Abstract—Carotid artery intima-media thickness (IMT) increases with advancing age in humans. The underlying mechanism of this increase is unknown, but data from animal studies suggest that a chronic increase in local distending pressure can act as a stimulus. To test this hypothesis, we studied a total of 129 healthy normotensive, nonobese, nonsmoking men aged 18 to 77 years. Brachial systolic blood pressure (SBP) was unchanged, but carotid SBP increased progressively with age ($P<0.05$). Carotid IMT and the ratio of carotid IMT to lumen (ultrasonography) increased progressively with age ($P<0.05$). Carotid IMT was $\approx50\%$ greater in the older compared with the young men. Carotid SBP was positively related to carotid IMT ($r=0.55$, $P<0.001$). After carotid SBP was taken into account (ANCOVA), the age-related difference in carotid IMT was no longer statistically significant ($P=0.22$). We conclude that carotid IMT increases with age in healthy men in the absence of elevations in peripheral SBP. Carotid SBP increases progressively with advancing age in this population and is significantly related to the corresponding carotid wall hypertrophy. These results support the hypothesis that chronic increases in local distending pressure may be an important mechanism in the wall thickening that occurs with human aging in central elastic arteries. (Arterioscler Thromb Vasc Biol. 2001;21:82-87.)

Key Words: arterial stiffness  ■  vascular compliance  ■  smooth muscle cells

Carotid arterial wall thickness, as assessed by intima-media thickness (IMT), increases with age in adult humans.\(^1\)\(\text{-}^3\) This elevation in carotid IMT is an independent risk factor for cardiovascular disease with age.\(^2\)\(\text{-}^4\)

It has been assumed that the age-related increase in IMT primarily represents a thickening of intimal layers that results from diffusive atherosclerosis.\(^5\)\(\text{-}^7\) This is probably the case in some individuals with major risk factors, such as hypertension, diabetes, and hyperlipidemia, who demonstrate marked increases in IMT. However, a growing body of evidence indicates that the modest increase in IMT with age may not simply reflect atherosclerosis.\(^8\)\(\text{-}^9\) Indeed, recently, an age-associated increase in carotid IMT was documented in healthy normotensive adults.\(^{10}\)

A complementary theory is that the increase in carotid IMT with age in healthy adults is primarily an adaptive response of intrinsic compositional elements of the arterial wall to progressive elevations in chronic arterial blood pressure.\(^{11}\) The fact that resting brachial artery systolic and arterial pulse pressures increase with age in the general population and correlate with the age-associated elevation in carotid IMT is consistent with this idea.\(^{5,12}\)

In our previous studies of primary cardiovascular aging in highly screened healthy adults, we have observed only modest or, in some cases, no elevations in brachial systolic arterial blood pressure (SBP) over the adult age range, particularly in men.\(^{13,15}\) This suggests a dissociation between peripheral blood pressure and IMT with age in this population, calling into question the presumed role of blood pressure as the key stimulus for arterial hypertrophy.

One possibility is that because of stiffening of the large elastic arteries in the cardiothoracic circulation (arteriosclerosis), the increase in SBP in this region with age is greater than that observed in the peripheral circulation, as has been suggested previously.\(^{16}\) If true, it would seem reasonable to hypothesize that it is the age-related increase in local (central circulatory) distending pressure that may be mechanistically linked to the increase in carotid IMT. In this context, animal experiments have demonstrated that an increase in local distending pressure is a major stimulus for the hypertrophy of smooth muscle cells and the synthesis of extracellular matrix in the arterial wall.\(^{17,19}\)

However, presently, there are no data addressing these issues in humans.

Accordingly, the present study tested the hypotheses that (1) the thickening of the carotid arterial wall with age in healthy humans can be observed in the absence of elevations in peripheral SBP, and (2) if so, it is related instead to elevations in local distending pressure (ie, carotid SBP). To test these hypotheses, we measured SBP and IMT in the common carotid artery with the use of arterial applanation tonometry and high-resolution ultrasonography, respectively, in healthy men matched for brachial SBP across age.
TABLE 1. Selected Subject Characteristics

<table>
<thead>
<tr>
<th>Variable</th>
<th>Young</th>
<th>Middle-Aged</th>
<th>Older</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>43</td>
<td>43</td>
<td>43</td>
</tr>
<tr>
<td>Age, y</td>
<td>27±1</td>
<td>50±1*</td>
<td>65±1†</td>
</tr>
<tr>
<td>Height, cm</td>
<td>178±1</td>
<td>177±1</td>
<td>176±1</td>
</tr>
<tr>
<td>Body mass, kg</td>
<td>81±2</td>
<td>83±2</td>
<td>80±2</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>25.7±0.7</td>
<td>26.4±0.6</td>
<td>26.4±0.6</td>
</tr>
<tr>
<td>Body fat, %</td>
<td>18±1</td>
<td>24±1*</td>
<td>25±1*</td>
</tr>
<tr>
<td>Waist circumference, cm</td>
<td>87±2</td>
<td>95±2*</td>
<td>95±2*</td>
</tr>
<tr>
<td>Energy expenditure, kcal·kg⁻¹·d⁻¹</td>
<td>42.6±1.4</td>
<td>41.9±1.4</td>
<td>39.2±1.3</td>
</tr>
<tr>
<td>Total cholesterol, mmol/L</td>
<td>3.9±0.1</td>
<td>4.5±0.1*</td>
<td>4.4±0.1*</td>
</tr>
<tr>
<td>LDL cholesterol, mmol/L</td>
<td>2.2±0.1</td>
<td>2.6±0.1*</td>
<td>2.6±0.1*</td>
</tr>
<tr>
<td>HDL cholesterol, mmol/L</td>
<td>1.2±0.1</td>
<td>1.1±0.1</td>
<td>1.1±0.1</td>
</tr>
<tr>
<td>Glucose, mmol/L</td>
<td>5.0±0.1</td>
<td>5.3±0.2</td>
<td>5.2±0.2</td>
</tr>
<tr>
<td>Insulin, µU/mL</td>
<td>5.1±0.4</td>
<td>6.9±0.6*</td>
<td>6.6±0.5*</td>
</tr>
<tr>
<td>Fibrinogen, g/L</td>
<td>2.5±0.1</td>
<td>2.9±0.1*</td>
<td>3.2±0.1†</td>
</tr>
<tr>
<td>Blood viscosity, mPa·s</td>
<td>4.5±0.1</td>
<td>4.5±0.1</td>
<td>4.3±0.1</td>
</tr>
<tr>
<td>Shear stress, dyne/cm²</td>
<td>11.9±0.8</td>
<td>10.8±0.8</td>
<td>8.7±0.5†</td>
</tr>
<tr>
<td>Dinamap brachial SBP, mm Hg</td>
<td>114±2</td>
<td>116±2</td>
<td>117±2</td>
</tr>
<tr>
<td>Dinamap brachial DBP, mm Hg</td>
<td>63±1</td>
<td>71±2*</td>
<td>70±1*</td>
</tr>
<tr>
<td>Dinamap brachial PP, mm Hg</td>
<td>54±1</td>
<td>46±1*</td>
<td>48±2*</td>
</tr>
<tr>
<td>Dinamap brachial MABP, mm Hg</td>
<td>81±1</td>
<td>87±1*</td>
<td>87±1*</td>
</tr>
<tr>
<td>RZ brachial SBP, mm Hg</td>
<td>112±2</td>
<td>115±2</td>
<td>117±2</td>
</tr>
<tr>
<td>RZ brachial DBP, mm Hg</td>
<td>66±1</td>
<td>75±1*</td>
<td>71±1*</td>
</tr>
<tr>
<td>Carotid SBP, mm Hg</td>
<td>100±2</td>
<td>111±2*</td>
<td>117±2†</td>
</tr>
<tr>
<td>Carotid DBP, mm Hg</td>
<td>63±1</td>
<td>71±2</td>
<td>70±1</td>
</tr>
<tr>
<td>Carotid PP, mm Hg</td>
<td>37±2</td>
<td>39±2</td>
<td>42±2</td>
</tr>
</tbody>
</table>

Values are mean±SE. DBP indicates diastolic blood pressure; PP, pulse pressure; MABP, mean arterial blood pressure; and RZ, random zero sphygmomanometer.

*P<0.05 vs young; †P<0.05 vs middle-aged.

Methods

Subjects

A total of 129 healthy men were studied. Subjects were grouped in consecutive 20-year age ranges starting from 18 years as follows: young, 18 to 37 years; middle-aged, 38 to 57 years; and older, 58 to 77 years (Table 1). Subjects were either sedentary or recreationally active. All subjects were normotensive (<140/90 mm Hg), nonobese, and free of overt chronic diseases as assessed by medical history, physical examination, and blood chemistries and hematological evaluation (eg, plasma glucose concentration <140 mg/dL and total cholesterol <240 mg/dL). Men aged >40 years were further evaluated by ECG at rest and ECG and blood pressure responses to incremental treadmill exercise performed to exhaustion.26 Candidates who had smoked in the past 4 years, who were taking medications, or who demonstrated significant intimal thickening, plaque formation, and/or characteristics of atherosclerosis were excluded. All subjects gave their written informed consent to participate. All procedures were reviewed and approved by the Human Research Committee of the University of Colorado.

Measurements

All measurements were performed while abstaining from caffeine and after a fast of at least 4 hours (a 12-hour overnight fast for determination of metabolic risk factors and blood viscosity). During the experimental sessions, each subject rested supine for at least 15 minutes in a quiet, temperature-controlled, semidarkened room.

Carotid Artery IMT

Carotid artery IMT was measured from the images derived from an ultrasound machine (Toshiba SSH-140) equipped with a high-resolution (7.5-MHz) linear-array transducer as originally described by Pignoli et al.3 All scans were performed by the same sonographer. The longitudinal 2D images were obtained at the proximal 1- to 2-cm straight portion of the common carotid artery. These images were recorded on a super VHS videotape recorder (Panasonic AG7350) for later offline analysis. The computer images were digitized with a video frame grabber (DT-3152, Data Translation) and stored in a PC computer.

Ultrasound carotid images were analyzed by use of computerized image analysis software as previously described.8 All image analyses were performed by the same investigator, who was blinded to the group assignment of subjects. IMT was defined as the distance from the leading edge of the lumen-intima interface to the leading edge of the media- adventitia interface.8 Lumen diameter was defined as the distance between the vessel far-wall boundary, corresponding to the interface between the lumen and intima, and a near-wall boundary, corresponding to the interface of the adventitia and media. These measurements were made at end diastole.8 At least 10 measurements of IMT and lumen diameter were taken at each segment, and the mean values were used for analysis. Plaque was considered to be present if a localized irregular thickening was at least 1.5 mm thick.21 In our laboratory, this technique has excellent day-to-day reproducibility (coefficient of variation ±1%) for the carotid IMT.

Carotid Arterial Blood Pressure

The pressure waveform and amplitude were obtained from the common carotid artery with a pencil-type probe incorporating a high-fidelity strain-gauge transducer (TCB-500, Millar Instruments) as previously described.22,23 This tonometer has been shown to register a pressure wave with harmonic content that does not differ from that of an intra-arterially recorded wave, and the use of the tonometer on an exposed artery records a waveform identical to that recorded intra-arterially.24 Because the baseline levels of carotid blood pressure are subjected to hold-down force, the pressure signal obtained by tonometry was calibrated by equating the carotid mean arterial pressure to the brachial artery measurement as previously described.23 Mean arterial blood pressure in the resting supine position does not vary significantly within the large conduit arteries.23 Systolic wall tension was calculated as the product of carotid systolic blood pressure and carotid radius at end systole. Systolic tensile stress was the ratio of systolic wall tension to the carotid IMT.

Brachial Arterial Blood Pressure

To avoid any possibility of investigator bias, peripheral arterial blood pressure was measured with a semiautomated device (Dinamap, Johnson & Johnson) over the brachial artery. Recordings were made in triplicate in the supine positions. All measurements conformed strictly to American Heart Association guidelines.26 Because of the controversy surrounding the accuracy of the semiautomated device, we also measured arterial blood pressure with a random zero sphygmomanometer (Hawksley & Sons) in the same subjects.

Blood Viscosity and Shear Stress

Blood viscosity and shear stress have been associated with elevated carotid IMT.27 Blood viscosities were measured at shear rates of 0.3 to 60 rpm at 37°C with use of a cone and plate viscometer (model DV-1+, Brookfield Engineering) as previously described.27 All measurements were performed within 2 hours after blood withdrawal. Viscosity values obtained at the highest shear rate (ie, 60 rpm) were reported. Shear stress was calculated as previously described28 by the following formula: 4π/3blood viscosity×blood flow/m², where r is the radius.

Ankle-Brachial Pressure Index

To exclude the possibility of overt peripheral artery disease, SBP of the posterior tibial artery was measured by using a Doppler flowmeter (Parks Medical) and a sphygmomanometer. The ratio of the ankle SBP to the brachial SBP (Dinamap) was taken as the ankle-arm pressure index. Peripheral artery disease was considered present when the index was <0.90.

Body Composition

Total fat mass and fat-free mass were determined by using dual energy x-ray absorptiometry (DPX-IQ, Lunar Corp) as previously
Figure 1. Brachial artery (left) and carotid artery (right) SBP in young, middle-aged, and older men. *P<0.05 vs young men; †P<0.05 vs middle-aged men.

Figure 2. Carotid artery IMT (top) and IMT/lumen ratio (bottom) in young, middle-aged, and older men. *P<0.05 vs young men; †P<0.05 vs middle-aged men.

Results

Selected subject characteristics are shown in Table 1. Body fat percentage, waist circumference, and brachial diastolic blood pressure were lower in the young compared with the middle-aged and older groups. There were no significant group differences in height, body mass, body mass index, estimated energy expenditure, or brachial SBP. Fasting plasma concentrations of total cholesterol, LDL cholesterol, insulin, and fibrinogen were performed in the clinical laboratory affiliated with the General Clinical Research Center, as previously described.

The carotid artery was greater in the middle-aged (31.5±0.7 mm Hg/mm) and older (32.3±0.8 mm Hg/mm) compared with the young (28.6±0.7 mm Hg/mm) men (P<0.01). Systolic tensile stress was lower in older (47.9±1.9 mm Hg/mm²) compared with young (60.7±2.1 mm Hg/mm²) and middle-aged (56.8±1.7 mm Hg/mm²) men (P<0.01).

As illustrated in Figure 2, carotid IMT and the carotid IMT/lumen ratio were progressively greater with age (all P<0.05). Carotid IMT was 50% greater in older compared with the young men. Carotid lumen diameter was not significantly different among the 3 groups (6.7±0.1 mm in young versus 7.0±0.1 mm in middle-aged versus 7.0±0.2 mm in older men, P=0.23).

Because many of the coronary heart disease risk factors were higher with age, we sought to exclude, as much as possible, the effects of these factors on carotid IMT. To do so, we identified subgroups of young and older subjects matched for coronary heart disease risk factors (Table 2). Carotid IMT and the IMT/lumen ratio remained significantly higher in older than in young subjects (Figure 3).

As shown in Figure 4, carotid artery SBP was positively related to carotid IMT in the pooled subject population (r=0.55, P<0.001). Significant associations also were present within the groups of young and older men (r=0.29 to 0.35, P<0.05). Moreover, when the influence of age was included in the pooled population, the association between carotid IMT and carotid SBP remained significant (r=0.40, P<0.01). Carotid pulse pressure was more modestly related to carotid IMT (r=0.24, P<0.05). In contrast, neither brachial SBP nor pulse pressure was significantly related to carotid IMT, whereas brachial diastolic blood pressure and mean arterial blood pressure were modestly related to carotid IMT (both r=0.26, P<0.05). Arterial systolic wall stress was not significantly associated with carotid IMT. Carotid IMT also was modestly associated with plasma fibrinogen (r=0.25), LDL cholesterol levels (r=0.25), and shear

Physical Activity

Estimated daily energy expenditure was assessed by the Stanford Physical Activity Questionnaire and was used as a measure of daily physical activity.

Metabolic Risk Factors

Fasting plasma concentrations of cholesterol, glucose, insulin, and fibrinogen were performed in the clinical laboratory affiliated with the General Clinical Research Center, as previously described.

Statistical Analyses

One-way ANOVA was used to assess the influence of age on variables of interest. In the case of a significant F value, a post hoc test using the Newman-Keuls method was used to identify differences among mean values. Univariate regression and correlation analyses as well as partial correlation analyses were used to determine the relationships between variables of interest. ANCOVA was used to examine the contribution of the correlated variables to age-related increases in carotid IMT. Forward stepwise multiple regression analyses were used to identify significant independent determinants for the age-related increase in carotid IMT. Only variables that had significant univariate correlations with carotid IMT were included in the model. Because blood viscosity and shear stress were obtained in only 50% of the subjects, analyses of these variables were performed on this sample of the overall study population. All data are reported as the mean±SE. Statistical significance was set a priori at P<0.05 for all comparisons.
stress \( (r = -0.33, \ p < 0.05) \), whereas no significant association was found with waist circumference.

When ANCOVA was performed with carotid SBP as the covariate, the age-related differences in carotid IMT were no longer statistically significant \( (p = 0.22) \). However, when carotid IMT was adjusted for carotid pulse pressure, brachial diastolic blood pressure, brachial mean arterial blood pressure, plasma fibrinogen, LDL cholesterol, or shear stress (ie, other variables that were significantly related to carotid IMT on the basis of univariate correlations), the age-related differences in carotid IMT were attenuated (range 1% to 20%) but remained highly significant \( (p < 0.01) \). The only variable that entered in the stepwise regression analysis was carotid SBP, accounting for 22% of the variability associated with carotid IMT \( (p < 0.001) \).

**Discussion**

The primary findings of the present study are as follows: First, carotid artery IMT increases progressively with age in healthy normotensive men in the absence of elevations in peripheral SBP. Second, carotid SBP also increases with advancing age and is positively related to the corresponding increases in carotid IMT in this population. Third, the significant increase in carotid IMT with age is abolished after statistically accounting for the corresponding increase in carotid SBP. Fourth, carotid SBP was the only independent predictor of the age-associated increase in carotid IMT. These results suggest that the age-related increase in intraluminal pressure exerting a distending force on the arterial wall may be an important mechanism contributing to arterial hypertrophy in the central elastic arteries.

In the present study, carotid IMT increased by \( \approx 50\% \) with age (young to older adulthood) in healthy men. This observation was independent of any changes in lumen diameter. We carefully screened for several major risk factors for cardiovascular disease, including smoking, elevated adiposity, and impaired lipid and carbohydrate metabolism. Importantly, brachial SBP was well within the normotensive range and was similar across the age groups. Thus, our data suggest a primary effect of aging (rather than comorbidities) in the increase in carotid artery wall thickness. These results are consistent with recent findings of increases in carotid IMT across age in healthy adults.10 However, in this previous study, brachial SBP was elevated with age, albeit within the normotensive range (ie, <140 mm Hg). Thus, our results extend these earlier findings by showing that carotid IMT increases with age in the absence of any increase in peripheral SBP or pulse pressure.

Previous observations in experimental animals indicate that sustained elevations in local distending pressure stimulate...
smooth muscle hypertrophy and synthesis of extracellular materials in the arterial wall. On the basis of these findings, we hypothesized that the greater carotid IMT with advancing age in humans may be associated with elevated carotid SBP. At least 4 lines of evidence from the present study support our hypothesis. First, mean levels of carotid IMT and carotid SBP increased progressively with age. Second, univariate correlational analyses indicate that carotid IMT was positively related to carotid SBP. Third, when ANCOVA was performed with carotid SBP as the covariate, the age-related differences in carotid IMT were no longer statistically significant. Fourth, stepwise regression analysis indicated that the only significant independent predictor of the age-related increase in carotid IMT was carotid SBP. Taken together, our findings are consistent with the hypothesis that the increase in local distending pressure with primary human aging acts to stimulate arterial hypertrophy in the large elastic arteries of the central circulation.

We have previously reported that tonic sympathetic nerve activation with age may be an important mechanism contributing to smooth muscle hypertrophy and subsequent arterial wall hypertrophy in peripheral muscular arteries (ie, the femoral artery). Although the amount of smooth muscle cells is less in the carotid compared with the femoral artery, we cannot discount the possibility that increased sympathetic tone contributed to the age-related increase in carotid IMT in the present study. Alternatively, it is possible that local distending pressure may have played a role in femoral arterial wall hypertrophy. However, because pulsatile distension is considerably smaller in the carotid compared with the femoral artery, we cannot discount the possibility that increased sympathetic tone contributed to the age-related increase in carotid IMT in the present study. Nevertheless, we cannot discount the possibility that genetic or other constitutional factors may have influenced the present findings regarding the relation between carotid IMT and carotid artery and the carotid bifurcation, which were not examined in the present study. In the general population, the carotid artery hypertrophy that occurs with primary aging may act to increase “baseline” wall thickness, from which the additional effects of adverse genetic or behavioral influences (eg, smoking, high-fat diet, and sedentary lifestyle) could cause further increases to pathological levels. Indeed, a recent longitudinal study demonstrated that most atherosclerotic lesions develop at sites with increased wall thickness. Alternatively, independent of atherosclerosis, carotid wall hypertrophy may contribute to age-associated reductions in central arterial compliance and consequent left ventricular pulsatile afterload, thereby increasing the risk of cardiovascular disease with age.

In the young men in the present study, SBP was lower in the carotid than in the brachial artery. In the older men, however, there was no central-peripheral SBP difference, primarily because of the age-related increase in carotid SBP. With advancing age, the central elastic arteries stiffen (develop arteriosclerosis), as indicated by an increase in pulse-wave velocity and earlier wave reflections. In contrast, there is no significant age-associated increase in the stiffness of the peripheral arteries. As such, the amplification of the pressure wave between central and peripheral arteries is gradually reduced with advancing age, such that carotid and brachial SBP become similar in older adults. These findings have important clinical implications. Many clinicians may assume that the traditionally emphasized peripheral-central arterial SBP difference is standard for all patient populations. Our findings indicate otherwise, suggesting that this relation changes dramatically with advancing age even in healthy adults.

In the present study, we observed no significant increases in carotid artery lumen diameter with advancing age in healthy men. This finding appears to be at odds with findings of previous population-based studies showing age-related increases in arterial lumen diameter with age. This discrepancy is likely due to the differences in the health status among subjects in each study. Indeed, it has recently been reported that carotid artery lumen diameter increases significantly with age in the population as a whole, whereas such an age-related increase is absent when only healthy adults are selected for analysis.

A major limitation of the present study is its cross-sectional study design. However, a recent epidemiological study demonstrated that the cross-sectional findings for carotid IMT are generally consistent with what is observed longitudinally. Nevertheless, we cannot discount the possibility that genetic or other constitutional factors may have influenced the present cross-sectional findings. We should also emphasize that our findings regarding the relation between carotid IMT and carotid SBP are based largely on correlational data. As such, a cause-and-effect association between these events cannot be proven from the present results. In this context, it is possible, for example, that an increase in local blood pressure may be secondary to an increase in carotid IMT.

In summary, we have shown that carotid IMT increases with advancing age in healthy humans in the absence of elevations in...
peripheral SBP. However, the increases in IMT are strongly and positively related to elevations in carotid SBP. These results support the hypothesis that chronic increases in local distending pressure may be an important mechanism in the wall thickening of central elastic arteries that occurs with human aging.

Acknowledgments

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