Impact of Gender and Menopausal Status on Relationships Between Biological Aging, as Indexed by Telomere Length, and Aortic Stiffness

Andrew R. Raymond,1 Gavin R. Norton,1 Angela J. Woodiwiss,1 and Richard L. Brooksbank1

BACKGROUND
Telomere length predicts cardiovascular disease (CVD) possibly through an impact of telomere attrition on aortic stiffness. Whether reduced biological aging and a lack of telomere length–aortic stiffness relationships in women contribute to the lower prevalence of CVD in women, prior to menopause, is uncertain.

METHODS
We evaluated the relationship between telomere length and carotid–femoral (aortic) pulse wave velocity (PWV) in 580 randomly recruited participants of Black African descent (age = 44±19 years; women: n = 361; premenopausal: n = 195). PWV was determined using carotid and femoral applanation tonometry (Sphygmocor). Relative leukocyte telomere length (T/S) was measured using quantitative real-time polymerase chain reaction assays.

RESULTS
Men and women had similar T/S. T/S was inversely correlated with age (r = −0.14, P < 0.001) and this association was similar in all (r = −0.14, P < 0.01) and premenopausal (r = −0.17, P < 0.05) women as in men (r = −0.14, P < 0.05). An inverse relationship between T/S and PWV was noted both before (r = −0.20, P < 0.001) and after (partial r = −0.14, P < 0.001) adjustments for confounders. No interaction between T/S and either sex or menopausal status was independently associated with PWV, and T/S was independently correlated with PWV in all (partial r = −0.14, P < 0.01) and premenopausal (partial r = −0.18, P < 0.05) women and in men (partial r = −0.15, P < 0.05).

CONCLUSIONS
Gender and premenopausal status do not affect age-related decreases in T/S and associations between T/S and PWV. In participants of African descent in whom telomere length did not differ by gender, the impact of gender prior to menopause on CVD is unlikely to be attributed to differences in the effect of biological aging on aortic stiffness.

Keywords: aortic stiffness; blood pressure; gender; hypertension; pulse wave velocity; telomere length.

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Telomeres are nucleoprotein structures at the end of eukaryotic chromosomes. Telomeres prevent chromosomal degradation and decrease in length progressively with each cell division.1 Telomere shortening is associated with life stress,2 and as such, telomere length reflects the biological age of an organism which may differ considerably from chronological age.3 A reduced leukocyte telomere length independently predicts the development of cardiovascular disease (CVD),4–8 associated with a number of large vessel structural changes that determine the atherosclerotic process,9–12 and as such is a marker of vascular aging.13 The relationship between telomere length and cardiovascular outcomes may in-part be attributed to an impact on aortic stiffness,14–16 a large vessel change that may contribute toward cardiovascular outcomes through an increase in central aortic, but not brachial blood pressure (BP), or through the facilitation of transmission of pulsatile energy into end-organ circulatory systems.17 Whether sex-specific age-related telomere attrition5,18 and a sex-specific impact of telomere shortening on aortic stiffness contributes toward the well-recognized sex differences in CVD prior to menopause19–22 is uncertain.

Telomeres may be longer in adult women as compared to men,14,23 an effect that could be attributed to a reduced age-related attrition of telomere length.14 Moreover, although a relationship between telomere length and aortic stiffness, as indexed by aortic pulse wave velocity (PWV), is now well described in men,14–16 in the only study conducted in women, no relationship between telomere length and aortic PWV was noted.14 Thus, it is possible that sex-specific telomere length–aortic PWV relationships contribute toward sex differences in CVD prior to menopause. However, the lack of relationship between telomere length and aortic PWV in women was reported in a study with a small sample size (n = 73)14 and hence may reflect a lack of statistical power to show such effects in this group. To determine whether telomere length–aortic stiffness relationships are indeed sex-specific or determined by menopause, in the present study, we evaluated the impact of gender and menopausal status on...
the relationship between telomere length and aortic PWV and central aortic hemodynamics in a large, randomly selected community-based sample.

METHODS

Study group

The present study was conducted according to the principles outlined in the Helsinki declaration. The Committee for Research on Human Subjects of the University of the Witwatersrand approved the protocol (approval number: M02-04-72 renewed as M07-04-69 and M12-04-108). Participants provided informed, written consent. All participants were part of a community-based study previously described. Briefly, nuclear families of black African descent, with siblings older than 16 years of age were randomly recruited from the South West Township of Johannesburg, South Africa. In a substudy, 587 sequentially recruited participants with all central aortic hemodynamics assessments available had leukocyte telomere length assessed. Of the sample, 7 participants were excluded from the analysis as telomere length measurements were extreme outliers (more than 2 SD beyond the mean).

Clinical, demographic, and anthropometric variables

A standardized questionnaire was used to obtain demographic, clinical, smoking, and alcohol intake history of the participants. Height and weight, for the determination of body mass index, were measured using standard methods, and participants were defined as obese if their body mass index was greater than 30 kg/m². The mean of 5 high quality nurse-derived BP measurements were obtained in the seated position, according to guidelines, using a standard mercury sphygmanomanometer as previously described. Hypertension was defined as the presence of antihypertensive treatment or a mean systolic BP (SBP) greater than 140 mm Hg or diastolic BP (DBP) greater than 90 mm Hg. Blood samples were obtained in the supine position after 10 minutes of rest in the morning between 10:00 and 12:00 hours. Standard laboratory blood tests for total cholesterol, high-density lipoprotein, low-density lipoprotein, and percentage glycated hemoglobin were performed. Diabetes mellitus was defined as the presence of glucose-lowering therapy and a poor glucose control as a percentage glycated hemoglobin were performed. Diabetes mellitus was defined as the presence of glucose-lowering therapy and a poor glucose control as a percentage glycated hemoglobin were performed. Diabetes mellitus was defined as the presence of glucose-lowering therapy and a poor glucose control as a percentage glycated hemoglobin were performed.

To identify possible factors that explain relations between telomere length and aortic PWV, after centrifugation, serum and plasma samples were obtained in order to measure plasma renin, angiotensinogen, aldosterone, and insulin concentrations. These samples were stored at −70 °C until the time of analysis. Plasma renin concentrations were measured using an immunoradiometric technique (Renin III Generation; Cisbio International, Ceze, France) (intra-assay coefficients of variation ranging from 1.1% for high concentrations to 4.5% for low concentrations and with a mean inter-assay coefficient of variation of 5.3%). Serum aldosterone concentrations were measured using an [125I]-radioimmunoassay (Coat-A-Count; Diagnostic Products Corporation, Los Angeles, CA) (intra-assay coefficients of variation ranging from 2.3% for high concentrations to 5.4% for low concentrations, and with a mean inter-assay coefficient of variation of 5.4%). Plasma angiotensinogen concentrations were determined using a human total angiotensinogen solid phase sandwich ELISA (Code No. 27412, Immuno-Biological Laboratories, Gunma, Japan) (intra-assay coefficients of variation ranging from 4.4% for high concentrations to 5.5% for low concentrations and with an inter-assay coefficient of variation ranging from 4.3% for high concentrations to 7.0% for low concentrations). Fasting plasma insulin concentrations were determined from an insulin immulite, solid phase, 2-site chemiluminescent immunometric assay (Diagnostic Products Corporation) and insulin resistance was estimated by the homeostasis model assessment of insulin resistance (HOMA-IR) using the formula (insulin (μU/ml) × glucose (mmol/l))/22.5.

Telomere length

Leukocyte DNA was extracted from whole blood samples using the salting out method and samples were stored at −20 °C until analyzed. Samples were stored in a Tris–EDTA buffer which minimizes DNA degradation during long-term storage. Furthermore, DNA samples were stored in aliquots from the master stock to prevent repeat freeze-thaw cycles, which further reduces the chances of DNA degradation. A quantitative real-time polymerase chain reaction method was used to determine relative telomere length of leukocyte DNA. This method determines the factor by which telomere repeat copy number (T) differs from the copy number of a single copy gene (S). The single copy gene used was the 36B4 gene (located on chromosome 12, which codes for acidic ribosomal phosphoprotein PO). This real-time quantitative polymerase chain reaction method has been validated and shown to give a measure of relative telomere length, which is proportional to mean telomere length as determined by Southern blotting. A standard sample was included in each amplification run and was used to standardize the entire sample T/S ratios. Standard curves were generated for both telomere and 36B4 reactions from a single DNA sample that was serially diluted to produce a range of DNA quantities (100–10 ng). The reaction efficiencies of 2.1 and 1.9, and the tube-to-tube variations of 0.09 and 0.001 for the 36B4 and telomere standard curves, respectively, were well within accepted limits. For the telomere and 36B4 standard curves, the slopes of the fitted regressions were −3.18 ± 0.3 and −3.75 ± 0.3 (P < 0.001 for both), respectively. The regression coefficients were 0.998 and 1.00 for the telomere and 36B4 standard curves, respectively, and the coefficients of variation of the S and T assays were 3.12% and 3.46%, respectively. The standard curves showed that there was a strong negative linear relationship between DNA concentration and crossing point. Samples that fell outside the linear concentration range of the standard curves were diluted and rerun in order to ensure they fell within the linear range of the standard curves.
Central aortic hemodynamics

Central aortic hemodynamics were determined as previously described.25–27 After participants had rested for 15 minutes in the supine position, arterial waveforms at the radial (dominant arm), carotid, and femoral artery pulses were recorded by applanation tonometry. Pressure waveforms were recorded during an 8-second period using a high-fidelity SPC-301 micromanometer (Millar Instrument, Houston, TX) interfaced with a computer employing SphygmoCor, version 9.0 software (AtCor Medical Pty, West Ryde, New South Wales, Australia). Aortic PWV was determined from sequential waveform measurements at carotid and femoral sites as previously described.27 The time delay in the pulse waves between the carotid and femoral sites was determined using an electrocardiograph-derived R wave as a fiducial point. Pulse transit time was taken as the average of 10 consecutive beats. The distance which the pulse wave travels was determined as the difference between the distance from the femoral sampling site to the suprasternal notch and the distance from the carotid sampling site to the suprasternal notch. Aortic PWV was calculated as the ratio of the distance to the transit time (m/s). To determine aortic BP, the pulse wave obtained from the radial tonometer recordings was calibrated by manual measurement (auscultation) of brachial BP taken immediately before the recordings. The radial pressure waveform was converted into a central (aortic) waveform using a validated generalized transfer function incorporated in SphygmoCor software. Central aortic SBP was derived from the aortic waveform and central aortic pulse pressure was calculated from the difference between central aortic SBP and DBP. The aortic forward wave pressure was determined from the difference between the pressure at the aortic first systolic shoulder and DBP. Aortic augmentation pressure was calculated as the difference between the second and the first systolic peak of the aortic waveform. All measurements were made by a single experienced technician unaware of the clinical history of the participants and with a low degree of intraobserver variability and a high degree of reproducibility.28–30

Statistical analysis

Statistical analysis was conducted using SAS software, version 9.3 (SAS Institute, Cary, NC). Data are represented as mean ± SD. As T/S was positively skewed (skewness = 4.91; kurtosis = 34.4, Shapiro–Wilk’s statistics = 0.59), it was log transformed which improved the distribution of the data (skewness = −0.25; kurtosis = 4.39, Shapiro–Wilk’s statistics = 0.94). As aortic PWV was also positively skewed (skewness = 1.88; kurtosis = 6.03, Shapiro–Wilk’s statistics = 0.86), it was log transformed which improved the distribution of the data (skewness = 0.30; kurtosis = 1.04, Shapiro–Wilk’s statistics = 0.98). Hence, all analyses were conducted on log-transformed T/S and log-transformed PWV. Multivariate regression analysis was performed to determine independent relationships. To ensure that the relationships identified in all women were not specifically attributed to effects produced after menopause, sensitivity analysis was conducted in premenopausal women only. As antihypertensive therapy may affect aortic PWV, sensitivity analysis was conducted in participants having never received antihypertensive therapy. Unadjusted or multivariate adjusted correlation coefficients were compared using Z-statistics.

RESULTS

Participant characteristics

Minor differences in PWV were noted between those with and those without leukocyte telomere length and all central aortic hemodynamics assessments (Supplementary Table 1). No differences in other characteristics between those with and those without leukocyte telomere length and all central aortic hemodynamics assessments available were noted (Supplementary Table 1). The characteristics of men, women, and premenopausal women in the study sample are shown in Table 1. The median log T/S values for men, women, and premenopausal women were −0.0120, 0.0060, and 0.0421, respectively. The quartile ranges for log T/S in men, women, and premenopausal women were −0.13 to 0.08, −0.13 to 0.12, and −0.10 to 0.16, respectively. No differences in telomere length were noted between men and women.

Factors associated with average relative leukocyte telomere length

There was an inverse correlation between age and T/S (Table 2) and both the strength (r values) and the magnitude (slopes or β coefficients) of the relationships were similar in all and premenopausal women as compared to men (Table 3). The similarity of the relationships noted between age and T/S in all women, premenopausal women, and men persisted with adjustments (Table 3). T/S was reduced in those who regularly consumed alcohol (Table 2). However, no relationships between T/S and other general risk factors for CVD were noted after adjustments for age and sex (Table 2). T/S was positively rather than inversely associated with aldosterone, aldosterone-to-renin ratio, and angiotensinogen (Supplementary Table 2). No relationships between T/S and HOMA-IR were noted (Supplementary Table 2).

Sex-specific relationships between average relative leukocyte telomere length and aortic PWV

On both bivariate (partial \( r = -0.20, CI = -0.27 \) to \(-0.12, P < 0.0001\)) and multivariate regression analysis with adjustments for age, sex, menopausal status, SBP, body mass index, smoking, drinking, antihypertensive treatment, and diabetes (defined as HbA1c > 6.1%) (partial \( r = -0.14, CI = -0.22 \) to \(-0.06, P < 0.001\)), relative leukocyte telomere length was associated with aortic PWV. The relationship between relative leukocyte telomere length and aortic PWV remained with further adjustments for HOMA-IR, aldosterone, the aldosterone-to-renin ratio, and angiotensinogen concentrations (partial \( r = -0.15, CI = -0.23 \) to \(-0.07, P < 0.001\)). No interaction between relative telomere length and gender (partial \( r = 0.022, P = 0.60\)) or relative telomere length
and menopausal status (partial $r = -0.017, P = 0.69$) was associated with aortic PWV after adjustment for the aforementioned confounders. The strength of the bivariate and multivariate adjusted relationships (slopes or β coefficients) between relative leukocyte telomere length and aortic PWV was similar in all women compared to men (Table 4). However, premenopausal women had a greater magnitude (β coefficients) of the relationship between relative leukocyte telomere length and aortic PWV compared to men (Table 4). Furthermore, in those not receiving antihypertensive treatment, both all women and premenopausal women had a greater magnitude (β coefficients) of the relationship between relative leukocyte telomere length and aortic PWV as compared to men (Table 4).

**Relationships between average relative leukocyte telomere length and additional aortic hemodynamic variables**

After adjustments for potential confounders (age and presence or absence of regular alcohol intake), no independent relationships were noted between T/S and either aortic SBP, aortic pulse pressure, aortic forward wave pressure, or augmentation pressure in either men or women (Table 5).

**DISCUSSION**

The main findings of the current investigation are as follows: In a relatively large, cross-sectional study conducted in a group of Black African ancestry in which telomere length did not differ by gender, age-related decreases in telomere length, an index of biological aging, were similar in men and women. Moreover, independent of chronological age, relationships between biological age, as indexed by relative leukocyte telomere length, is associated with aortic stiffness in a relatively large study sample ($n = 361$) of women; that this relationship also exists in a sample of premenopausal women ($n = 195$); and that this relationship in all and premenopausal women is equivalent in strength and greater in magnitude than the relationship that exists in a similar sample size ($n = 195$) of men. These findings noted in a group of African ancestry are in contrast to the relationships noted between relative leukocyte telomere length and aortic pulse pressure in either men or women.
Telomeres and Aortic Stiffness

Telomere length and aortic PWV in a small study sample of Caucasian men (n = 120), but not women (n = 73) previously reported on. Possible explanations for the discrepancy between the present and this prior study include differences in ethnicity or alternative demographic and clinical characteristics in women. The exact differences in demographic or clinical characteristics that explain this discrepancy are not apparent.

The results of the present study indicate that gender and premenopausal status are not independent determinants of the impact of biological aging on aortic stiffness. Hence, the present results support the notion that biological aging of large arteries may in-part be encoded in DNA as opposed to being determined by biological factors such as sex hormones.

Moreover, the present results also suggest that it is unlikely that the well-described sex differences that exist in cardiovascular outcomes prior to menopause can be attributed to sex-specific effects of biological aging, as indexed by relative leukocyte telomere length, on aortic stiffness.

One possible reason for our lack of ability to show sex-specific telomere length–aortic stiffness relationships in the present study is that with adjustments for chronological age, women had a similar biological age, as indexed by telomere length, as men. In addition, relationships between chronological and biological age, as indexed by relative telomere length, were similar in men and premenopausal women. This is in contrast to the shorter telomeres noted in men as compared to women in prior studies where telomere

### Table 2. Relationships between relative leukocyte telomere length (log T/S) and risk factors for cardiovascular disease

<table>
<thead>
<tr>
<th>Log T/S vs.</th>
<th>Continuous analysis (n = 580)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>−0.14 (−0.22 to −0.06)</td>
</tr>
<tr>
<td>Body mass index</td>
<td>−0.03 (−0.11 to 0.05)</td>
</tr>
<tr>
<td>Systolic blood pressure</td>
<td>−0.02 (−0.10 to 0.06)</td>
</tr>
<tr>
<td>Diastolic blood pressure</td>
<td>−0.02 (−0.10 to 0.06)</td>
</tr>
<tr>
<td>Total cholesterol (n = 550)</td>
<td>−0.03 (−0.12 to 0.05)</td>
</tr>
<tr>
<td>LDL cholesterol (n = 541)</td>
<td>−0.03 (−0.12 to 0.05)</td>
</tr>
<tr>
<td>Follicle-stimulating hormone</td>
<td>−0.07 (−0.18 to 0.03)</td>
</tr>
</tbody>
</table>

Abbreviations: CI, confidence interval; DM, diabetes mellitus or an HbA1c > 6.1%; LDL, low-density lipoprotein; reg., regular.

*Adjusted for age and sex.
†Determined from log T/S.

### Table 3. Impact of gender and menopausal status on relationships between age and relative leukocyte telomere length (log T/S)

<table>
<thead>
<tr>
<th>Age vs. log T/S</th>
<th>Pearson’s/partial r (95% CI)</th>
<th>β coefficient (±SE)</th>
<th>P value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bivariate relationships</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Men (n = 219)</td>
<td>−0.14 (−0.27 to −0.01)</td>
<td>−0.0016 ± 0.0008</td>
<td>0.04</td>
</tr>
<tr>
<td>Women (n = 361)</td>
<td>−0.14 (−0.24 to −0.04)</td>
<td>−0.0019 ± 0.0007</td>
<td>0.007</td>
</tr>
<tr>
<td>Premenopausal (n = 195)</td>
<td>−0.17 (−0.30 to −0.03)</td>
<td>−0.0044 ± 0.0019</td>
<td>0.02</td>
</tr>
</tbody>
</table>

*Abbreviation: CI, confidence interval.
†For relationships.
‡Adjusted for presence or absence of regular alcohol consumption. No differences were noted in Pearson’s r, partial r, or β coefficients between all or premenopausal women vs. men (P > 0.05 for all).
As substantial variations in telomere length occur between populations, it is possible that these population differences in telomere length may translate into differences in gender vs. telomere length relationships. Importantly, however, the present study suggests that previously reported gender differences in telomere length are unlikely to be attributed to hormonal differences between men and women. We suggest that alternative demographic or phenotypic differences that may occur between women and men and that characterize different populations may explain sex effects on telomere length.

The clinical implications of the present study warrant consideration. In this regard, several studies have demonstrated that leukocyte telomere length is independently associated with CVD. However, the exact mechanisms explaining

### Table 4. Impact of gender and menopausal status on relationships between relative leukocyte telomere length (log T/S) and aortic PWV (log aortic PWV)

<table>
<thead>
<tr>
<th></th>
<th>Log T/S vs. log aortic PWV</th>
<th>P value&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pearson's r (95% CI)</td>
<td>β coefficient (±SE)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Bivariate relationships</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>All participants</td>
<td>-0.20 (-0.32 to -0.06)</td>
<td>-0.292 ± 0.099</td>
</tr>
<tr>
<td>Men (n = 219)</td>
<td>-0.20 (-0.29 to -0.10)</td>
<td>-0.327 ± 0.086</td>
</tr>
<tr>
<td>Premenopausal (n = 195)</td>
<td>-0.24 (-0.37 to -0.10)</td>
<td>-0.597 ± 0.176&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Not receiving antihypertensive therapy</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Men (n = 187)</td>
<td>-0.17 (-0.31 to -0.03)</td>
<td>-0.286 ± 0.121</td>
</tr>
<tr>
<td>Women (n = 269)</td>
<td>-0.24 (-0.35 to -0.12)</td>
<td>-0.465 ± 0.115&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Premenopausal (n = 191)</td>
<td>-0.26 (-0.38 to -0.12)</td>
<td>-0.673 ± 0.184&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>Multivariate adjusted&lt;sup&gt;c&lt;/sup&gt; relationships</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>All participants</td>
<td>-0.15 (-0.28 to -0.01)</td>
<td>-0.302 ± 0.142</td>
</tr>
<tr>
<td>Men (n = 219)</td>
<td>-0.14 (-0.25 to -0.04)</td>
<td>-0.343 ± 0.129</td>
</tr>
<tr>
<td>Premenopausal (n = 195)</td>
<td>-0.18 (-0.32 to -0.04)</td>
<td>-0.597 ± 0.243&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Not receiving antihypertensive therapy</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Men (n = 187)</td>
<td>-0.13 (-0.27 to 0.02)</td>
<td>-0.274 ± 0.158</td>
</tr>
<tr>
<td>Women (n = 269)</td>
<td>-0.21 (-0.32 to 0.09)</td>
<td>-0.567 ± 0.167&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Premenopausal (n = 191)</td>
<td>-0.18 (-0.32 to -0.04)</td>
<td>-0.599 ± 0.244&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

**Abbreviations:** CI, confidence interval; PWV, pulse wave velocity.

<sup>a</sup>For relationships.

<sup>b</sup>Indicates P < 0.05 when comparing β coefficients vs. men.

<sup>c</sup>Adjusted for age, sex, menopausal status, systolic blood pressure, body mass index, regular smoking, regular alcohol consumption, antihypertensive treatment, diabetes mellitus or an HbA1c > 6.1%. No differences were noted in Pearson’s r or partial r between all or premenopausal women vs. men (P > 0.05).

### Table 5. Multivariate adjusted relationships between relative leukocyte telomere length (log T/S) and central aortic hemodynamics in men and women

<table>
<thead>
<tr>
<th></th>
<th>Men (n = 219)</th>
<th>Women</th>
<th>All (n = 361)</th>
<th>Premenopausal (n = 195)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Partial r&lt;sup&gt;a&lt;/sup&gt;</td>
<td>P value</td>
<td>Partial r&lt;sup&gt;a&lt;/sup&gt;</td>
<td>P value</td>
</tr>
<tr>
<td>Aortic SBP</td>
<td>0.01</td>
<td>0.92</td>
<td>-0.04</td>
<td>0.48</td>
</tr>
<tr>
<td>Aortic PP</td>
<td>0.02</td>
<td>0.78</td>
<td>-0.03</td>
<td>0.54</td>
</tr>
<tr>
<td>Forward wave pressure</td>
<td>0.03</td>
<td>0.60</td>
<td>-0.03</td>
<td>0.56</td>
</tr>
<tr>
<td>Augmentation pressure</td>
<td>-0.03</td>
<td>0.63</td>
<td>-0.02</td>
<td>0.66</td>
</tr>
</tbody>
</table>

**Abbreviations:** PP, pulse pressure; SBP, systolic blood pressure.

<sup>a</sup>Adjusted for age and the presence or absence of regular alcohol consumption.
the relationships between leukocyte telomere length and CVD have yet to be fully elucidated. In this regard, together with prior results demonstrating an independent relationship between leukocyte telomere length and aortic stiffness in 120 men,14 in 163 men with coronary artery disease,16 and in 112 healthy men,16 the results of the present study conducted in the largest study sample evaluated to date (n = 580, 361 women and 219 men) support the notion that telomere attrition may in-part be associated with CVD through relationships with aortic stiffness. Importantly, the present study suggests that the impact of telomere attrition on aortic stiffness, if indeed a causal relationship, may occur to a greater extent in premenopausal women as compared to men. Although the present study was not designed to evaluate the potential explanations for independent relationships between telomere attrition and aortic stiffness, assuming that these relationships represent cause and effect, they could be attributed to the impact of telomere attrition on vascular smooth muscle senescence,10 oxidative stress,10 large artery calcification,11 or general atherosclerotic changes,9 any of which may directly or indirectly influence the stiffness of the aorta. Importantly, however, in the present study, we were unable to show inverse relationships between indexes of renin–angiotensin II activation or insulin resistance and telomere length, factors that are well recognized as mediating increases in large artery oxidative stress. Hence, the mechanisms that explain relationships between telomere length and aortic stiffness require further study.

Although the present study provides clear evidence that telomere length is independently associated with aortic stiffness in either men or women, it is important to emphasize that these relationships do not necessarily explain relationships between telomere length and the atherosclerotic process.9–12 In this regard, although atherosclerotic changes are associated with increases in aortic stiffness, the pathophysiological processes responsible for increases in aortic stiffness contribute toward arteriosclerotic rather than atherosclerotic changes. However, there is now clear evidence from meta-analyses of a number of large studies that aortic stiffness, as determined from carotid–femoral PWV, is a strong and independent risk factor for cardiovascular events.33,34

Although in the present study, telomere length was independently associated with aortic stiffness, no independent relationships between telomere length and central aortic BP were noted. It may therefore be argued that any impact of telomere attrition on aortic stiffness is unlikely to contribute to the development of CVD, as aortic stiffness is thought to mediate organ damage in-part by enhancing aortic BP. However, increases in aortic stiffness may produce adverse end-organ changes by decreasing wave reflection from the periphery (which will decrease aortic BP) and increase pulsatile energy in the peripheral vasculature.17 Hence, irrespective of effects on aortic BP, telomere attrition may contribute toward cardiovascular events through increases in aortic stiffness.

A limitation of the present study is the cross-sectional design which prevents us from drawing firm conclusions on cause and effect. In this regard, future longitudinal studies are required to evaluate whether changes in relative telomere length are independently related to changes in aortic stiffness, and whether these relationships are similar in premenopausal women as compared to men. Further, in participants included in the present substudy, a modestly higher PWV was noted as compared to those not included in the study. Hence, the results may be biased toward those with higher rather than lower PWV values. However, PWV was similar between men and women in the study sample. When making comparisons between our mean PWV values, please note that these measurements were calculated from subtracted distances (distance from suprasternal notch to femoral measurement site minus distance from suprasternal notch to carotid measurement site), prior to the recommendation that a standardized approach of 80% of the direct carotid–femoral distance be used.35 Hence, our PWV values may be marginally different from actual invasive measurements. As estrogen and progesterone concentrations across all phases of the menstrual cycle cannot be determined in large community-based studies such as the present study, we did not assess relationships between estrogen or progesterone concentrations and telomere length. Hence, although relations between age and telomere length and telomere length and PWV were similar between men, all women, and premenopausal women, we may have missed more subtle effects of sex steroids in women.

SUPPLEMENTARY MATERIAL

Supplementary materials are available at American Journal of Hypertension (http://ajh.oxfordjournals.org).

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DISCLOSURE

The authors declared no conflict of interest.

REFERENCES


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