THE GERONTOLOGICAL SOCIETY OF AMERICA®

Journals of Gerontology: Medical Sciences
cite as: *J Gerontol A Biol Sci Med Sci*, 2017, Vol. 72, No. 7, 965–970
doi:10.1093/gerona/glw223
Advance Access publication November 9, 2016

OXFORD

Research Article

Cystatin C and the Risk of Frailty and Mortality in Older Men

Allyson Hart,[1,2] Terri L. Blackwell,[3] Misti L. Paudel,[4,5] Brent C. Taylor,[5,6] Eric S. Orwoll,[7] Peggy M. Cawthon,[3] and Kristine E. Ensrud[2,5,6]; for the Osteoporotic Fractures in Men (MrOS) Study Group

[1]Division of Nephrology, Hennepin County Medical Center, Minneapolis, Minnesota. [2]Department of Medicine, University of Minnesota, Minneapolis. [3]Research Institute, California Pacific Medical Center, San Francisco. [4]NORC at the University of Chicago, Health Care Department, Bethesda, Maryland. [5]Division of Epidemiology and Community Health, University of Minnesota, Minneapolis. [6]Center for Chronic Disease Outcomes Research, VA Health Care System, Minneapolis, Minnesota. [7]Bone and Mineral Unit, Oregon Health and Science University, Portland.

Address correspondence to Allyson Hart, MD, MS, Division of Nephrology, 701 Park Avenue, Suite S5, Minneapolis, MN 55415. E-mail: hart1044@umn.edu

Received April 21, 2016; Accepted October 18, 2016

Decision Editor: Stephen Kritchevsky, PhD

Abstract

Background: This study examines the association between cystatin C (cysC) levels and risks of progression of frailty status or death in older men.

Methods: Prospective study of 2,613 men without overt frailty aged 67 years and older enrolled in the MrOS ancillary sleep study. Baseline measurements included serum cysC, serum creatinine, and frailty status. Repeat frailty status, performed an average of 3.4 years later, was assessed as an ordinal outcome of robust, intermediate stage (prefrail), frail or dead.

Results: Mean age was 75.7 years. Men with higher cysC were older and had a higher comorbidity burden. After adjusting for age, clinical site, and race, higher cysC was associated with nearly twofold greater odds of being classified as intermediate stage versus robust (OR quartile 4 vs 1; 1.82, 95% confidence interval [CI] 1.35–2.45), a threefold greater odds of frailty versus robust (OR quartile 4 vs 1; 3.13, 95% CI 2.03–4.82), and a more than fivefold greater odds of death versus robust (OR quartile 4 vs 1; 5.48, 95% CI 2.98–10.08). Results were similar for cysC-based estimated glomerular filtration rate (eGFR). This relationship was attenuated but persisted after adjusting for additional potential confounders including baseline frailty status, body mass index, smoking status, comorbidity burden, self-reported disability, and serum albumin. In contrast, neither serum creatinine nor creatinine-based eGFR was associated in a graded manner with higher risks of development of frailty or death.

Conclusions: In this cohort of older men without overt frailty, higher cysC and cysC-based eGFR, but not creatinine or creatinine-based estimates of GFR, were associated with increased risks of frailty or death. These findings suggest that higher cysC level may be a promising biomarker for unsuccessful aging as manifested by increased risks of frailty and death.

Keywords: Cystatin C—Creatinine—Frailty—Mortality—Men

Chronic kidney disease (CKD), typically defined as an estimated glomerular filtration rate (eGFR) of less than 60 mL/min/1.73 m^2, is common among older adults (1,2) and has been associated with higher rates of prevalent and incident physical impairment, cognitive impairment, and frailty (3,4). Frailty, characterized by decreased physiologic reserve and increased vulnerability to physical decline and death, is frequently defined using the Cardiovascular Health Study (CHS) index proposed by Fried and colleagues in which three or more of the following criteria are present: shrinking (eg, unintentional weight loss), self-reported exhaustion or poor energy, weakness manifested by low grip strength, slow walking speed, and low physical activity. Participants with none of the criteria are considered

to be robust, whereas those with one or two criteria are considered to be in an intermediate stage (5). In an analysis of CHS data, frailty defined by the CHS index was predictive of an increased risk of falling, hospitalization, disability, and mortality (6). Although multiple instruments have been developed to operationalize the construct of frailty, the CHS index has been extensively studied and the predictive value has been validated in multiple cohorts (7–10).

Serum creatinine-based eGFR ($eGFR_{Cr}$) presents challenges in elderly individuals with mild-to-moderate reductions in kidney function due to its dependence on muscle mass and lower accuracy at higher eGFR levels (11). These concerns have generated interest in the use of cystatin C (cysC), an alternative measure of kidney function that is less dependent on muscle mass (12) and has better accuracy than $eGFR_{Cr}$ alone (13). CysC has been found to have a stronger association than $eGFR_{Cr}$ with risks of mortality, end-stage kidney disease, cardiovascular events (14,15), and hip fracture (16,17). However, the association between higher cysC and incident frailty among older adults without overt frailty has not been fully evaluated. A previous longitudinal study (18) reported that eGFR using cysC ($eGFR_{CysC}$), but not $eGFR_{Cr}$, was associated with greater risk of incident frailty in a cohort of older adults, but this analysis did not evaluate frailty status as a continuum ranging from robust, intermediate stage, frail to dead.

To test the hypothesis whether older men with higher cysC (and lower $eGFR_{CysC}$) at baseline are at risk of progression of frailty status and death at follow-up, we measured cysC in a cohort of 2,429 men aged 67 years and older without overt frailty at baseline enrolled in the Outcomes of Sleep Disorders of Older Men (MrOS Sleep) study and reassessed frailty status and vital status an average of 3.4 years later. In secondary analyses, we examined whether serum creatinine and $eGFR_{Cr}$ were associated with these outcomes.

Methods

Participants

The Osteoporotic Fractures in Men (MrOS) Study is a prospective multicenter observational study of a cohort of 5,994 community-dwelling men aged 65 years and older recruited between 2000 and 2002 from six clinical sites in the United States. Detailed descriptions of the study design and recruitment have been previously published (19,20). In order to participate, men needed to be able to walk without assistance, must not have had a bilateral hip replacement, and must not have had a medical condition that (in the judgment of the investigator) would result in imminent death. These inclusion criteria were designed to yield a cohort of men that was reasonably representative of a broad population of older men. No inclusion or exclusion criteria were based on risk of osteoporotic fracture. 3,135 men enrolled in an ancillary sleep study examination (56% of active surviving participants, >100% of recruitment goal) between 2003 and 2005 (baseline for this analysis). Of these, 2,613 were both not overtly frail at baseline and had adequate sera for cysC and creatinine measurement. After excluding 184 men who did not have frailty data at the follow-up visit an average (*SD*) of 3.42 (0.46) years later, 2,429 men comprised the analytical cohort (Supplementary Figure 1). The institutional review board at each center approved the protocol, and written informed consent was obtained from all participants.

Biochemical Data

Measures of renal function were performed on previously frozen (−70°C) stored serum samples collected at baseline; serum creatinine and cysC were measured from the same sample. Serum creatinine was analyzed using the Roche Modular P chemistry analyzer (Enzymatic/Roche Diagnostics, Indianapolis, IN) in 2013; inter-assay coefficient of variation (CV) for serum creatinine is 3.7% at 0.82 mg/dL and 2.3% at 3.62 mg/dL. This assay is calibrated to be isotope dilution mass spectrometry traceable. CysC was measured using the Roche Modular P chemistry analyzer (Turbidimetric/Gentian AS, Moss, Norway) in 2013; inter-assay CV is 4.1% at 0.94 mg/L and 2.8% at 3.29 mg/L. This assay is calibrated to the international cysC reference material ERM-DA471/IFCC. Both assays were performed at the Fairview University Medical Center Clinical Laboratory (Minneapolis, MN).

Using the Chronic Kidney Disease Epidemiology (CKD-EPI) Collaboration equation based on age, gender, and cysC alone, $eGFR_{cysC}$ was calculated (13). Using the CKD-EPI equation that includes variables for standardized creatinine, age, gender, and race, $eGFR_{Cr}$ was calculated (21). CysC + creatinine-based eGFR ($eGFR_{Cr\text{-}CysC}$) was calculated using the CKD-EPI equation including variables for cysC, creatinine, age, gender, and race (13).

C-reactive protein was measured using the ELISA assay kit from ALPCO (C-reactive protein sensitive ELISA), interleukin-6 and tumor necrosis factor-α were assayed using the Human ProInflammatory I 4-Plex Ultra-Sensitive Kit by MSD (catalog K15009C-4). Serum albumin was measured using serum collected between 2000 and 2002 using a Roche COBAS Integra 800 automated analyzer (Roche Diagnostics) with inter-assay CV of 1.98%.

Other Measurements

At baseline, demographic information, health status, social support, and smoking status were ascertained through self-administered questionnaires. Men were asked whether they had received a physician diagnosis of the nine selected medical conditions (see footnote of Table 1). In addition, dementia was defined as a score less than 80 on the Modified Mini-Mental State Examination (22–24). A summary comorbidity score was calculated as the sum of these conditions (range 0–10). Physical activity was assessed using the Physical Activity Scale for the Elderly (PASE) score (25). Height was measured with Harpenden stadiometers and weight was measured on balance beam or digital scales. Body mass index was calculated as kilograms per square meter. Walking speed was measured on a 6-m walking course and expressed as meters per second averaged over two trials. Grip strength (kg) was measured using a Jamar hand dynamometer (Lafayette Instrument, Lafayette, IN). Men were asked about their ability to perform five instrumental activities of daily living (IADLs) (26–28).

Frailty Status Assessment

Frailty status at baseline was defined using criteria similar to those proposed by Fried and colleagues using data collected in the CHS study (5). Cutpoints where applicable are provided in Supplementary Table 1:

(1) Shrinking as identified by an unintentional weight loss of 5% or more over an average of 3.36 (*SD* 0.48) years prior to baseline;
(2) Weakness as identified by a grip strength at baseline in the lowest quintile stratified by body mass index (quartiles);
(3) Poor energy as identified by an answer of "no" to the question "Do you feel full of energy?" from the Geriatric Depression Scale (29) administered at baseline;
(4) Slowness as identified by a walk speed at baseline in the lowest quintile stratified by standing height (median); and
(5) Low physical activity level at baseline as identified by a PASE score in the lowest quintile.

Table 1. Baseline Characteristics of 2,429 Men Overall and by Serum Cystatin C Quartile

	Overall	Q1	Q2	Q3	Q4	*p* Value
Characteristic	(*N* = 2,429)	(*n* = 605)	(*n* = 573)	(*n* = 624)	(*n* = 627)	
Age (y), mean (*SD*)	75.7 (5.2)	73.7 (4.4)	75.1 (4.8)	76.1 (5.0)	78.0 (5.6)	<.001
Caucasian race, *n* (%)	2201 (90.6)	516 (85.3)	523 (91.3)	578 (92.6)	584 (93.1)	<.001
Self-reported health status, *n* (%)						<.001
Poor, very poor, or fair	255 (10.5)	43 (7.1)	47 (8.2)	67 (10.8)	98 (15.6)	
Good or excellent	2172 (89.5)	561 (92.9)	526 (91.8)	556 (89.3)	529 (84.4)	
Current or past smoker, *n* (%)	1449 (59.7)	356 (58.8)	344 (60.0)	360 (57.8)	389 (62.0)	.46
Number of select medical conditions, *n* (%)[a]						<.001
0–1	1370 (56.6)	399 (66.1)	358 (62.8)	347 (55.9)	266 (42.6)	
2–3	946 (39.1)	195 (32.3)	192 (33.7)	253 (40.7)	306 (49.0)	
≥4	103 (4.3)	10 (1.7)	20 (3.5)	21 (3.4)	52 (8.3)	
PASE physical activity score, mean (*SD*)	157.4 (68.8)	168.3 (70.2)	158.4 (65.0)	157.0 (70.9)	146.5 (67.0)	<.001
Body mass index (kg/m^2), mean (*SD*)	27.2 (3.7)	26.4 (3.3)	26.6 (3.5)	27.5 (3.5)	28.0 (4.2)	<.001
Number of IADL impairments (0–5), mean (*SD*)	0.2 (0.6)	0.1 (0.5)	0.2 (0.5)	0.2 (0.6)	0.4 (0.8)	<.001
Baseline serum albumin (g/dL), mean (*SD*)	4.3 (0.2)	4.3 (0.2)	4.3 (0.2)	4.3 (0.2)	4.2 (0.2)	<.001
Frailty status at baseline, *n* (%)						<.001
Robust	917 (37.8)	294 (48.6)	235 (41.0)	230 (36.9)	158 (25.2)	
Intermediate stage	1512 (62.3)	311 (51.4)	338 (59.0)	394 (63.1)	469 (74.8)	
$eGFR_{Cr}$ (mL/min/1.73 m^2), mean (*SD*)	71.5 (14.3)	82.8 (8.6)	76.5 (9.8)	70.7 (10.0)	56.9 (13.1)	<.001
Inflammatory markers						
CRP (μg/mL), mean (*SD*)	2.7 (5.6)	2.0 (2.8)	2.2 (4.0)	2.9 (7.1)	3.7 (6.8)	<.001
IL-6 (pg/mL), mean (*SD*)	1.7 (3.8)	1.3 (3.8)	1.4 (3.9)	1.7 (2.3)	2.3 (4.6)	<.001
TNF-α, mean (*SD*)	5.4 (2.2)	4.5 (1.6)	5.0 (1.3)	5.4 (2.6)	6.6 (2.4)	<.001

Note: Quartile ranges (mg/L): <0.91, 0.91 to <1.04, 1.04 to <1.22, ≥1.22.

CRP = C-reactive protein; $eGFR_{cr}$ = creatinine-based estimated glomerular filtration rate; IADL = instrumental activities of daily living; IL-6 = interleukin-6; TNF-α = tumor necrosis factor α; PASE = Physical Activity Scale for the Elderly.

[a]Selected medical conditions include fracture after age 50, dementia (Modified Mini-Mental State Examination score < 80), transient ischemic attack or stroke, diabetes mellitus, hypertension, coronary heart disease (including myocardial infarction, bypass surgery, coronary angioplasty, or angina), congestive heart failure, chronic obstructive pulmonary disease, Parkinsonism, or cancer (except non-melanoma skin cancer).

At baseline, men with none of the above components were categorized as robust, those with one or two components were categorized as intermediate stage, and those with three or more components were categorized as frail. Because we are evaluating the outcome of incident frailty, men who were categorized as frail at baseline were excluded from the analytical cohort. Frailty status at the follow-up exam an average of 3.4 years later was defined using the same criteria cutpoints from the baseline examination (Supplementary Table 1). Because frailty and mortality are competing events, frailty status at follow-up and mortality between the baseline and follow-up exams was analyzed by considering four outcome levels at follow-up: robust, intermediate stage, frail, or dead.

Statistical Analysis

All analyses were prespecified. The primary predictor variables were serum cysC and $eGFR_{cysC}$ expressed as quartiles. The referent group was the lowest quartile (Q1) for analyses examining cysC and the highest quartile (Q4) for analyses examining $eGFR_{cysC}$. Secondary predictors included quartiles of serum creatinine (referent group Q1) as well as quartiles of $eGFR_{Cr}$ (referent group Q4) and quartiles of $eGFR_{cysC\text{-}Cr}$ (referent group Q4).

Differences in baseline characteristics by quartile of cysC were compared using chi-square (for categorical data) and analysis of variance (for normally distributed continuous data) and Kruskal–Wallis (for skewed continuous data) tests. Given the ordinal nature of the frailty status outcome variable (robust, intermediate stage, frail, or dead), multinomial logistic regression was used to simultaneously calculate the odds at follow-up of being dead versus robust, frail versus robust, or intermediate stage versus robust.

Base models were adjusted for age, race, and clinical site; then additionally adjusted for baseline frailty status (intermediate stage vs robust). In forming the multivariate model, prespecified characteristics that may be related to cysC level or frailty status (ie, body mass index, comorbidity burden, smoking status, IADL impairment, and serum albumin) were added to the base + baseline frailty status model. A test for linear trend was performed for predictor variables categorized in quartiles by expressing each predictor quartile as an ordinal variable. To explore the effect of mediation by inflammation, inflammatory markers (C-reactive protein, interleukin-6, and tumor necrosis factor-alpha) were added one at a time to the multivariate model for cysC to examine whether the results were similar in effect size or statistical significance.

In sensitivity analyses, we expressed cysC, $eGFR_{cysC}$, creatinine, $eGFR_{Cr}$, and $eGFR_{CysC\text{-}Cr}$ as continuous variables. In addition, we evaluated $eGFR_{cysC}$, $eGFR_{Cr}$, and $eGFR_{CysC\text{-}Cr}$ as binary variables of <60 mL/min/1.73 m^2 versus ≥60 mL/min/1.73 m^2 to examine the association of CKD with risks of progression of frailty status and death at follow-up. All significance levels reported were two sided and all analyses were conducted using SAS version 9.4 (SAS Institute, Cary, NC).

Results

The characteristics of the analytical cohort (*n* = 2,429) overall and by quartile (Q) of cysC (Q1: <0.91, Q2: 0.91 to <1.04, Q3: 1.04 to <1.22, Q4: ≥1.22 mg/L) are shown in Table 1. The mean age of the cohort was 75.7 years. 2,201 (90.6%) were Caucasian and 2,172 (89.5%) reported their health status as "good" or "excellent."

Among our cohort of men without overt frailty at baseline, the frailty status of 917 (37.8%) men was classified as robust, whereas that of 1,512 (62.5%) men was classified as intermediate stage. Mean (*SD*) was 1.09 (0.27) mg/L for serum cysC, 69.8 (19.3) mL/min/1.73 m^2 for $eGFR_{cysC}$, 1.05 (0.23) mL/dL for serum creatinine, and 71.5 (14.3) mL/min/1.73 m^2 for $eGFR_{Cr}$. Using a definition of CKD as an eGFR less than 60 mL/min/1.73 m^2, 787 (32.4%) men had CKD when eGFR was calculated using cysC, whereas 508 (20.9%) had CKD when eGFR was calculated using creatinine. Only 159 (6.6%) had moderate CKD with an $eGFR_{cysC-Cr}$ less than 45 mL/min/1.73 m^2 and only 16 (0.66%) had advanced CKD with an $eGFR_{cysC-Cr}$ less than 30 mL/min/1.73 m^2.

The mean time to follow-up was 3.4 (0.5) years. After adjusting for age, clinical site, and race, higher cysC was associated with greater odds of being classified as intermediate stage (vs robust), frail (vs robust), or dead (vs robust). Compared with men in the Q1 of cysC (referent), men in Q4 had a twofold greater odds of intermediate stage (OR 1.82, 95% confidence interval [CI] 1.35–2.45), a threefold greater odds of frailty (OR 3.13, 95% CI 2.03–4.82), and a more than fivefold greater odds of death (OR 5.48, 95% CI 2.98–10.08) (Table 2). The associations appeared graded in nature (p for linear trend across cysC quartiles ≤ .001).

After further adjustment for baseline frailty status, body mass index, smoking status, comorbidity burden, IADL impairments, and serum albumin, associations of cysC with frailty and death were attenuated but persisted. Compared with men in Q1 of cysC, men in Q4 had 60% higher odds of frailty (OR 1.60, 95% CI 0.98–2.60; p trend .045) and a nearly threefold greater odds of death (OR 2.94, 95% CI 1.50–5.76; p trend .005). However, the association between higher cysC and intermediate stage was no longer significant (Q4 vs Q1 OR 1.20, 95% CI 0.86–1.69; p trend .62). Further adjustment for inflammatory markers to the multivariate model did not substantially alter the association between cysC and frailty or death (data not shown). Results were similar when cysC was expressed as a continuous variable; each 1 *SD* increase in cysC was associated with a 1.3-fold odds of being frail (vs robust) and a 1.5-fold odds of being dead (vs robust) at follow-up (Table 2). The strength of the associations between higher cysC and greater odds of frailty and death at follow-up was nearly identical to that between lower $eGFR_{cysC}$ and these outcomes at follow-up (Supplementary Table 2).

Conversely, neither lower $eGFR_{Cr}$ (Supplementary Table 3) nor higher serum creatinine (data not shown) was associated with greater odds of intermediate stage, frailty, or death at follow-up in base or multivariable models (p value for linear trend across quartiles ranged from .19 to 0.67). When eGFR was calculated using a formula based on both serum cysC and serum creatinine, the inclusion of serum creatinine with cysC in the calculation appeared to slightly attenuate the association of eGFR with frailty status and death at follow-up, as the strength of the association between $eGFR_{cysC-Cr}$ and these outcomes was somewhat smaller in magnitude to that for $eGFR_{cysC}$ alone (Supplementary Table 4).

Discussion

In this cohort of community-dwelling older men who were not overtly frail at baseline, higher serum cysC and lower $eGFR_{cysC}$ were associated in a graded manner with greater odds of incident frailty or death at follow-up. However, we found no evidence of an association between higher serum creatinine (or lower $eGFR_{Cr}$) and risks of frailty or death at follow-up.

Our findings contribute to a growing body of literature suggesting that cysC is a more sensitive predictor for adverse age-related outcomes

Table 2. Association Between Cystatin C and Frailty Status and Death at Follow-Up

	Odds Ratio (95 % Confidence Interval)		
	Intermediate Stage vs Robust	Frail vs Robust	Dead vs Robust
Base model[a]			
Quartile 1	1.00 (referent)	1.00 (referent)	1.00 (referent)
Quartile 2	1.06 (0.82, 1.38)	1.10 (0.71, 1.71)	2.05 (1.10, 3.85)
Quartile 3	0.97 (0.75, 1.27)	1.48 (0.98, 2.25)	2.43 (1.33, 4.47)
Quartile 4	1.82 (1.35, 2.45)	3.13 (2.03, 4.82)	5.48 (2.98, 10.08)
p Trend	.001	<.001	<.001
Continuous, per 1 *SD* increase (0.27)	1.30 (1.15, 1.46)	1.69 (1.45, 1.97)	1.94 (1.64, 2.30)
Base model + Baseline frailty status			
Quartile 1	1.00 (referent)	1.00 (referent)	1.00 (referent)
Quartile 2	0.99 (0.75, 1.30)	0.99 (0.63, 1.57)	1.88 (1.00, 3.55)
Quartile 3	0.88 (0.67, 1.16)	1.29 (0.84, 2.00)	2.17 (1.17, 4.02)
Quartile 4	1.50 (1.10, 2.06)	2.40 (1.53, 3.77)	4.39 (2.37, 8.16)
p Trend	.07	<.001	<.001
Continuous, per 1 *SD* increase (0.27)	1.19 (1.05, 1.35)	1.52 (1.30, 1.78)	1.77 (1.48, 2.10)
Multivariable model[b]			
Quartile 1	1.00 (referent)	1.00 (referent)	1.00 (referent)
Quartile 2	0.95 (0.71, 1.27)	0.89 (0.55, 1.45)	1.80 (0.92, 3.52)
Quartile 3	0.84 (0.63, 1.13)	1.12 (0.71, 1.78)	1.89 (0.98, 3.65)
Quartile 4	1.20 (0.86, 1.69)	1.60 (0.98, 2.60)	2.94 (1.50, 5.76)
p Trend	.62	.045	.005
Continuous, per 1 *SD* increase (0.27)	1.09 (0.95, 1.25)	1.29 (1.08, 1.53)	1.54 (1.27, 1.87)

Note: Quartile ranges (mg/L): <0.91, 0.91 to <1.04, 1.04 to <1.22, ≥1.22.

IADL = instrumental activities of daily living.

[a]Adjusted for age, race, and clinic. [b]Adjusted for age, race, clinic, baseline frailty status (intermediate stage vs robust), body mass index, smoking (ever vs never), number of comorbidities, number of IADL impairments, and serum albumin.

than creatinine or creatinine-based eGFR (4,15,17,18,30,31). These results may in part be explained by the improved sensitivity of cysC in identifying reduced kidney function in older adults with less muscle mass, especially among those with mild-to-moderate reductions in function. Studies have also noted a J-shaped association between creatinine and poor outcomes (4,15–17), suggesting that low serum creatinine is often more indicative of low muscle mass (and presumably decreased biological reserve) than excellent kidney function and that population-based equations for eGFR fail to account for this phenomenon. Indeed, 11.5% of the cohort would be reclassified as having CKD as defined by $eGFR_{cysC}$ less than 60 mL/min/1.73 m^2 compared with CKD defined by $eGFR_{Cr}$.

A previous study reported that the accuracy of equations used to estimate true (directly measured) GFR is better when both cysC and creatinine are included compared with those based on either creatinine or cysC alone (13) and that analysis included almost 1,000 individuals older than 65 years. The benefit of combining cysC and creatinine to estimate GFR was again noted in a study of 610 adults aged 70 years and older (32). In our analysis, the association between $eGFR_{cysC\text{-}Cr}$ and risks of frailty or death actually appeared to be somewhat attenuated compared with that between $eGFR_{cysC}$ or cysC and these outcomes. These results suggest that the association of cysC with incident frailty and death is not entirely due to cysC being a marker of kidney function. In addition, the association of cysC with frailty or death was observed despite the cohort being predominantly comprised of men with at most mild reductions in kidney function; indeed, the lowest quartile of $eGFR_{cysC}$ included men with eGFR less than 55 mL/min/1.73 m^2. The concept that cysC may be a predictor of frailty beyond its use to define GFR was supported by an analysis of the CHS cohort by Dalrymple and colleagues (18). In that analysis, although the risks of frailty differed somewhat in magnitude between two different $eGFR_{cysC}$ prediction equations when eGFR was expressed as a categorical variable, the hazard ratio of risk of frailty per 10 mL/min/1.73 m^2 decrease in eGFR was nearly identical regardless of which $eGFR_{cysC}$ prediction equation was used.

Other potential mechanisms may drive the association of cysC with risks of frailty or death, including the role of inflammation (33). We have previously examined the effect of potential biological mediators, including calciotropic hormones and inflammatory markers, on the associations between cysC and between prevalent frailty status (4) in a cross-sectional analysis of the MrOS study cohort. Adjustment for these potential biological mediators had little, if any, impact on the association between cysC and prevalent frailty status. Similarly, in this longitudinal analysis, the association between cysC and frailty or death remained largely unchanged after further adjustment for inflammatory markers. It is possible that higher cysC is indicative of alterations in biochemical pathways that, in combination with mild decrements in kidney function, synergistically increase risk of frailty or death.

This study includes several strengths, including its large sample size and the inclusion of a comprehensively characterized cohort of community-dwelling older men who were not selected for CKD. We were able to prospectively evaluate a validated measure of frailty status as well as death and included an evaluation of frailty status as a continuum from robust to intermediate stage to frail to death. Failing to include death as a level of frailty could erroneously underestimate the association between cysC and poor outcomes, as men who died would not be classified as frail. However, this study has several limitations. We relied on surrogate markers of kidney function rather than direct measurements. Recent investigations indicate that equations using creatinine + cysC-based estimates of GFR are more accurate than those based on either marker alone, but error remains a concern particularly at the higher levels of GFR of ≥60 mL/min/1.73 m^2. The use of single measurements of cysC and creatinine may increase variability; however, this variability would likely bias our findings toward the null hypothesis of no association. Our cohort was comprised of community-dwelling, predominantly Caucasian men, and findings may not be generalizable to other population groups; our results should be confirmed in other cohort studies of older adults. Very few men in the MrOS cohort had advanced CKD, limiting our ability to assess the association between cysC and frailty or death among those with markedly impaired kidney function. Measures used to identify frailty status included some frailty components that were similar but not identical to the original CHS frailty index definition. However, previous studies have reported that the associations between this frailty index (8,9) and risk of adverse health outcomes are similar in magnitude to those between the CHS frailty index and risk of adverse outcomes. Finally, as with any observational study, the possibility for residual confounding persists.

In conclusion, higher serum cysC (and lower $eGFR_{cysC}$) among older men without overt frailty was associated with increased risks of progression of frailty status and death at follow-up. These relationships were not observed for serum creatinine or $eGFR_{Cr}$. These results suggest that cysC itself may be a promising biomarker for unsuccessful aging as manifested by increased risks of frailty and death.

Supplementary Material

Supplementary data is available at *The Journals of Gerontology, Series A: Biomedical Sciences and Medical Sciences* online.

Funding

The Osteoporotic Fractures in Men (MrOS) Study is supported by the National Institutes of Health funding. The following institutes provide support: the National Institute on Aging (NIA), the National Institute of Arthritis and Musculoskeletal and Skin Diseases (NIAMS), the National Center for Advancing Translational Sciences (NCATS), and NIH Roadmap for Medical Research under the following grant numbers: U01 AG027810, U01 AG042124, U01 AG042139, U01 AG042140, U01 AG042143, U01 AG042145, U01 AG042168, U01 AR066160, and UL1 TR000128.

The National Heart, Lung, and Blood Institute (NHLBI) provides funding for the MrOS Sleep ancillary study "Outcomes of Sleep Disorders in Older Men" under the following grant numbers: R01 HL071194, R01 HL070848, R01 HL070847, R01 HL070842, R01 HL070841, R01 HL070837, R01 HL070838, and R01 HL070839.

A.H. was also supported by the National Center for Advancing Translational Sciences in the National Institutes of Health Award Number UL1TR000114.

The funding agencies had no direct role in the conduct of the study, the collection, management, analyses, and interpretation of the data, or preparation or approval of the manuscript. This manuscript is the result of work supported with resources and use of facilities of the Minneapolis VA Health Care System.

Declaration of Interest

The authors report no declarations of interest.

References

1. Coresh J, Selvin E, Stevens LA, et al. Prevalence of chronic kidney disease in the United States. *JAMA*. 2007;298:2038–2047. doi:10.1001/jama.298.17.2038
2. Stevens LA, Li S, Wang C, et al. Prevalence of CKD and comorbid illness in elderly patients in the United States: results from the Kidney Early

Evaluation Program (KEEP). *Am J Kidney Dis*. 2010;55(3 suppl 2):S23–S33. doi:10.1053/j.ajkd.2009.09.035
3. Anand S, Johansen KL, Kurella TM. Aging and chronic kidney disease: the impact on physical function and cognition. *J Gerontol A Biol Sci Med Sci*. 2014;69:315–322. doi:10.1093/gerona/glt109
4. Hart A, Paudel ML, Taylor BC, et al. Cystatin C and frailty in older men. *J Am Geriatr Soc*. 2013;61:1530–1536. doi:10.1111/jgs.12413
5. Fried LP, Tangen CM, Walston J, et al. Frailty in older adults: evidence for a phenotype. *J Gerontol A Biol Sci Med Sci*. 2001;56:M146–M156. doi:10.1093/gerona/56.3.M146
6. Boyd CM, Xue QL, Simpson CF, Guralnik JM, Fried LP. Frailty, hospitalization, and progression of disability in a cohort of disabled older women. *Am J Med*. 2005;118:1225–1231. doi:10.1016/j.amjmed.2005.01.062
7. Bandeen-Roche K, Xue QL, Ferrucci L, et al. Phenotype of frailty: characterization in the women's health and aging studies. *J Gerontol A Biol Sci Med Sci*. 2006;61:262–266. doi:10.1093/gerona/61.3.262
8. Cawthon PM, Marshall LM, Michael Y, et al. Frailty in older men: prevalence, progression, and relationship with mortality. *J Am Geriatr Soc*. 2007;55:1216–1223. doi:10.1111/j.1532-5415.2007.01259.x
9. Ensrud KE, Ewing SK, Cawthon PM, et al. A comparison of frailty indexes for the prediction of falls, disability, fractures, and mortality in older men. *J Am Geriatr Soc*. 2009;57:492–498. doi:10.1111/j.1532-5415.2009.02137.x
10. Woods NF, LaCroix AZ, Gray SL, et al. Frailty: emergence and consequences in women aged 65 and older in the Women's Health Initiative Observational Study. *J Am Geriatr Soc*. 2005;53:1321–1330. doi:10.1111/j.1532-5415.2005.53405.x
11. Baxmann AC, Ahmed MS, Marques NC, et al. Influence of muscle mass and physical activity on serum and urinary creatinine and serum cystatin C. *Clin J Am Soc Nephrol*. 2008;3:348–354. doi:10.2215/CJN.02870707
12. Westhuyzen J. Cystatin C: a promising marker and predictor of impaired renal function. *Ann Clin Lab Sci*. 2006;36:387–394.
13. Inker LA, Schmid CH, Tighiouart H, et al. Estimating glomerular filtration rate from serum creatinine and cystatin C. *N Engl J Med*. 2012;367:20–29. doi:10.1056/NEJMoa1114248
14. Shlipak MG, Sarnak MJ, Katz R, et al. Cystatin C and the risk of death and cardiovascular events among elderly persons. *N Engl J Med*. 2005;352:2049–2060. doi:10.1056/NEJMoa043161
15. Shlipak MG, Matsushita K, Arnlov J, et al. Cystatin C versus creatinine in determining risk based on kidney function. *N Engl J Med*. 2013;369:932–943. doi:10.1056/NEJMoa1214234
16. Ensrud KE, Parimi N, Cauley JA, et al. Cystatin C and risk of hip fractures in older women. *J Bone Miner Res*. 2013;28:1275–1282. doi:10.1002/jbmr.1858
17. Ensrud KE, Parimi N, Fink HA, et al. Estimated GFR and risk of hip fracture in older men: comparison of associations using cystatin C and creatinine. *Am J Kidney Dis*. 2014;63:31–39. doi:10.1053/j.ajkd.2013.05.022
18. Dalrymple LS, Katz R, Rifkin DE, et al. Kidney function and prevalent and incident frailty. *Clin J Am Soc Nephrol*. 2013;8:2091–2099. doi:10.2215/CJN.02870313
19. Blank JB, Cawthon PM, Carrion-Petersen ML, et al. Overview of recruitment for the osteoporotic fractures in men study (MrOS). *Contemp Clin Trials*. 2005;26:557–568. doi:10.1016/j.cct.2005.05.005
20. Orwoll E, Blank JB, Barrett-Connor E, et al. Design and baseline characteristics of the osteoporotic fractures in men (MrOS) study–a large observational study of the determinants of fracture in older men. *Contemp Clin Trials*. 2005;26:569–585. doi:10.1016/j.cct.2005.05.006
21. Levey AS, Stevens LA, Schmid CH, et al. A new equation to estimate glomerular filtration rate. *Ann Intern Med*. 2009;150:604–612.
22. Kurella M, Chertow GM, Fried LF, et al. Chronic kidney disease and cognitive impairment in the elderly: the health, aging, and body composition study. *J Am Soc Nephrol*. 2005;16:2127–2133. doi:10.1681/ASN.2005010005
23. McDowell I, Kristjansson B, Hill GB, Hébert R. Community screening for dementia: the Mini Mental State Exam (MMSE) and Modified Mini-Mental State Exam (3MS) compared. *J Clin Epidemiol*. 1997;50:377–383.
24. Teng EL, Chui HC. The Modified Mini-Mental State (3MS) examination. *J Clin Psychiatry*. 1987;48:314–318.
25. Washburn RA, Smith KW, Jette AM, Janney CA. The Physical Activity Scale for the Elderly (PASE): development and evaluation. *J Clin Epidemiol*. 1993;46:153–162. doi:10.1016/0895-4356(93)90053-4
26. Cawthon PM, Fink HA, Barrett-Connor E, et al. Alcohol use, physical performance, and functional limitations in older men. *J Am Geriatr Soc*. 2007;55:212–220. doi:10.1111/j.1532-5415.2007.01062.x
27. Fitti JE, Kovar MG. The Supplement on Aging to the 1984 National Health Interview Survey. *Vital Health Stat 1*. 1987;21:1–115.
28. Pincus T, Summey JA, Soraci SA Jr, Wallston KA, Hummon NP. Assessment of patient satisfaction in activities of daily living using a modified Stanford Health Assessment Questionnaire. *Arthritis Rheum*. 1983;26:1346–1353. doi:10.1002/art.1780261107
29. Sheikh JI, Yesavage JA. Geriatric depression scale (GDS): recent evidence and development of a shorter version. *Clin Gerontol*. 1986;5:165–173. doi:10.1300/J018v05n01_09
30. Hart A, Ishani A, Paudel ML, et al. Cystatin C and risk of frailty and mortality in older men. *J Am Soc Nephrol*. 2012;23:402A.
31. Liu CK, Lyass A, Massaro JM, D'Agostino RB Sr, Fox CS, Murabito JM. Chronic kidney disease defined by cystatin C predicts mobility disability and changes in gait speed: the Framingham Offspring Study. *J Gerontol A Biol Sci Med Sci*. 2014;69:301–307. doi:10.1093/gerona/glt096
32. Schaeffner ES, Ebert N, Delanaye P, et al. Two novel equations to estimate kidney function in persons aged 70 years or older. *Ann Intern Med*. 2012;157:471–481. doi:10.7326/0003-4819-157-7-201210020-00003
33. Stevens LA, Schmid CH, Greene T, et al. Factors other than glomerular filtration rate affect serum cystatin C levels. *Kidney Int*. 2009;75:652–660. doi:10.1038/ki.2008.638