arteries in response to pressure fixation. This finding has been classified as a form of atrophy, but no loss of media is evident. It seems more likely that a change in viscoelastic properties was detected after pressure fixation. To determine whether our data in primates were compatible with the concept of medial viscoelastic change in atherosclerotic arteries, we examined medial thinning in pressure-perfused and non-perfused arteries (Figure 5). Pressure perfusion caused a significant increase in medial thinning in atherosclerotic animals but not in controls. We propose that these focal changes may indicate a more general change in viscoelastic properties. Such a change would be compatible with the remodeling of the media in atherosclerotic vessels into a thinner tissue enclosing an enlarged cross-sectional area.

**Is the Arterial Shape Change in Atherosclerosis Compensatory or Degenerative?**

In these studies of primate atherosclerosis, we have confirmed the sustained maintenance of lumen size despite massive intimal thickening. Vascular resistance remained only minimally elevated after years of an atherogenic diet. Extensive morphologic changes occurred in the media with focal atrophy and generalized thinning, but both overall medial mass and medial function were nonetheless preserved. We suggest that the increase in the cross-sectional area of the atherosclerotic artery is largely compensatory, an adaptation to intimal thickening that preserves conduit function. The prestenotic phase of atherosclerosis may thus include major adaptive responses of the arterial wall that permit long intervals of minimal vascular dysfunction, despite marked structural changes in the wall before further lesion growth causes occlusive luminal change or loss of medial function and ectasia.

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