Quantitative morphologic study of intimal thickening at
the human carotid bifurcation: II. The compensatory
enlargement response and the role of the intima
in tensile support

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Abstract

Arteries enlarge where intimal plaques form, tending to preserve lumen cross sectional area but causing an increase in mural tangential tension due to the increase in radius. To characterize the compensatory enlargement process at the carotid bifurcation and to evaluate the possible contribution of intima thickening to mural tensile support during the enlarging process, we assessed the relationships among intimal thickening, artery size and estimated tensile stress at 9 sequential axial levels in 42 human carotid bifurcations obtained during post-mortem examinations of 36 adults with no clinical or anatomical evidence of cerebrovascular disease. Right and left bifurcations were available for 6 patients. The arteries were fixed under conditions of controlled pressure distention and histologic sections were prepared at 0.5 cm axial intervals. We determined vessel radius \( r \), intima thickness \( IT \), media thickness \( MT \), intima area \( IA \), lumen area \( LuA \) and the area encompassed by the internal elastic lamina (IELA), i.e. the lumen area if there were no intimal thickening. Although \( IT, IA \) and \( r \) were greatest in the proximal sinus region, there was a highly significant linear relationship between \( IA \) and \( IELA \) at each axial level; correlation coefficients ranged from 0.64 to 0.97 with \( P < 0.001 \) at each level. Stenosis \( (IA \times IELA \times 100) \) ranged from 10.8 ± 8.0% at the common carotid level immediately proximal to the bifurcation angle to 22.3 ± 17.9% at the level immediately distal to the angle, but \( LuA \) remained nearly constant at each level regardless of \( IA \). When wall tensile stress \( TS \) was computed using only media thickness \( (MT) \), values ranged from 8.2 ± 2.5 to 17 ± 7.0 \( \times 10^5 \) dyn/cm\(^2\) \((1 \text{ dyn} = 10^{-5} \text{ N})\) with the maximum occurring at the proximal sinus. When, however, total wall thickness \( (IT + MT) \) was used, \( TS \) was similar at all levels with a mean of 6.5 ± 2.5 \( \times 10^5 \) dyn/cm\(^2\), \((\text{range, 5.5 ± 1.8 to 7.3 ± 2.7} \times 10^5 \text{ dyn/cm}^2)\) with no significant differences among the 9 levels, including those with little or no intimal thickening. We conclude that during stages of minimal or moderate intimal thickening and plaque formation at the carotid bifurcation, artery size \( (IELA) \) at each level increases with \( IA \), thereby preserving \( LuA \). The sum of intima and media thickness corresponds to the increase in radius such that estimated tangential mural tensile stress remains normal at each level.

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1. Introduction

Intimal thickening, including the development of manifest human atherosclerotic plaques, is associated with vessel enlargement [1-3]. This compensatory phenomenon tends to preserve lumen cross sectional area, but little is known concerning the nature of the mural adjustments to the increase in wall tension which corresponds to the altered radius under these conditions. In view of the cell and matrix fiber content of non-atherosclerotic intimal thickening and of the matrix content and stratified organization of atherosclerotic plaques, it is not unreasonable to consider a possible contribution of the thickened intima to the tensile support of the artery wall, particularly at stages when lesions are uncomplicated and artery enlargement is prominent. As part of an ongoing study of intimal thickening, plaque formation and plaque complication at the carotid bifurcation [4-7], we have utilized carotid bifurcations with relatively simple non-obstructive, uncomplicated intimal thickenings and plaques to assess this possibility.

Forty-two bifurcations were obtained at autopsy from patients with no clinical history or anatomic evidence of cerebrovascular disease. We analysed axial sections of pressure-fixed specimens at 9 sequential levels to determine the extent to which vessel enlargement is associated with intimal thickening, including plaque formation, at each level and whether the intimal thickening associated with vessel enlargement could contribute to the maintenance of baseline tensile stress levels, i.e. levels which prevail where intimal thickening and vessel enlargement are minimal or absent.

2. Materials and methods

Carotid bifurcations consisting of a segment of the common carotid artery, and the contiguous internal and external carotid branches were removed at autopsy from 36 individuals who died at the University of Chicago Medical Center. Both right and left bifurcations were obtained from 6 of the patients, such that a total of 42 bifurcations were studied, 23 right and 19 left. Each specimen included a segment of the internal carotid artery beyond the sinus. Arteries were selected for the study only from patients with no previous clinical history or anatomic evidence of cerebrovascular disease. The ages of the patients ranged from 27 to 91 years with a mean of 62 ± 14. Two-thirds of the patients were between 50 and 80 years of age.

Specimens were stored unopened in cold physiologic saline solution until fixation from 2 to 4 h after excision. At the time of fixation cannulas were tied into the transected ends of each specimen. After flushing with physiologic saline solution to remove blood clots, each specimen was attached to a pressure perfusion system, distended with 10% buffered formalin at 37°C and immersed in a container containing the fixative utilizing procedures described in detail elsewhere [4]. Fixation as carried out for 90 min with intraluminal pressure maintained and monitored at 100 mmHg. Under these conditions the vessels are rendered rigid and there is no discernable change in dimensions or geometric configuration when the distending pressure is released. Fixed specimens were flushed free of fixative and the lumen filled with a radio-opaque mixture of barium sulfate in molten gelatin at 37°C with intralumenal pressure maintained at 100 mmHg. The specimens were then immersed in fixative at 4°C to solidify the barium sulfate-gelatin mixture. This procedure permitted radiographic study of the lumen in several projections and preservation of vessel configuration during processing for histologic study. Before sectioning, the position of the wall opposite the flow divider was indexed by placing a continuous axial row of sutures in the adventitia of the walls of the common and internal carotid branches opposite the flow divider.

Each bifurcation was then sectioned into transverse blocks at 0.5-cm intervals perpendicular to the axial direction of the common or internal carotid segments (Fig. 1). Blocks were numbered in sequence away from the branch angle; up to 4
blocks were obtained from the common carotid (C1 to C4) and up to 5 were obtained from the internal carotid artery (I1 to I5). Depending on the dimensions of individual specimens, I1 was usually at the proximal sinus, I2 and/or I3 were at the mid-sinus, I4 was generally at the distal sinus and I5 was distal to the sinus. The samples were embedded in paraffin, sectioned at 5 \( \mu m \) and stained with hematoxylin and eosin and with the Weigert von Gieson and Gomori trichrome-aldehyde fuchsin methods for connective tissue fiber identification.

Images of the histologic sections were projected onto a digitizing tablet coupled to a computer and, for each, the contours of the lumen, internal elastic lamina and outer media were traced \([4,8,9]\). Data were processed to display the contours and to compute vessel, radius (\( r \)), lumen area (LuA), intima area (IA) and the area encompassed by the internal elastic lamina (IELA). Intima thickness (IT) and media thickness (MT) were computed both as the average at each level and at 24 polar sectors about the circumference of each vessel. IELA was considered to be the potential lumen area, i.e. the lumen area if there were no intimal thickening. Thus, the degree of cross sectional lumen stenosis was expressed as the percent of the IELA occupied by IA. Tensile stress (TS) was calculated from the modified law of Laplace, according to the expression \( TS = P r/d \) where \( P \) was the distending pressure in dyn/cm\(^2\), \( r \) the radius and \( d \) the wall thickness in centimeters (1 dyn = 10\(^{-5}\) N). TS was computed using the media thickness and also using total wall thickness, i.e. media plus intima. Results are expressed as means \( \pm \) standard error of the mean. Statistical analyses included Student's \( t \)-test or Welch's \( t \)-test for comparisons among the levels of section, while relationships of variables were studied by regression analysis. Significance was assumed if \( P \) was less than 0.05.

3. Results

There was considerable variation in overall size of the bifurcation region, so that sections could be obtained at levels C1, I1 and I2 for all 42 specimens and at levels C2 and I3 from 39 and 40 of the specimens, respectively. C3 could be obtained from 37 bifurcations and I4 from 21. C4 and I5 were available for only 11 specimens each. The lumen outline of each section of each bifurcation was circular or slightly oval (Fig. 2). All but 3 of the 285 sections had regular lumen surface contours at each level. The few exceptional specimens had complex atherosclerotic plaques which caused focal irregular contours at level I1 in 2 and at level I2 in 1. The distribution of sections at each level and the determinations for LuA, IA, IELA and percent stenosis are given in Table 1. LuA, IA and IELA were greatest in the immediate vicinity of the bifurcation, (at C1, I1 and I2). IA and IELA were greatest at level I1 and decreased with distance from the bifurcation angle both proximally and distally, particularly for the internal carotid sections. Percent cross sectional stenosis was also greatest at the proximal sinus level (I1) and tended to decrease distally and proximally.

The parallel changes in IA and IELA resulted in significant positive correlations between these
values at all of the internal carotid levels and at all of the common carotid levels but C4, where IA was minimal. IELA is plotted against IA for each of the levels in Fig. 3. Regression of IELA on IA was linear at each sample level. Values of \( R \) ranged from 0.64 at I1, where IA was \( 9.6 \pm 12.3 \text{ mm}^2 \), to 0.97 at I5 where IA was \( 4.1 \pm 3.8 \text{ mm}^2 \) (\( P < 0.001 \) for each of the section levels). There was, however, no significant correlation between LuA and percent stenosis at any level. Of the 285 sections studied, 278 (97.5%) had less than 40% stenosis. Average stenosis ranged from \( 11.8 \pm 7.9\% \) at C3 to \( 22.3 \pm 17.9\% \) at the proximal sinus level (I1), but LuA was not markedly reduced by IA at any level. In Fig. 4 LuA is plotted against percent stenosis for all sections at level C1. Regardless of intimal area or percent stenosis, LuA does not fall below 10 mm\(^2\). Results were similar for each level.

Mean intima thickness (IT), media thickness (MT) and tangential tensile wall stress (TS) for each level are given in Table 2. IT was greatest at the proximal internal carotid level (I1) and tended to decrease with distance from the bifurcation angle for both the common and internal carotid levels, but the range and mean were similar for both vessels. In contrast, MT was markedly dif-

**Table 1**

<table>
<thead>
<tr>
<th>Bifurcation level(^a)</th>
<th>No. sections</th>
<th>Lumen area (mm(^2))</th>
<th>Intima area (mm(^2))</th>
<th>IEL area(^b) (mm(^2))</th>
<th>Stenosis(^c) (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I-5</td>
<td>11</td>
<td>14.6 ± 1.4</td>
<td>4.1 ± 3.8</td>
<td>18.7 ± 4.7</td>
<td>19.3 ± 11.9</td>
</tr>
<tr>
<td>I-4</td>
<td>21</td>
<td>15.8 ± 5.0</td>
<td>3.9 ± 5.5</td>
<td>19.7 ± 7.5</td>
<td>15.8 ± 15.8</td>
</tr>
<tr>
<td>I-3</td>
<td>40</td>
<td>18.1 ± 8.1</td>
<td>3.8 ± 5.6</td>
<td>21.8 ± 11.2</td>
<td>13.5 ± 16.6</td>
</tr>
<tr>
<td>I-2</td>
<td>42</td>
<td>24.7 ± 12.3</td>
<td>7.1 ± 9.9</td>
<td>31.7 ± 17.1</td>
<td>17.7 ± 17.0</td>
</tr>
<tr>
<td>I-1</td>
<td>42</td>
<td>28.9 ± 14.4</td>
<td>9.6 ± 12.3</td>
<td>38.5 ± 18.8</td>
<td>22.3 ± 17.9</td>
</tr>
<tr>
<td>C-1</td>
<td>42</td>
<td>29.3 ± 9.6</td>
<td>6.4 ± 5.3</td>
<td>37.0 ± 14.3</td>
<td>15.1 ± 9.0</td>
</tr>
<tr>
<td>C-2</td>
<td>39</td>
<td>27.5 ± 8.9</td>
<td>4.2 ± 3.2</td>
<td>31.6 ± 10.7</td>
<td>12.4 ± 9.0</td>
</tr>
<tr>
<td>C-3</td>
<td>37</td>
<td>25.6 ± 7.5</td>
<td>3.7 ± 3.1</td>
<td>29.7 ± 9.3</td>
<td>11.8 ± 7.9</td>
</tr>
<tr>
<td>C-4</td>
<td>11</td>
<td>23.5 ± 6.0</td>
<td>2.6 ± 1.5</td>
<td>26.1 ± 5.6</td>
<td>10.8 ± 8.0</td>
</tr>
</tbody>
</table>

\(^a\)Bifurcation levels are numbered in sequence from bifurcation level. I levels refer to sections of internal carotid artery, C levels refer to sections of common carotid artery.

\(^b\)IEL area, area encompassed by the internal elastic lamina.

\(^c\)Stenosis, IA/IELA × 100.
The relationship is linear and significant at each level ($P < 0.001$). Correlation coefficients range from 0.65 to 0.97.

Fig. 3. The area encompassed by the internal elastic lamina (IEL area) is plotted against the intima area for each level of section. The relationship is linear and significant at each level ($P < 0.001$). Correlation coefficients range from 0.65 to 0.97.

Different for the internal carotid and common carotid levels. MT at the common carotid levels was nearly twice that measured at the internal carotid levels ($P < 0.001$) regardless of IT or IA, but there was, however, no difference in MT among the internal carotid levels, nor among the common carotid levels. Calculated levels of tangential mural tensile stress (TS) are shown in Table 2 using MT alone and using the sum of MT and IT for wall thickness. When only MT is used, a wide range of TS values was noted (8.2 $\pm$ 1.9 $\times$ 10$^5$ to 17.3 $\pm$ 7.0 $\times$ 10$^5$ dyn/cm$^2$), with the maximum value oc-
Fig. 4. A plot of lumen area against percent stenosis for all sections at level C1. The relationship is not statistically significant and the lumen area does not fall below 10 mm² regardless of the percent cross sectional stenosis. Similar findings prevail at each section level.

4. Discussion

In recent studies, we have attempted to provide information concerning the configuration and composition of the vessels comprising the human carotid bifurcation, particularly in relation to the distribution of intimal thickening and atherosclerosis before the onset of the complications of ulceration and thrombosis. In an initial series of studies [4,5] we showed that intimal thickening and plaques form mainly opposite the flow divider in the sinus of the internal carotid artery and in the immediately proximal adjacent common carotid artery segment. Using scale models, these regions were shown to be sites of low and oscillating wall shear stress. Where shear stress was relatively high and unidirectional, the vessels were largely spared. In order to establish overall relationships between wall shear stress and plaque localization a limited number of sections were taken, one each at the subjacent common carotid artery, the proximal sinus, the mid sinus, and at the internal carotid distal to the sinus. The present group of studies was undertaken to ascertain if the helical flow patterns evident in the scale models [4,5] corresponded at levels 11 (P < 0.05 compared with 15; P < 0.01 compared with 12; P < 0.001 compared with 13 and 14 and compared with C1, C2, C3 and C4). At level 11, IT and IA are greatest and MT is least. When, however, IT plus MT is used as the value for MT, the range of TS values is reduced to between 5.5 ± 1.8 and 7.3 ± 2.7 x 10⁵ dyn/cm² (mean, 6.5 ± 2.5) and there is no significant difference among the section levels.

### Table 2
Tensile stress at each section level

<table>
<thead>
<tr>
<th>Bifurcation level</th>
<th>Intima thickness (mm)</th>
<th>Media thickness (mm)</th>
<th>Tangential wall stress</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>TSₘ a (dyn/cm² x 10⁵)</td>
</tr>
<tr>
<td>I-5</td>
<td>0.273 ± 0.221</td>
<td>0.323 ± 0.130</td>
<td>12.3 ± 6.2</td>
</tr>
<tr>
<td>I-4</td>
<td>0.235 ± 0.301</td>
<td>0.318 ± 0.116</td>
<td>11.4 ± 3.1</td>
</tr>
<tr>
<td>I-3</td>
<td>0.197 ± 0.284</td>
<td>0.319 ± 0.103</td>
<td>12.2 ± 5.2</td>
</tr>
<tr>
<td>I-2</td>
<td>0.370 ± 0.579</td>
<td>0.321 ± 0.107</td>
<td>14.0 ± 5.0</td>
</tr>
<tr>
<td>I-1</td>
<td>0.466 ± 0.542</td>
<td>0.302 ± 0.125</td>
<td>17.3 ± 7.0</td>
</tr>
<tr>
<td>C-1</td>
<td>0.284 ± 0.203</td>
<td>0.316 ± 0.139</td>
<td>9.7 ± 3.3</td>
</tr>
<tr>
<td>C-2</td>
<td>0.200 ± 0.159</td>
<td>0.520 ± 0.114</td>
<td>8.4 ± 2.0</td>
</tr>
<tr>
<td>C-3</td>
<td>0.191 ± 0.143</td>
<td>0.528 ± 0.120</td>
<td>8.2 ± 1.9</td>
</tr>
<tr>
<td>C-4</td>
<td>0.148 ± 0.092</td>
<td>0.507 ± 0.126</td>
<td>8.2 ± 2.5</td>
</tr>
</tbody>
</table>

Values are the means for each level.

aTSₘ, tensile stress using intima as wall thickness.
bTSₘᵦ, tensile stress using the sum of intima and media as wall thickness.
oriented to the detailed axial and circumferential distribution of intimal thickenings [10] and, for the present report, to characterize the effects of intimal thickening and plaque formation on the artery wall. For these investigations a new set of carotid bifurcations was necessary with sequential blocks of the common and internal carotid arteries taken at 9 contiguous axial levels at 0.5 cm intervals. In the first paper of the present series [10], we report that intimal thickening is indeed distributed in a manner corresponding to the previously demonstrated complex helical flow pattern [3,4]. Thickening forms toward the flow divider side in the common carotid artery at some distance from the angle of bifurcation but the maximum intimal thickness shifts along the medial side wall toward the far wall region opposite the flow divider at the angle. The intima is then thickest in the proximal sinus opposite the flow divider. A lesser degree of intimal thickening occurs distally as maximum thickness veers back toward the flow divider side at the lateral side wall. At the internal carotid distal to the sinus, intimal thickening is minimal. The findings suggested that a single direction of interrogation by ultrasonography at any level may not provide an accurate picture of the extent of plaque development, but that assessments of the wall opposite the flow divider ('far wall') at the proximal internal and immediately adjacent common carotid levels (II and C1) are likely to detect maximum intimal thickening or corresponding alteration of mechanical properties, at least for minimal and moderate uncomplicated intimal thickenings [11].

In the present communication we have provided evidence that, as in other locations [1-3], the vessels at the human carotid bifurcation enlarge in relation to intima area so that lumen cross sectional area tends to be maintained at near-normal levels for extended periods. In human coronary arteries lumen narrowing does not occur on the average until about 40% of the area encompassed by the internal elastic lamina (IELA), i.e. the potential lumen area, has been occupied by plaque [1]. Of the 285 sections which form the basis of the present study, 279 (98%) had less than 40% cross sectional stenosis and lumen area was preserved even at the II and C1 levels where intimal area and percent cross sectional stenosis were greatest. These findings indicate that similar compensatory mechanisms are operative in both the coronary and carotid vessels with this degree of disease. Otherwise stated, neither plaque cross sectional area nor intimal thickness per se are reliable indicators of lumen narrowing, at least for cross sectional stenoses of less than 40%. Conversely, a normal lumen diameter or cross sectional area does not indicate that no intimal thickenings or plaques are present.

Under normal circumstances, the thickness of the artery media decreases with distance from the heart in keeping with the decrease in radius and therefore in tangential tension [12]. Although media thickness varies with diameter, media microarchitecture of homologous arteries tends to be similar [13,14]. In the present study, all 4 levels of the common carotid had the expected elastic artery structure and were of nearly identical diameter and media thickness in keeping with the absence of branches in this segment. Corresponding to the likely flow division at the bifurcation, the mean lumen area of the internal carotid artery was 20% less than that of common carotid artery while the media thickness of the muscular-type internal carotid was 60% less than that of the elastic-type common carotid. Thus, the marked and consistent difference in media thickness between common carotid level C1 and adjacent internal carotid level II should be taken into account when lesions are detected by ultrasonographic determinations of intima-media thickness. Because of the marked difference in media thickness at the two levels, equal degrees of intima-media thickness of the carotid segment immediately proximal to the bifurcation (C1) and at the internal carotid segment just distal to the bifurcation (II) do not necessarily reflect equal degrees of intimal thickening.

Our findings also provide relatively strong support for the second hypothesis set forth at the outset of this study, i.e. that intimal thickening contributes to mural tensile support. When we estimated tensile stress on the basis of pressure, radius and media thickness values covered a wide range (8.1 ± 1.9 to 17.3 ± 7.0 x 10⁵) from level to level, the highest value occurring at the level of maximum intimal thickness at the proximal sinus...
even those which result in little or moderate in-fibers which are likely to affect the mechanical cells forming compact structures often similar in ample, are usually composed of oriented fibers and properties of the artery wall. Fibrous caps for ex-timal thickening, contain collagen and elastin with artery enlargement. Although the pathogenetic died lipid-containing plaques is not clear, it appears that at least some of the non-atheroscler-otic intimal thickenings and characteristic stratif-

It should not be surprising that the intimal thickening which appears in relation to the distribution of mechanical stresses or with plaque formation appears to compensate for the increased wall tension due to the increased diameter associated with artery enlargement. Although the pathogenetic relationship between fibrocellular, non-atheroscler-otic intimal thickenings and characteristic stratified lipid-containing plaques is not clear, it appears that at least some of the non-atherosclerotic thickenings are adaptive remodelling responses [15–19]. Furthermore, atherosclerotic plaques, even those which result in little or moderate intimal thickening, contain collagen and elastin fibers which are likely to affect the mechanical properties of the artery wall. Fibrous caps for example, are usually composed of oriented fibers and cells forming compact structures often similar in both thickness and curvature to the media of the unininvolved sector of the artery cross section [20,21]. It is therefore reasonable to expect that, under some conditions, plaque matrix elements may furnish mechanical tissue support during the organizing and modelling processes which accompany lesion evolution. Modern methods for ‘early detection’ of plaques by changes in wall composition and/or mechanical properties are based on the assumption that such relationships are usual. Structural and mechanical end-points which corre-spond to potential plaque instability at more advanced stages may be more difficult to define [6,22,23] and are evaluated in subsequent communications in this series. Identification of the mechanisms which underlie and regulate these inter-active adaptive phenomena and define their limits should provide sounder criteria for evaluating plaque progression, regression and potential instability than are available at present.

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