Cystatin C Associates with Arterial Stiffness in Older Adults

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ABSTRACT

Large arteries commonly become stiff in kidney failure, but few studies have investigated arterial stiffness in earlier stages of kidney disease. We evaluated the association between kidney function and aortic pulse wave velocity (aPWV) and its potential modification by race, diabetes, or coronary heart disease in older adults. We measured aPWV in 2468 participants in the Health Aging and Body Composition (Health ABC) study; mean age was 73.7 yr, 40% were black, and 24% had diabetes. After categorizing kidney function into three groups on the basis of cystatin C level, multivariable analysis revealed that the medium and high cystatin C groups associated with a 5.3% (95% confidence interval 0.8 to 10.0%) and 8.0% (95% confidence interval 2.2 to 14.1%) higher aPWV than the low cystatin C group; however, chronic kidney disease, as defined by estimated GFR <60 ml/min per 1.73 m², did not significantly associate with aPWV. We did not identify interactions between cystatin C and race, diabetes, or coronary heart disease. In conclusion, stiffness of large arteries, a major risk factor for cardiovascular disease, may partially mediate the association between cystatin C and cardiovascular risk in older adults.


Cardiovascular disease (CVD) is the primary cause of mortality in patients with kidney failure, accounting for >50% of deaths. Large vessel arterial stiffness is believed to be one of the mechanisms underlying this elevated risk. Recent studies have demonstrated that dialysis patients have less compliant arteries compared with control subjects matched for age and blood pressure (BP).1 Furthermore, both pulse pressure and increased aortic pulse wave velocity (aPWV),2,3 manifestations of large vessel stiffness, are independent risk factors for adverse outcomes in this population. Recent studies have demonstrated that even mildly decreased kidney function is a powerful and independent risk factor for CVD outcomes.4–6 One candidate explanation for this association is that individuals with early stages of kidney disease may have abnormalities in large arterial compliance. This mechanism could be important in older...
adults, who have increased risk for both kidney disease and CVD.

The Health Aging and Body Composition (Health ABC) study is an ideal cohort to study the association of kidney function with vascular stiffness in older adults, because the participants encompass a broad range of kidney function and have large representation of black individuals and patients with diabetes and coronary heart disease (CHD). In addition, Health ABC measured both creatinine and cystatin C at baseline. As a measure of kidney function, cystatin C offers the advantage of not being influenced to the same extent as serum creatinine by gender, race, and muscle mass and particularly in older adults may be a better marker of kidney function.7 Furthermore, cystatin C has been shown to have a more linear relationship with measures of kidney function, cystatin C offers the advantage of being modified by race, diabetes, or CHD. We also evaluated whether any associations would be

\[ \text{cystatin C} = \text{baseline measurement} \]

\[ \text{eGFR} = \text{estimated glomerular filtration rate} \]

\[ \text{aPWV} = \text{arterial pulse wave velocity} \]

\[ \text{CHD} = \text{coronary heart disease} \]

\[ \text{ACEI} = \text{angiotensin-converting enzyme inhibitor} \]

\[ \text{ARB} = \text{angiotensin receptor blocker} \]

\[ \text{BMI} = \text{body mass index} \]

\[ \text{IFG} = \text{impaired fasting glucose} \]

\[ \text{IGT} = \text{impaired glucose tolerance} \]

\[ \text{PVD} = \text{peripheral vascular disease} \]

\[ \text{SD} = \text{standard deviation} \]

\[ \text{yr} = \text{years} \]

\[ \text{mg/L} = \text{milligrams per liter} \]

\[ \text{cm/s} = \text{centimeters per second} \]

\[ \text{m}^2 = \text{square meter} \]

\[ \text{eGFR} (\text{ml/min per 1.73 m}^2; \text{mean} = \text{value for group} \]

\[ \text{BMI} (\text{kg/m}^2; \text{mean} = \text{value for group} \]

\[ \text{Cystatin C Level} = \text{baseline measurement} \]

\[ \text{Age} (\text{yr}; \text{mean} = \text{value for group} \]

\[ \text{Male} (\text{n [%]}) = \text{number of males in group} \]

\[ \text{Black} (\text{n [%]}) = \text{number of blacks in group} \]

\[ \text{BMI} (\text{kg/m}^2; \text{mean} = \text{value for group} \]

\[ \text{Current smoking} (\text{n [%]}) = \text{number of current smokers in group} \]

\[ \text{Prevalent CHD} (\text{n [%]}) = \text{number of patients with prevalent CHD in group} \]

\[ \text{Hypertension} (\text{n [%]}) = \text{number of hypertensives in group} \]

\[ \text{Prevalent PVD} (\text{n [%]}) = \text{number of patients with prevalent PVD in group} \]

\[ \text{Diabetes status} (\text{n [%]}) = \text{number of diabetics in group} \]

\[ \text{IFG/IGT} = \text{impaired fasting glucose/impaired glucose tolerance} \]

\[ \text{ACEI use} (\text{n [%]}) = \text{number of patients using ACEI in group} \]

\[ \text{ARB use} (\text{n [%]}) = \text{number of patients using ARB in group} \]

\[ \text{Table 1. Characteristics of participants by baseline cystatin C level}\]

\[ \text{Characteristic} \]

\[ \text{Low <0.84 mg/L} \]

\[ \text{(n = 454)} \]

\[ \text{Medium 0.84 to 1.18 mg/L} \]

\[ \text{(n = 1497)} \]

\[ \text{High 1.19 mg/L} \]

\[ \text{(n = 517)} \]

\[ \text{P} \]

\[ \text{Age (yr; mean ± SD)} \]

\[ 73.0 ± 2.8 \]

\[ 73.7 ± 2.8 \]

\[ 74.3 ± 2.9 \]

\[ <0.0001 \]

\[ \text{Male (n [%])} \]

\[ 149 (33) \]

\[ 743 (50) \]

\[ 285 (55) \]

\[ <0.0001 \]

\[ \text{Black (n [%])} \]

\[ 219 (48) \]

\[ 593 (40) \]

\[ 181 (35) \]

\[ 0.0001 \]

\[ \text{BMI (kg/m}^2; \text{mean ± SD)} \]

\[ 26.1 ± 4.7 \]

\[ 27.5 ± 4.7 \]

\[ 28.3 ± 5.0 \]

\[ <0.0001 \]

\[ \text{Current smoking (n [%])} \]

\[ 39 (9) \]

\[ 159 (11) \]

\[ 52 (10) \]

\[ 0.4500 \]

\[ \text{Prevalent CHD (n [%])} \]

\[ 73 (16) \]

\[ 301 (20) \]

\[ 153 (30) \]

\[ <0.0001 \]

\[ \text{Hypertension (n [%])} \]

\[ 265 (58) \]

\[ 919 (61) \]

\[ 372 (72) \]

\[ <0.0001 \]

\[ \text{Prevalent PVD (n [%])} \]

\[ 16 (3.5) \]

\[ 64 (4.3) \]

\[ 53 (10.3) \]

\[ <0.0001 \]

\[ \text{Diabetes status (n [%])} \]

\[ \text{none} \]

\[ 213 (47) \]

\[ 696 (47) \]

\[ 209 (40) \]

\[ 0.0560 \]

\[ \text{IFG/IGT} \]

\[ 123 (27) \]

\[ 427 (29) \]

\[ 156 (30) \]

\[ \text{diabetes} \]

\[ 99 (22) \]

\[ 325 (22) \]

\[ 140 (27) \]

\[ \text{Triglycerides (mg/dl; mean ± SD)} \]

\[ 118.8 ± 62.0 \]

\[ 140.4 ± 81.0 \]

\[ 156.8 ± 105.4 \]

\[ <0.0001 \]

\[ \text{HDL (mg/dl; mean ± SD)} \]

\[ 61.2 ± 18.6 \]

\[ 54.1 ± 16.2 \]

\[ 49.8 ± 15.9 \]

\[ <0.0001 \]

\[ \text{LDL (mg/dl; mean ± SD)} \]

\[ 124.6 ± 32.5 \]

\[ 122.5 ± 34.2 \]

\[ 117.4 ± 37.6 \]

\[ 0.0030 \]

\[ \text{CRP (mg/L; mean ± SD)} \]

\[ 2.7 ± 5.1 \]

\[ 2.7 ± 3.6 \]

\[ 4.1 ± 7.1 \]

\[ <0.0001 \]

\[ \text{Creatinine (mg/dl; mean ± SD)} \]

\[ 0.87 ± 0.15 \]

\[ 1.01 ± 0.18 \]

\[ 1.36 ± 0.73 \]

\[ <0.0001 \]

\[ \text{eGFR (ml/min per 1.73 m}^2\text{; mean ± SD)} \]

\[ 84.7 ± 15.3 \]

\[ 73.9 ± 12.7 \]

\[ 57.0 ± 15.0 \]

\[ <0.0001 \]

\[ \text{ACEI use (n [%])} \]

\[ 51 (11) \]

\[ 209 (14) \]

\[ 104 (20) \]

\[ 0.0002 \]

\[ \text{ARB use (n [%])} \]

\[ 10 (2) \]

\[ 27 (2) \]

\[ 23 (5) \]

\[ 0.0030 \]

aACEI, angiotensin-converting enzyme inhibitor; ARB, angiotensin receptor blocker; BMI, body mass index; IFG, impaired fasting glucose; IGT, impaired glucose tolerance; PVD, peripheral vascular disease.

bP values obtained by ANOVA or χ² test.
CKD and mean aPWV \((P = 0.13\) unadjusted analysis and \(P = 0.50\) in adjusted analysis; Figure 1). Adjusted winsorized spline analysis suggested a linear association of cystatin C with log-transformed aPWV in the middle 95% of the cystatin C distribution, whereas the relationship with creatinine was not linear (Figure 2).

In continuous analysis, there was a significant association between cystatin C and creatinine with aPWV in unadjusted models (2.97\% [95\% confidence interval (CI) 1.40 to 4.56; \(P = 0.0002\]) higher aPWV per SD higher cystatin C and 1.98\% [95\% CI 0.40 to 3.60; \(P = 0.01\]) higher aPWV per SD higher creatinine). In the fully adjusted models, the relationship with both cystatin C and creatinine was attenuated and no longer significant (1.19\% [95\% CI −0.58 to 2.98; \(P = 0.19\]) higher aPWV per SD higher cystatin C and 0.96\% [95\% CI −2.64 to 0.76; \(P = 0.2732\]). In additional adjusted analyses using cystatin C winsorized at 95\% of the distribution, there was a 2.10\% (95\% CI 0.30 to 3.94; \(P = 0.02\)) higher aPWV per SD higher cystatin C and a 0.92\% (95\% CI −1.02 to 2.91; \(P = 0.35\)) higher aPWV per SD increase in creatinine.

When cystatin C was divided into low, medium, and high groups, there was a significant association between cystatin C and aPWV in unadjusted and fully adjusted analyses. There was no association, however, between the presence of CKD and aPWV (Table 2). In separate models that included mean arterial pressure rather than systolic (SBP) or diastolic BP (DBP), that excluded SBP and DBP, and that adjusted for oral steroid use, the associations were not appreciably changed (data not shown).

There were no interactions between cystatin C and race \((P = 0.81)\), diabetes \((P = 0.88)\), or CHD \((P = 0.41)\) in the adjusted models. When the analyses were stratified into these subgroups, the percentage change in aPWV seemed somewhat higher in white than in black patients (Table 3). The percentage

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**Figure 1.** (A and B) Unadjusted (A) and adjusted (B) geometric mean aPWV by cystatin C groups and CKD.

**Figure 2.** (A) Adjusted spline of cystatin C with aPWV. (B) Adjusted spline of creatinine with aPWV. Adjusted for age, gender, race, site, body mass index, alcohol use (≤ 1 drink per day versus >1 drink per day), smoking, angiotensin-converting enzyme inhibitor use, angiotensin receptor blocker use, prevalent congestive heart failure, prevalent CHD, diabetes, ankle arm index, triglycerides, LDL cholesterol, HDL cholesterol, C-reactive protein, IL-6, heart rate, peripheral vascular disease, SBP, and DBP. Tick marks along the x axis indicate values for individual participants.
changes were similar among patients with and without diabetes and in patients with or without CHD. No associations were noted between the presence of CKD and aPWV in any of the subgroups (Table 3).

**DISCUSSION**

In Health ABC, a cohort of well-functioning ambulatory older adults, kidney function as assessed by different levels of cystatin C was independently associated with PWV. This relationship was not modified by race, the presence of diabetes, or CHD. No association was observed between CKD and aPWV.

Arterial stiffness is known to be increased in patients with kidney failure, an association that has been partially attributed to an increase in the calcium content of the arterial wall. Furthermore, it is now believed that increased aPWV is an important risk factor for long-term outcomes in patients with kidney failure. There are few studies of aPWV in the earlier stages of CKD.

Our results on arterial stiffness and kidney function are important because our cohort exclusively included older adults, who are particularly at increased risk for both CKD and CVD; used cystatin C– and creatinine-based eGFR; and included a large population of black individuals and patients with diabetes and CHD. We noted that cystatin C groups but not CKD were independently associated with aPWV. Our results are in contrast to other studies, which found associations between creatinine clearance or creatinine-based eGFR with arterial stiffness. For example, in a cohort of 1290 individuals with normal BP or essential hypertension, kidney function was inversely associated with aPWV in individuals in the lowest tertile of creatinine clearance (mean 68.5 ml/min per 1.73 m²). Similarly, a study of 3387 healthy individuals with a mean age of 52 yr (mean creatinine clearance 92 ml/min) demonstrated that aPWV was independently associated with creatinine clearance even after adjustment for confounding variables including proteinuria. Finally, in a recent study comparing 95 patients who had CKD (mean GFR 31 ml/min per 1.73 m²) with 121 patients who had hypertension and no CKD, aortic stiffness was significantly higher in patients who had hypertension with CKD than without CKD, despite similar age and BP in each of the groups.

Unlike in the aforementioned studies, creatinine, eGFR, and CKD did not have an independent association with aPWV, although cystatin C groups did. These results suggest that the multiple determinants of creatinine generation, particularly muscle mass, in older adults may limit its utility as a measure of kidney function. Cystatin C is less influenced by gender, race, and muscle mass and may therefore more accurately reflect kidney function in this patient population.

We acknowledge, however, that it is possible that actual kidney function is not independently associated with aPWV in this cohort and that the observed association between cystatin C groups and aPWV may be due to an unknown effect of cystatin C that is independent of GFR. For example, in the Prevention of Renal and Vascular End-Stage Disease (PREVEND) Study, cystatin C concentrations were significantly associated with increased age, male gender, increased weight and height, current smoking, and higher C-reactive protein levels even after adjustment for creatinine clearance. Other factors, such as hyperthyroidism and glucocorticoids, have also been associated with higher concentrations of cystatin C. In Health ABC, thyroid-stimulating hormone was not measured at baseline, so we were unable to adjust for it; however, when oral steroids were added into the models, the results did not change appreciably. Furthermore, we adjusted for the other potential determinants in our analyses. It is also important to note that GFR was not measured directly in the latter studies. A recent study by Menon et al. demonstrated that cystatin C is highly correlated with measured GFR, suggesting that these other determinants are less likely to play a major role. Finally, there is little evidence to support a pathophysiologic role of cystatin C, independent of GFR, in arterial stiffness.

There are several potential mechanisms to explain the link between kidney function and arterial stiffness. First, kidney disease may directly promote arterial stiffness through its effect on hypertension and salt retention. Salt retention, in addition to affecting volume status and hypertension, may have direct trophic effects on the vasculature. It has also been suggested that sodium may modify vascular tone by affecting the sympathetic nervous system. Second, changes in kidney function

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**Table 2. Unadjusted and adjusted percentage change in aPWV by cystatin C and CKD**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Unadjusted Model</th>
<th>Adjusted Model*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cystatin groups</td>
<td></td>
<td></td>
</tr>
<tr>
<td>low (n = 454)</td>
<td>Reference</td>
<td>Reference</td>
</tr>
<tr>
<td>medium (n = 1497)</td>
<td>6.3 (2.1 to 10.7)</td>
<td>5.3 (0.8 to 10.0)</td>
</tr>
<tr>
<td>high (n = 517)</td>
<td>13.8 (8.4 to 19.4)</td>
<td>8.0 (2.2 to 14.1)</td>
</tr>
<tr>
<td>CKD</td>
<td></td>
<td></td>
</tr>
<tr>
<td>not present CKD (n = 1934)</td>
<td>Reference</td>
<td>Reference</td>
</tr>
<tr>
<td>present (n = 533)</td>
<td>2.9 (−0.9 to 6.8)</td>
<td>1.2 (−2.7 to 5.3)</td>
</tr>
</tbody>
</table>

*Adjusted for age, gender, race, site, BMI, alcohol use (≤1 drink per day versus >1 drink per day), smoking, ACEI use, ARB use, prevalent congestive heart failure, prevalent CHD, diabetes, ankle arm index, triglycerides, LDL cholesterol, HDL cholesterol, C-reactive protein, IL-6, heart rate, PVD, SBP, and DBP.
Table 3. Adjusted percent change in aPWV by cystatin C, presence of CKD, and risk groups.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Reference</th>
<th>Cystatin C</th>
<th>CHD not Present (n = 1093)</th>
<th>CHD Present (n = 527)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>low (n = 454)</td>
<td>Reference</td>
<td>Percentage Change (95% CI)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>medium (n = 1497)</td>
<td>Reference</td>
<td>10.1 (0.6 to 22.0)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>high (n = 517)</td>
<td>Reference</td>
<td>4.4 (-2.5 to 11.8)</td>
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<tr>
<td>CKD</td>
<td></td>
<td>present (n = 993)</td>
<td>Reference</td>
<td>-11.1 (-16 to -5.6)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>not present (n = 1824)</td>
<td>Reference</td>
<td>4.5 (2.5 to 7.1)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>n = 1893</td>
<td>Reference</td>
<td>11.1 (-3.4 to 25.4)</td>
</tr>
</tbody>
</table>

*Adjusted for age, gender, race, BMI, alcohol use, smoking, ACEI use, ARB use, prevalent congestive heart failure, prevalent CHD, diabetes, ankle arm index, triglycerides, IDL cholesterol, HDL cholesterol, C-reactive protein, IL-6, systolic and diastolic blood pressure, heart rate, PVD, SBP, and DBP.

The cross-sectional nature of our analysis does not permit us to distinguish the direction of this association.

We were especially interested in evaluating whether the relationship between kidney function and arterial stiffness would be modified by race, diabetes, or the presence of CHD. Several studies have suggested that the prevalence of arterial stiffness may be higher in black than in white individuals, possibly because the severity of hypertension and diabetes is worse in black individuals. Similarly, studies in both CKD and non-CKD have reported higher PWV in those with diabetes in comparison with those without diabetes. Finally, one could hypothesize that prevalent CHD may modify the relationship between kidney function and aPWV. Although diabetes and black race were associated with higher aPWV in this study, they did not modify the association between cystatin C and aPWV. In fact, in subgroup analysis, the relationship between cystatin C and aPWV was perhaps slightly stronger in white compared with black individuals, although the interaction term was not significant (NS). Similarly, the interaction terms were NS and the effect sizes similar in those with diabetes versus those without diabetes.

Our study has several limitations. Most important, as mentioned already, in any cross-sectional study, the direction of the association cannot be ascertained and no causal inference can be made. In addition we cannot be certain that the association of cystatin C with aPWV is solely due to its approximation of kidney function, because GFR was not measured directly in this study. Furthermore, this study did not measure albumin excretion rate or hemoglobin levels, potentially important risk factors for arterial stiffness. It is also possible that there is residual confounding from other variables that were included in the analysis and unmeasured confounding from variables that were not evaluated in this study. Finally, because the Health ABC study was composed of ambulatory older adults and those with kidney disease were primarily in stage 3 CKD (eGFR 30 to 60 ml/min per 1.73 m²), the associations between cystatin C and aPWV in younger populations, among older
adults with functional disability, or in individuals with more advanced CKD cannot be established from this study. Furthermore, any study that includes only older adults may be subject to survival bias.

In summary higher levels of cystatin C but not the presence of CKD was independently associated with aPWV in well-functioning older adults. This relationship was not modified by race, the presence of diabetes, or CHD. Large arterial stiffness, a major risk factor for CVD, may partially mediate the strong association between cystatin C and CVD risk in older adults.

CONCISE METHODS

Study Sample
The Health ABC is a longitudinal study of the impact of changes in weight and body composition on age-related physiologic and functional changes. Participants aged 70 to 79 yr were recruited from Medicare eligibility lists from March 1997 through July 1998 at two field centers in Pittsburgh, PA, and Memphis, TN. White patients were recruited from a random sample of the lists; black patients were recruited from all age-eligible individuals residing in the respective communities. The cohort consists of 1491 (48%) men and 1584 (52%) women, 42% of whom were black and 24% of whom had diabetes. aPWV data were missing for 354 participants because of equipment problems. An additional 233 participants had waveforms that were unusable because there was no clearly defined initial upstroke of the waveform, the trace was contaminated with a large venous flow component, no consistent flow waveform was found, or the electrocardiogram timing signal used to synchronize waveform averaging was too noisy for reliable use.

Adequate specimens for analysis of cystatin C and creatinine were available for 2468 individuals with adequate aPWV measures, the sample for this analysis. All participants gave informed written consent, and the protocol was approved by the institutional review boards of the clinical sites and the Data Coordinating Center (University of California, San Francisco, San Francisco, CA).

Primary Outcome: aPWV
The methods for measurement of aPWV in Health ABC have been described in detail. In brief, aPWV was measured from simultaneous Doppler flow signals obtained from the right carotid and right femoral arteries with nondirectional transcutaneous Doppler flow probes (model 810A, 9.0- to 10-MHz probes; Parks Medical Electronics, Aloha, OR). Digitized data were recorded by custom programming for subsequent analysis. Less compliant vessels are associated with a faster aPWV. Results from all acceptable runs were averaged for the final aPWV measure used in the analyses. Replicate measures of aPWV in 14 individuals revealed intraclass correlations of 0.88 between sonographs and 0.84 between readers.

Ankle brachial BPs were used as a measure of occlusive disease to the lower extremities. Pressures were taken in the right arm and both ankles (posterior tibial artery) with standard BP cuffs and a pencil Doppler. The SBP of the ankle was divided by the SBP of the arm to create the ankle arm index. Measurements were performed twice, and the results were averaged. The lower value between the two legs was used, and an ankle arm index of 0.90 was considered evidence of occlusive disease.

Predictor: Kidney Function
Three different measures of kidney function were evaluated. Cystatin C was measured using plasma specimens that had been stored at −70°C at the Health ABC core laboratory (University of Vermont, Burlington, VT) using a BNII nephelometer (Dade Behring, Deerfield, IL), which used a particle-enhanced immuno nephelometric assay (N Latex Cystatin C). The intraindividual coefficient of variation was 7.7%. The assay range is 0.195 to 7.330 mg/L, with the reference range for young, healthy individuals reported as 0.530 to 0.950 mg/L. Creatinine was assayed by a colorimetric technique on a Johnson & Johnson Vitros 950 analyzer (New Brunswick, NJ); the intraindividual coefficient of variation was approximately 1.5% in Health ABC reproducibility studies.

GFR was estimated using the simplified Modification of Diet in Renal Disease (MDRD) equation: GFR (ml/min per 1.73m²) = 186 × serum Cr (mg/dl) −1.154 × age −0.203 × 0.742 (if female) × 1.212 (if black). CKD was defined as an eGFR < 60 ml/min per 1.73 m². Low (<0.84 mg/L), medium (0.84 to 1.18 mg/L), and high (≥1.19 mg/L) cystatin C groups were based on mortality associations observed in a previous study.

Covariates
Covariates included sociodemographic factors (age, gender, race, clinical site, education level), lifestyle factors (current smoking [defined by current versus former or never], alcohol use [defined by > 1 drink per day versus < 1 drink per day], and body mass index), and comorbid conditions (impaired fasting glucose [defined as fasting glucose from 100 to 125 mg/dl], impaired glucose tolerance [defined as a 2-h glucose tolerance of 140 to 200 mg/dl], diabetes [defined by use of hypoglycemic agents, self-report, fasting plasma glucose ≥126 mg/dl, or an oral glucose tolerance test >200 mg/dl], hypertension [defined either by self-report plus use of antihypertensive medications or by measured SBP >140 mmHg or DBP >90 mmHg], heart failure, CHD [defined as CHD, myocardial infarction, angina, coronary artery bypass], chronic obstructive pulmonary disease, peripheral arterial disease, and cerebrovascular disease). Prevalent comorbid conditions were determined by an algorithm that includes self-report, medication use, and medical records. Additional covariates included blood glucose, total cholesterol, HDL cholesterol, triglycerides, albumin, insulin, TNF-α, and C-reactive protein. We also specifically adjusted for use of angiotensin-converting enzyme inhibitors and angiotensin receptor blockers because they may directly affect level of kidney function as well as arterial compliance; therefore, they are potential confounding variables in the relationship between the two.

Statistical Analysis
We compared the distribution of each covariate among cystatin C risk groups using the χ² test for categorical variables and ANOVA for continuous variables. Unadjusted and adjusted geometric mean aPWV were calculated by cystatin C groups and CKD status. After
examination of the distribution of aPWV, the natural log transformation was used to meet normality assumptions for linear regression modeling. We used generalized additive models to determine the appropriate functional form of cystatin C and to evaluate any potential threshold effects in the association of cystatin C with log-transformed PWV. Multivariate linear regression models were used to evaluate the association between kidney function measures and aPWV. Models were initially unadjusted and subsequently adjusted for demographics, comorbid conditions, and medications.

Winsorized splines using the 2.5th and 97.5th percentiles were created to evaluate the relation between either cystatin C or creatinine with log-transformed PWV. Winsorizing involves ranking all values of a variable, then redefining the most extreme values to the percentiles of choice; in this case the 2.5th and 97.5th percentiles. We initially modeled cystatin C as a continuous variable to determine the percentage change in aPWV per SD increase of cystatin C. In additional analyses as a result of outliers with low PWV in those with cystatin C >2 mg/L, the analysis was repeated using cystatin C winsorized at the 2.5th and 97.5th percentiles of its distribution. For consistency, analyses were repeated using winsorized creatinine. We used a transformation of the $\beta$ coefficients [100 * ($e^\beta - 1$)] to obtain the percentage increase or decrease in aPWV per SD increase in cystatin C or creatinine. Because the splines did suggest a possible nonlinear association of cystatin and PWV, especially in those with cystatin C $\geq$2 mg/L, we also modeled cystatin C as a categorical variable represented as low (<0.84 mg/L), medium (0.84 to 1.18 mg/L), and high (>1.18 mg/L) on the basis of our previous work. In the case of eGFR <60 ml/min per 1.73 m², the percentage increase in aPWV corresponds to a percentage change from the reference eGFR category of >60 ml/min per 1.73 m².

Because race, diabetes, and CHD could modify the relationship between cystatin C and aPWV, we evaluated interactions with these covariates and conducted stratified analyses within subgroups. Finally, in separate models, we adjusted for mean arterial pressure rather than SBP or DBP, performed an additional model that excluded SBP and DBP because they may be mediators by which aPWV is related to kidney function, and adjusted for oral steroid use because it may have an effect on cystatin C independent of GFR. Spline analyses were conducted using S-Plus V6.1 (Insightful Corp., Seattle, WA). All other analyses were conducted in SAS 9.1.3 (SAS Institute, Cary, NC).

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DISCLOSURES

None.

REFERENCES


