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Comparison of Measured GFR, Serum Creatinine, Cystatin C, and Beta-Trace Protein to Predict ESRD in African Americans With Hypertensive CKD

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Abstract

Background—Identification of persons with chronic kidney disease (CKD) who are at highest risk to progress to end stage renal disease (ESRD) is necessary to reduce the burden of kidney failure. The relative utility of traditional markers of kidney function, including estimated glomerular filtration rate (GFR) and serum creatinine, and emerging markers of kidney function, including cystatin C and beta-trace protein (BTP), to predict ESRD and mortality has yet to be established.

Study Design—Randomized clinical trial followed by an observational cohort study.

Setting & Participants—865 African American individuals with hypertensive CKD enrolled in a clinical trial of two levels of blood pressure control and three different antihypertensive drugs as initial therapy and subsequently followed by an observational cohort study.

Predictors—Quintile of measured GFR (mGFR) by iothalamate clearance, serum creatinine, serum creatinine-based estimated GFR (eGFR_{SCr}), cystatin C, and BTP.

Outcomes and Measurements—Incidence of ESRD and mortality.

Results—A total of 246 participants reached ESRD over a median follow-up of 102 months. The incidence rate of ESRD was higher with higher quintiles of each marker. The association between

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higher BTP and ESRD was stronger than those for the other markers, including mGFR. All the markers remained significantly associated with ESRD after adjustment for mGFR and relevant covariates (all $p < 0.05$), with BTP retaining the strongest association (HR for highest versus lowest quintile, 5.7; 95% CI, 2.2-14.9). Associations with the combined endpoint of ESRD or mortality ($n=390$) were weaker, but remained significant for cystatin C ($p=0.05$) and BTP ($p=0.004$).

Limitations—The ability of these markers to predict ESRD and mortality in other racial and ethnic groups and among individuals with CKD due to other causes is unknown.

Conclusions—Plasma BTP and cystatin C may be useful adjuncts to serum creatinine and mGFR in evaluating risk for progression of kidney disease.

Keywords

End-stage renal disease; beta trace protein; cystatin C; serum creatinine; iothalamate glomerular filtration rate

Decreased kidney function increases risk of end-stage renal disease (ESRD) and all-cause mortality.¹⁻³ ESRD, in turn, is associated with a substantially increased risk of mortality and morbidity.⁴ Accurate identification of persons with chronic kidney disease (CKD) who are at highest risk of progressing to ESRD is critical to tailor therapy to reduce its incidence and sequelae.

Directly determining measured glomerular filtration rate (mGFR) by iothalamate clearance is considered the gold standard method to assess kidney function. This procedure, however, is burdensome and impractical in many settings. Endogenous markers of kidney function are attractive alternatives in these settings. Serum creatinine and, more recently serum cystatin C, have most commonly been used to assess kidney function. Both markers, however, are influenced by factors other than kidney function, resulting in inaccurate estimates of GFR.⁵ For example, serum levels of creatinine are positively associated with greater muscle mass and dietary meat intake and inversely associated with concomitant illness.⁵ Although cystatin C is less sensitive to inter-individual differences in muscle mass, this marker is increased in persons with diabetes, inflammation, and higher body mass index.⁵

Beta trace protein (BTP) has been proposed as alternative marker of kidney function.⁶⁻⁸ BTP, also known as lipocalin prostaglandin D2 synthase, is a low molecular weight factor that is a member of the lipocalin protein family. Serum BTP levels were strongly correlated with GFR in small studies of kidney transplant patients and patients with CKD, though data are limited in other populations.^{9, 10} Limited data are available on the association of BTP levels with subsequent outcomes.

We compared the ability of mGFR, serum creatinine, estimated GFR based on serum creatinine ($eGFR_{SCR}$), cystatin C, and BTP to predict ESRD and mortality among African Americans with hypertensive CKD enrolled in the African American Study of Kidney Disease and Hypertension (AASK) clinical trial and cohort study.

Methods

Study Design

AASK was a 3×2 factorial multicenter randomized clinical trial conducted from 1995 to 2001. The study was designed to test the effects of three anti-hypertensive medications, an ACE-inhibitor (ramipril), a β -blocker (metoprolol), and a calcium channel blocker (amlodipine) as initial therapy in a drug regimen, and two levels of blood pressure control (low: mean arterial pressure (MAP) of 92 mmHg or lower, and usual: MAP of 102-107

mmHg) on progression of chronic kidney disease. The study population included 1,094 non-diabetic, self-identified African Americans, aged 18 to 70 years, with an mGFR assessed by iothalamate clearance between 20 and 65 mL/min/1.73 m² with no identified causes of chronic kidney disease other than hypertension.¹¹ At the conclusion of the trial, participants were invited to enroll in a cohort study, which was initiated to identify factors that predict progression of kidney disease within the setting of recommended management of CKD due to hypertension (i.e., use ACE inhibitors and ARB antihypertensive drugs and a low blood pressure goal (< 92 mmHg)).¹⁰ At the end of the clinical trial a total 759 participants who had not yet reached ESRD were being followed; 689 enrolled in the cohort study.¹² Incidence of ESRD was defined as beginning maintenance dialysis or receipt of a kidney transplant.^{13, 14} Study participants were seen in clinic at 3, 6, and every six months thereafter during the clinical trial and at baseline and every 12 months thereafter during the cohort study. Details of the design of the clinical trial and cohort study have been previously published.¹⁵

Measurements of GFR and Endogenous Markers

Kidney function was measured twice by iothalamate clearance at baseline of the clinical trial. We used the second of these measurements for comparison to the single measurements of the other markers, and the mean of these two measurements for adjusted analyses. Serum creatinine, cystatin C, and BTP also were measured from samples obtained at baseline. Serum creatinine was measured in a central laboratory with the rate-Jaffe method with an alkaline picrate assay. Cystatin C and BTP were measured by particle-enhanced nephelometry (Dade/Siemens). Estimated GFR based on serum creatinine was calculated using an equation specific to the AASK population.¹⁶

Other Measurements

The baseline visit consisted of a physical examination, questionnaires (including self-reporting of any history of cardiovascular disease), and 24-hour urine specimen collection on the day prior to their first pre-randomization mGFR,¹⁴ measurement of blood pressure (using Hawksley random-zero sphygmomanometers), and performance of a 12-lead electrocardiogram (EKG). Urinary protein excretion was expressed as the urinary protein-creatinine ratio from a 24-hour urine collection.

Statistical Analysis

Correlation of log(mGFR) with the inverse of each marker was assessed. Linear or logistic regression was used to test for trends across quintiles of mGFR. Event-free survival time was defined from time of randomization to ESRD, mortality, censoring or administrative withdrawal at the end of the study (June, 2007). ESRD was censored at death and mortality was censored at ESRD. Cox proportional hazards competing risk models were used to account for this informative censoring.^{17, 18} Multivariate models adjusted for age, sex, prevalent coronary heart disease, total high density lipoprotein (HDL) cholesterol, urinary protein-creatinine ratio, education, body mass index (BMI), smoking status, trial treatment group (i.e., blood pressure goal and anti-hypertensive medication), and mean baseline mGFR. The proportionality assumption was assessed using Schoenfeld residuals and log-log plots.¹⁹ Model discrimination was assessed using Harrell's C statistic for fully-adjusted models.²⁰

The continuous net reclassification improvement [NRI(>0)] at 102 months of follow-up (median follow-up) was calculated from fully-adjusted Poisson models and defined as the sum of those classified upward to higher risk among those with an event plus those classified downward to a lower risk among non-events less the sum of those classified downward to lower risk among those with an event plus those classified to a higher risk

among those without an event.²¹ We also report the NRI among those participants with an event ($\text{NRI}_{\text{event}}$) and without an event ($\text{NRI}_{\text{event-free}}$). These models did not account for informative censoring. All analyses were conducted using Stata 10.1 software.²²

Results

Baseline Characteristics

Serum creatinine, cystatin C, and BTP measurements were available for 865 participants at baseline. The mean age of participants was 54.8 years and 61% were male (**Table 1**). The mean mGFR was 46.5 mL/min/1.73 m². Lower mGFR was associated with younger age, female sex, and greater urinary protein-creatinine ratio. All of the other markers also were strongly associated with mGFR.

Serum creatinine had the weakest correlation with mGFR (0.65), whereas eGFR_{SCr} had the highest correlation (0.77; **Table 2**). Cystatin C and BTP were less well correlated with mGFR (0.73 and 0.71, respectively), but were most strongly correlated with each other (0.79).

Association with ESRD

A total of 246 participants reached ESRD over a mean follow-up of 102.7 months. Higher quintiles of each marker were strongly associated with ESRD, with cystatin C and BTP having the strongest associations, which reached statistical significance among the second quintile (**Table 3**). Adjustment for mGFR attenuated these associations, but each marker remained significantly associated with ESRD. Subsequent adjustment for covariates attenuated these associations further, but all remained significantly associated with ESRD, with BTP retaining the strongest association. Harrell's C statistic was slightly higher for BTP (0.857) than for mGFR (0.852), serum creatinine (0.849), eGFR_{SCr} (0.853), and cystatin C (0.851).

Net Reclassification Improvement (NRI) for ESRD

The addition of any individual marker significantly improved risk classification. The addition of eGFR quintiles to a model with mGFR and covariates had an $\text{NRI}(> 0)$ of 0.56 ($p < 0.001$; $\text{NRI}_{\text{event}} = 0.30$, $\text{NRI}_{\text{event-free}} = 0.26$). Including BTP resulted in an $\text{NRI}(> 0)$ of 0.40 ($p < 0.001$; $\text{NRI}_{\text{event}} = 0.27$, $\text{NRI}_{\text{event-free}} = 0.13$). Serum creatinine improved classification of those without an event ($\text{NRI}_{\text{event-free}} = 0.21$) more than those with an event ($\text{NRI}_{\text{event}} = 0.08$), whereas cystatin C improved classification of those with an event ($\text{NRI}_{\text{event}} = 0.35$), but not those without an event ($\text{NRI}_{\text{event-free}} = -0.10$).

Association with Mortality or ESRD

Results were similar for analyses of mortality or ESRD ($n=390$). Each marker was significantly associated with the combined outcome in crude analyses and after adjustment for mGFR, but only cystatin C and BTP remained significantly associated after further adjustment for covariates (**Table 4**). BTP was most strongly associated with the combined outcome. The models had lower discriminatory power for the combined outcome than for ESRD alone (Harrell's C for mGFR=0.769, compared to 0.754 for serum creatinine, 0.757 for eGFR_{SCr} , 0.758 for cystatin C and 0.761 for BTP).

Net Reclassification Improvement (NRI) for Mortality or ESRD

Improvements in reclassification also were lower for the combined outcome than for ESRD alone. Addition of BTP to a model with mGFR and covariates resulted in an $\text{NRI}(> 0)$ of 0.29 ($p < 0.001$; $\text{NRI}_{\text{event}} = 0.27$, $\text{NRI}_{\text{event-free}} = 0.02$). Additional of cystatin C improved

classification of those with an event ($\text{NRI}_{\text{event}} = 0.30$) but not those without an event ($\text{NRI}_{\text{event-free}} = -0.16$), as did eGFR_{SCr} ($\text{NRI}_{\text{event}} = 0.14$, $\text{NRI}_{\text{event-free}} = -0.036$), whereas serum creatinine improved classification of those without an event ($\text{NRI}_{\text{event-free}} = 0.22$), but not those with an event ($\text{NRI}_{\text{event}} = -0.09$).

Discussion

Identification of individuals with chronic kidney disease who are most likely to progress to ESRD or mortality is central in the clinical management of CKD. Such identification, however, is hampered by the shortcomings of current methods to assess kidney function by serum creatinine.^{23, 24} This has led to the search for other biomarkers of kidney function. Long-term follow-up of a cohort of African Americans with hypertensive chronic kidney disease afforded us the opportunity to compare the usefulness of mGFR, serum creatinine, eGFR_{SCr} , cystatin C, and BTP to predict ESRD and mortality.

Higher concentrations of each marker were significantly associated with higher risk of incident ESRD, but BTP predicted ESRD more strongly than did the other markers, including mGFR. Each of the other markers remained significantly associated with ESRD after adjustment for baseline mGFR, but BTP was again more strongly associated with ESRD than were the other markers. The addition of any marker to a model with covariates and mGFR significantly improved risk classification of study participants. Higher concentrations of each marker were also significantly associated with ESRD or mortality, but these associations were weaker than for ESRD alone. Again, BTP was more strongly associated with this outcome than were the other markers, remaining significant after adjustment for mGFR and relevant covariates.

Synthesis of beta-trace protein occurs predominately in the central nervous system and it is one of the main components of cerebrospinal fluid.²⁵ It may be a useful marker of kidney function because of its low molecular weight (23-29 kDa) and its lack of affinity for protein binding.^{25, 26} It acts as a prostaglandin D synthase, promoting conversion of prostaglandin H₂ to prostaglandin D₂.⁸ Previous studies on BTP and kidney function have reported that an eGFR equation based on serum BTP had lower bias and greater precision than eGFR based on serum creatinine.²⁷ It is uncertain, however, what non-filtration factors affect circulating levels of BTP.

A recent study of 227 European men and women with nondiabetic CKD and reduced kidney function at baseline explored the ability of serum creatinine, cystatin C, and BTP to predict CKD progression, defined as doubling of baseline creatinine and/or ESRD.²⁸ The concentrations of all three markers were significantly associated with GFR cross-sectionally. All three markers also were similarly predictive of CKD progression. Our results are in general agreement with these findings, though in our study BTP predicted ESRD more strongly than did cystatin C, serum creatinine, or eGFR_{SCr} . The results for serum creatinine as a predictor of outcomes in the previous study may be biased by using a change in serum creatinine as one component of the outcome definition. Other studies have reported that serum creatinine is less sensitive to small changes in kidney function compared to cystatin C and BTP.^{6, 29, 30} The range of GFR levels among the AASK population (i.e., 20-65 mL/min/1.73 m²), however, is generally within the range in which serum creatinine is a relatively accurate estimate of GFR.

Cystatin C has been proposed as a better marker of kidney function than serum creatinine, in part because it is less sensitive to differences in muscle mass.^{31, 32} Cystatin C is a stronger predictor of cardiovascular events, cardiovascular mortality, and all-cause mortality than is serum creatinine or eGFR based on serum creatinine.^{33, 34} Among 825 participants with

stage 3 or 4 non-diabetic CKD in the Modification of Diet in Renal Disease (MDRD) Study, 1/cystatin C had a stronger association with all-cause mortality and CVD mortality than did 1/creatinine.³¹ The associations of 1/creatinine with kidney failure, however, was slightly stronger than that for 1/cystatin C (relative risk per 1 standard deviation of 2.81 for 1/creatinine and 2.36 for 1/cystatin C). Similarly, in our study, the association between cystatin C and ESRD was generally similar to the association between serum creatinine and ESRD, but cystatin C was more strongly associated with the combined outcome of ESRD or death than was serum creatinine or eGFR_{SCr}.

A limited number of previous studies have found serum creatinine, cystatin C, and beta trace protein to better predict kidney outcomes than mGFR.^{28, 31} In our study, beta trace protein and cystatin C predicted ESRD more strongly than did mGFR. Each of the endogenous markers also predicted ESRD after adjustment for mGFR. This may be related to imprecision in mGFR measurement, including hourly and daily variations and deviations from measurement protocol.³⁵ Previous studies have shown that between-day coefficients of variation (CV) can vary between 6.3-16.6% when using I-iothalamate clearance.^{36, 37} We were not able to explicitly test this hypothesis. Alternatively, these markers may represent non-GFR factors that are related to progression to ESRD. It is important to note that the inclusion criteria for the AASK study required participants to have an mGFR between 20 and 65 mL/min per 1.73 m². This restricted range of mGFR values may limit the maximum statistical association between mGFR and outcomes. No such restriction was placed on the other markers, allowing them to have their full statistical association with outcomes. This may explain, in part, the residual association between these markers and outcomes after adjustment for mGFR.

In our study, ESRD was defined by kidney transplant or initiation of dialysis. As the decision to implement either treatment is likely to be influenced by the physicians' interpretation of the measured mGFR, serum creatinine, and eGFR, we could expect that the association between these markers and ESRD would be biased upwards. Despite this bias, higher BTP was more strongly associated with ESRD than these markers, even after adjustment for mGFR.

Each of the endogenous markers was reasonably well-correlated with mGFR. Serum creatinine had a weaker correlation with mGFR than did cystatin C or BTP. However, estimated GFR based on serum creatinine, age, and sex had the strongest correlation with mGFR. This may be due to the strong dependency of creatinine on muscle mass, for which age and sex act as proxies in the GFR-estimating equations. The correlation of eGFR_{SCr} with mGFR may be higher than in other studies, as the estimating equation used was developed in a pilot study for the AASK trial.¹⁶

The results of our analysis are promising but are limited by use of a select population. The AASK study enrolled African Americans with hypertensive kidney disease with no other identifiable cause of kidney disease. Further work needs to be done in a more generalizable population. The ability of these markers to predict mortality, cardiovascular disease, and other outcomes remains to be better studied. The use of the AASK study has distinctive advantages, as well. The study includes a high-risk group with well-controlled blood pressure. Baseline characteristics, including mGFR and proteinuria, were systematically collected and outcomes were carefully and thoroughly ascertained over a long follow-up period.

In summary, we found that BTP predicts ESRD more strongly than other markers of kidney function evaluated, including cystatin C, serum creatinine, eGFR_{SCr} and mGFR. The results of this study suggest that, although these endogenous markers have relatively similar

correlations with mGFR, the markers may be differentially associated with progression to ESRD, and that BTP and cystatin C may be useful adjuncts to serum creatinine or mGFR in evaluating risk for progression of kidney disease. Further work needs to be done to determine the best way to combine information from various markers to evaluate risk for specific adverse outcomes.

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References

1. Go AS, Chertow GM, Fan D, McCulloch CE, Hsu CY. Chronic kidney disease and the risks of death, cardiovascular events, and hospitalization. *The New England journal of medicine*. Sep 23; 2004 351(13):1296–1305. [PubMed: 15385656]
2. Klag MJ, Whelton PK, Randall BL, Neaton JD, Brancati FL, Stamler J. End-stage renal disease in African-American and white men. 16-year MRFIT findings. *JAMA*. Apr 23-30; 1997 277(16):1293–1298. [PubMed: 9109467]
3. Shlipak MG, Sarnak MJ, Katz R, et al. Cystatin C and the risk of death and cardiovascular events among elderly persons. *The New England journal of medicine*. May 19; 2005 352(20):2049–2060. [PubMed: 15901858]
4. US Renal Data System, USRDS 2009 Annual Data Report: Atlas of Chronic Kidney Disease and End-Stage Renal Disease in the United States. National Institutes of Health, National Institute of Diabetes and Digestive and Kidney Diseases; Bethesda, MD: 2009.
5. Stevens LA, Schmid CH, Greene T, et al. Factors other than glomerular filtration rate affect serum cystatin C levels. *Kidney Int*. Mar; 2009 75(6):652–660. [PubMed: 19119287]

6. Priem F, Althaus H, Birnbaum M, Sinha P, Conradt HS, Jung K. Beta-trace protein in serum: a new marker of glomerular filtration rate in the creatinine-blind range. *Clin Chem.* Apr; 1999 45(4):567–568. [PubMed: 10102918]
7. Kobata M, Shimizu A, Rinno H, et al. Beta-trace protein, a new marker of GFR, may predict the early prognostic stages of patients with type 2 diabetic nephropathy. *J Clin Lab Anal.* 2004; 18(4): 237–239. [PubMed: 15202116]
8. Melegos DN, Grass L, Pierratos A, Diamandis EP. Highly elevated levels of prostaglandin D synthase in the serum of patients with renal failure. *Urology.* Jan; 1999 53(1):32–37. [PubMed: 9886584]
9. Poge U, Gerhardt TM, Stoffel-Wagner B, et al. beta-Trace protein is an alternative marker for glomerular filtration rate in renal transplantation patients. *Clin Chem.* Aug. 2005; 51(8):1531–1533.
10. Donadio C, Lucchesi A, Ardini M, Donadio E, Giordani R. Serum levels of beta-trace protein and glomerular filtration rate--preliminary results. *J Pharm Biomed Anal.* Aug 8; 2003 32(4-5):1099–1104. [PubMed: 12900000]
11. Agodoa LY, Appel L, Bakris GL, et al. Effect of ramipril vs amlodipine on renal outcomes in hypertensive nephrosclerosis: a randomized controlled trial. *JAMA.* Jun 6; 2001 285(21):2719–2728. [PubMed: 11386927]
12. Lewis J, Greene T, Appel L, et al. A comparison of iothalamate-GFR and serum creatinine-based outcomes: acceleration in the rate of GFR decline in the African American Study of Kidney Disease and Hypertension. *J Am Soc Nephrol.* Dec; 2004 15(12):3175–3183. [PubMed: 15579521]
13. Appel LJ, Middleton J, Miller ER 3rd, et al. The rationale and design of the AASK cohort study. *J Am Soc Nephrol.* Jul; 2003 14(7 Suppl 2):S166–172. [PubMed: 12819323]
14. Gassman JJ, Greene T, Wright JT Jr. et al. Design and statistical aspects of the African American Study of Kidney Disease and Hypertension (AASK). *J Am Soc Nephrol.* Jul; 2003 14(7 Suppl 2):S154–165. [PubMed: 12819322]
15. Sika M, Lewis J, Douglas J, et al. Baseline characteristics of participants in the African American Study of Kidney Disease and Hypertension (AASK) Clinical Trial and Cohort Study. *Am J Kidney Dis.* Jul; 2007 50(1):78–89. 89, e71. [PubMed: 17591527]
16. Toto RD, Kirk KA, Coresh J, et al. Evaluation of serum creatinine for estimating glomerular filtration rate in African Americans with hypertensive nephrosclerosis: results from the African-American Study of Kidney Disease and Hypertension (AASK) Pilot Study. *J Am Soc Nephrol.* Feb; 1997 8(2):279–287. [PubMed: 9048347]
17. StataCorp. *stcrreg* command. Statistical Software: Release 11. StataCorp LP; College Station, Tx: 2009.
18. Fine JP. A Proportional Hazards Model for the Subdistribution of a Competing Risk. *Journa of the American Statistical Association.* 1999; 94(446):496–509.
19. Stucky CC, Gray RJ, Wasif N, Dueck AC, Pockaj BA. Increase in contralateral prophylactic mastectomy: echoes of a bygone era? Surgical trends for unilateral breast cancer. *Ann Surg Oncol.* Oct; 2010 17(Suppl 3):330–337. [PubMed: 20853055]
20. Harrell FE Jr. Lee KL, Mark DB. Multivariable prognostic models: issues in developing models, evaluating assumptions and adequacy, and measuring and reducing errors. *Stat Med.* Feb 28; 1996 15(4):361–387. [PubMed: 8668867]
21. Pencina MJ, D'Agostino RB Sr. Steyerberg EW. Extensions of net reclassification improvement calculations to measure usefulness of new biomarkers. *Stat Med.* Jan 15; 2011 30(1):11–21. [PubMed: 21204120]
22. StataCorp. *Stata Statistical Software: Release 11.* StataCorp LP; College Station, Tx: 2009.
23. Hsu CY, Chertow GM, Curhan GC. Methodological issues in studying the epidemiology of mild to moderate chronic renal insufficiency. *Kidney Int.* May; 2002 61(5):1567–1576. [PubMed: 11967006]
24. Shemesh O, Golbetz H, Kriss JP, Myers BD. Limitations of creatinine as a filtration marker in glomerulopathic patients. *Kidney Int.* Nov; 1985 28(5):830–838. [PubMed: 2418254]

25. Bokenkamp A, Franke I, Schlieber M, et al. Beta-trace protein--a marker of kidney function in children: "Original research communication-clinical investigation". *Clin Biochem. Sep; 2007* 40(13-14):969-975. [PubMed: 17588556]
26. Hoffmann A, Nimtz M, Conradt HS. Molecular characterization of beta-trace protein in human serum and urine: a potential diagnostic marker for renal diseases. *Glycobiology. Jun; 1997* 7(4): 499-506. [PubMed: 9184830]
27. White CA, Akbari A, Doucette S, et al. A novel equation to estimate glomerular filtration rate using beta-trace protein. *Clin Chem. Nov; 2007* 53(11):1965-1968. [PubMed: 17761751]
28. Spanaus KS, Kollerits B, Ritz E, Hersberger M, Kronenberg F, von Eckardstein A. Serum creatinine, cystatin C, and beta-trace protein in diagnostic staging and predicting progression of primary nondiabetic chronic kidney disease. *Clin Chem. May; 2010* 56(5):740-749. [PubMed: 20224047]
29. Mussap M, Dalla Vestra M, Fioretto P, et al. Cystatin C is a more sensitive marker than creatinine for the estimation of GFR in type 2 diabetic patients. *Kidney Int. Apr; 2002* 61(4):1453-1461. [PubMed: 11918752]
30. Laterza OF, Price CP, Scott MG. Cystatin C: an improved estimator of glomerular filtration rate? *Clin Chem. May; 2002* 48(5):699-707. [PubMed: 11978596]
31. Menon V, Shlipak MG, Wang X, et al. Cystatin C as a risk factor for outcomes in chronic kidney disease. *Annals of internal medicine. Jul 3; 2007* 147(1):19-27. [PubMed: 17606957]
32. Stevens LA, Coresh J, Schmid CH, et al. Estimating GFR using serum cystatin C alone and in combination with serum creatinine: a pooled analysis of 3,418 individuals with CKD. *Am J Kidney Dis. Mar; 2008* 51(3):395-406. [PubMed: 18295055]
33. Madero M, Sarnak MJ. Association of cystatin C with adverse outcomes. *Curr Opin Nephrol Hypertens. May; 2009* 18(3):258-263. [PubMed: 19374014]
34. Astor BC, Levey AS, Stevens LA, Van Lente F, Selvin E, Coresh J. Method of glomerular filtration rate estimation affects prediction of mortality risk. *J Am Soc Nephrol. Oct; 2009* 20(10): 2214-2222. [PubMed: 19762497]
35. Kwong YT, Stevens LA, Selvin E, et al. Imprecision of urinary iothalamate clearance as a gold-standard measure of GFR decreases the diagnostic accuracy of kidney function estimating equations. *Am J Kidney Dis. Jul; 2010* 56(1):39-49. [PubMed: 20537455]
36. Levey AS, Greene T, Schluchter MD, et al. Glomerular filtration rate measurements in clinical trials. Modification of Diet in Renal Disease Study Group and the Diabetes Control and Complications Trial Research Group. *J Am Soc Nephrol. Nov; 1993* 4(5):1159-1171. [PubMed: 8305642]
37. Perrone RD, Steinman TI, Beck GJ, et al. Utility of radioisotopic filtration markers in chronic renal insufficiency: simultaneous comparison of ¹²⁵I-iothalamate, ¹⁶⁹Yb-DTPA, ^{99m}Tc-DTPA, and inulin. The Modification of Diet in Renal Disease Study. *Am J Kidney Dis. Sep; 1990* 16(3):224-235. [PubMed: 2205098]

Table 1

Baseline characteristics

	Quintile of mGFR (in mL/min/1.73 m ²)					p-trend
	1 (≥58.8)	2 (51.1-58.7)	3 (43.2-51.1)	4 (32.4-43.1)	5 (<32.3)	
No.	173	173	173	173	173	-
Age, years	55.9 +/- 10.9	56.7 +/- 9.6	55.5 +/- 10.3	53.1 +/- 10.0	52.7 +/- 11.6	<0.001
Female	30.1	35.2	42.8	45.1	41.6	0.01
Prevalent heart disease	49.1	47.4	56.1	55.5	50.3	0.4
Current smoking	24.3	27.2	27.2	32.9	29.5	0.1
Body mass index, kg/m ²	30.6 +/- 5.8	31.0 +/- 6.3	30.6 +/- 6.0	30.2 +/- 6.8	30.1 +/- 6.9	0.2
Less than high school education	34.1	46.8	42.2	37.0	36.4	0.7
Non-HDL cholesterol, mg/dL	163.6 +/- 41.7	166.6 +/- 44.2	165.7 +/- 44.6	157.7 +/- 42.3	163.8 +/- 49.8	0.4
UPCR (mg/g)	3.7 [1.9; 9.4]	4.3 [2.2; 11.3]	5.7 [2.6; 24.7]	13.4 [4.6; 57.7]	40.6 [11.2; 106.8]	<0.001
mGFR, mL/min/1.73 m ²	62.3 +/- 2.6	55.0 +/- 2.2	47.4 +/- 2.2	37.7 +/- 3.1	26.9 +/- 3.5	-
SCr, mg/dL	1.54 +/- 0.31	1.64 +/- 0.33	1.79 +/- 0.47	2.18 +/- 0.58	2.94 +/- 0.78	<0.001
eGFR _{SCr} , mL/min/1.73 m ²	54.5 +/- 7.1	50.7 +/- 7.0	47.0 +/- 7.3	40.1 +/- 7/2	31.5 +/- 6.1	<0.001
SCysC, mg/L	1.24 +/- 0.22	1.37 +/- 0.28	1.52 +/- 0.30	1.88 +/- 0.42	2.43 +/- 0.53	<0.001
BTP, mg/dL	0.94 +/- 0.20	1.06 +/- 0.28	1.21 +/- 0.30	1.55 +/- 0.44	2.17 +/- 0.60	<0.001

Continuous variables are given as mean +/- standard deviation or median [25th; 75th percentile]; categorical variables as percentage.

eGFR_{SCr}: Estimated glomerular filtration rate based on serum creatinine; HDL, high-density lipoprotein; mGFR, measured glomerular filtration rate; UPCR, urinary protein-creatinine ratio; SCr, serum creatinine; SCysC, serum cystatin C; BTP, beta-trace protein.

Conversion factors for units: non-HDL cholesterol in mg/dL to mmol/L, x0.02586, SCr in mg/dL to micromol/L, x88.4; GFR in mL/min/1.73 m² to mL/s/1.73 m², x0.01667

Table 2

Correlation among kidney function markers

	Log (mGFR)	1 / SCr	Log (eGFR _{SCr})	1 / SCysC	1 / BTP	mGFR
1 / SCr	0.66	1.00	-	-	-	-
Log (eGFR _{SCr})	0.78	0.86	1.00	-	-	-
1 / SCysC	0.72	0.70	0.73	1.00	-	-
1 / BTP	0.71	0.70	0.71	0.79	1.00	-
mGFR	0.99	0.65	0.77	0.73	0.71	1.00

mGFR: measured glomerular filtration rate; BTP: beta-trace protein; SCr, serum creatinine; SCysC, serum cystatin C; eGFR_{SCr}: estimated glomerular filtration rate based on serum creatinine

Table 3

Incidence rate ratio for ESRD.

	Quintile of Kidney Function Marker					p-trend
	1	2	3	4	5	
<u>Unadjusted</u>						
mGFR	Ref	1.38 (0.71, 2.68)	2.68 (1.48, 4.84)	5.41 (3.12, 9.39)	12.96 (7.52, 22.31)	<0.001
SCr	Ref	1.36 (0.77, 2.41)	1.90 (1.07, 3.38)	4.64 (2.79, 7.73)	10.57 (6.54, 17.11)	<0.001
eGFR _{SCr}	Ref	1.28 (0.65, 2.50)	2.44 (1.34, 4.47)	4.90 (2.80, 8.60)	13.70 (7.99, 23.49)	<0.001
SCysC	Ref	3.92 (1.79, 8.59)	4.77 (2.21, 10.29)	10.78 (5.17, 22.45)	25.27 (12.25, 52.12)	<0.001
BTP	Ref	3.96 (1.62, 9.69)	7.21 (3.06, 17.00)	13.22 (5.72, 30.53)	37.75 (16.59, 85.90)	<0.001
<u>Adjusted for mGFR</u>						
SCr	Ref	1.09 (0.61, 1.94)	1.22 (0.67, 2.21)	1.90 (1.08, 3.33)	2.92 (1.66, 5.14)	<0.001
eGFR _{SCr}	Ref	1.01 (0.51, 2.00)	1.47 (0.79, 2.73)	1.96 (1.05, 3.64)	3.65 (1.90, 7.01)	<0.001
SCysC	Ref	3.70 (1.68, 8.18)	2.90 (1.32, 6.37)	4.45 (2.05, 9.65)	5.85 (2.59, 13.24)	<0.001
BTP	Ref	3.68 (1.50, 9.04)	5.10 (2.13, 12.22)	6.69 (2.76, 16.20)	13.10 (5.30, 32.42)	<0.001
<u>* Adjusted for covariates^a and mGFR</u>						
SCr	Ref	0.85 (0.47, 1.55)	0.97 (0.52, 1.81)	1.15 (0.62, 2.14)	1.67 (0.83, 3.37)	0.03
eGFR _{SCr}	Ref	1.15 (0.57, 2.30)	1.30 (0.67, 2.50)	1.81 (0.94, 3.51)	3.13 (1.57, 6.26)	<0.001
SCysC	Ref	2.94 (1.26, 6.83)	2.04 (0.88, 4.72)	2.17 (0.92, 5.12)	3.12 (1.27, 7.62)	0.04
BTP	Ref	3.22 (1.27, 8.16)	3.02 (1.21, 7.49)	3.69 (1.46, 9.33)	5.70 (2.19, 14.87)	<0.001

Values shown are incidence rate ratio (95% confidence interval).

eGFR_{SCr}, serum creatinine-based estimated glomerular filtration rate; BTP, Beta-trace Protein; SCysC, serum cystatin C; SCr, serum creatinine; mGFR, measured glomerular filtration rate; ESRD, end-stage renal disease.

* Adjusted for age, sex, prevalent coronary heart disease, total high density lipoprotein cholesterol, urinary protein-creatinine ratio, education, body mass index, smoking status, blood pressure goal, anti-hypertensive medication, and mean baseline glomerular filtration rate

Table 4

Incidence rate ratio for ESRD or mortality.

		Quintile of Kidney Function Marker				p-trend
	1	2	3	4	5	
Unadjusted						
mGFR	Ref	1.71 (1.10, 2.71)	2.61 (1.71, 4.01)	5.00 (3.35, 7.46)	9.90 (6.68, 14.67)	<0.001
SCr	Ref	1.44 (0.98, 2.11)	1.71 (1.15, 2.54)	3.64 (2.54, 5.22)	5.88 (4.17, 8.29)	<0.001
eGFR _{SCr}	Ref	1.09 (0.72, 1.66)	2.00 (1.37, 2.93)	3.14 (2.19, 4.51)	6.08 (4.29, 8.62)	<0.001
SCysC	Ref	1.91 (1.23, 2.96)	2.54 (1.67, 3.87)	4.89 (3.29, 7.28)	9.52 (6.45, 14.05)	<0.001
BTP	Ref	1.99 (1.28, 3.11)	2.76 (1.81, 4.29)	4.86 (3.22, 7.33)	11.18 (7.51, 16.65)	<0.001
Adjusted for mGFR						
SCr	Ref	1.18 (0.81, 1.74)	1.14 (0.76, 1.71)	1.66 (1.11, 2.47)	1.78 (1.17, 2.72)	0.002
eGFR _{SCr}	Ref	0.84 (0.55, 1.28)	1.16 (0.78, 1.73)	1.21 (0.79, 1.85)	1.44 (0.89, 2.31)	0.04
SCysC	Ref	1.79 (1.15, 2.78)	1.69 (1.09, 2.60)	2.41 (1.55, 3.73)	2.88 (1.75, 4.75)	<0.001
BTP	Ref	1.79 (1.15, 2.80)	1.99 (1.28, 3.10)	2.57 (1.63, 4.04)	4.05 (2.46, 6.66)	<0.001
Adjusted for covariates* and mGFR						
SCr	Ref	0.98 (0.65, 1.47)	0.97 (0.63, 1.51)	1.17 (0.74, 1.85)	1.25 (0.74, 2.13)	0.25
eGFR _{SCr}	Ref	0.84 (0.55, 1.28)	1.14 (0.76, 1.70)	1.16 (0.76, 1.78)	1.41 (0.87, 2.29)	0.07
SCysC	Ref	1.38 (0.88, 2.16)	1.41 (0.91, 2.19)	1.50 (0.95, 2.37)	1.77 (1.06, 2.97)	0.05
BTP	Ref	1.63 (1.04, 2.56)	1.56 (1.00, 2.44)	1.76 (1.10, 2.80)	2.32 (1.38, 3.92)	0.004

Values shown are incidence rate ratio (95% confidence interval).

eGFR_{SCr}, serum creatinine-based estimated glomerular filtration rate; BTP, Beta-trace Protein; SCysC, serum cystatin C; SCr, serum creatinine; mGFR, measured glomerular filtration rate; ESRD, end-stage renal disease.

* Adjusted for age, sex, prevalent coronary heart disease, total high density lipoprotein cholesterol, urinary protein-creatinine ratio, education, body mass index, smoking status, blood pressure goal, anti-hypertensive medication, and mean baseline glomerular filtration rate