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Associations of Urine Epidermal Growth Factor With Kidney and Cardiovascular Outcomes in Individuals With CKD in SPRINT

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Introduction: Urine epidermal growth factor (uEGF) has been found to be inversely associated with kidney function loss, whereas its associations with cardiovascular disease (CVD) and mortality have not been studied.

Methods: We measured baseline uEGF levels among 2346 Systolic Blood Pressure Intervention Trial (SPRINT) participants with an estimated glomerular filtration rate (eGFR) $<$ 60 ml/min per 1.73 m^2. A linear mixed-effects model was used to investigate the associations of uEGF with the annual eGFR change; Cox proportional hazards regression models were used to analyze its associations with the $\geq$30% eGFR decline, CVD, and all-cause mortality outcomes. To account for the competing risk of death, the Fine and Gray method was utilized for acute kidney injury (AKI) and end-stage kidney disease (ESKD) outcomes.

Results: At baseline, the study participants had mean age of 73 $\pm$ 9 years, mean eGFR of 46 $\pm$ 11 ml/min per 1.73 m^2, and median urine albumin-to-creatinine ratio (UACR) of 15 mg/g (interquartile range: 7–49). In the multivariable-adjusted analysis including baseline urine albumin and eGFR, each 50% lower uEGF concentration was associated with 0.74% (95% confidence interval [CI]: 0.29–1.19) per year faster decline in eGFR and 1.17 times higher risk of $\geq$30% eGFR decline (95% CI: 1.00–1.36). Lower uEGF concentrations were found to be associated with increased risks of ESKD, AKI, CVD, and all-cause mortality; however, these associations did not reach statistical significance when the models were controlled for baseline urine albumin and eGFR.

Conclusion: Among hypertensive adults with chronic kidney disease (CKD), lower baseline uEGF concentration was associated with faster eGFR decline independent of baseline albuminuria and eGFR; but not with ESKD, AKI, CVD, and all-cause mortality.

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CKD poses a worldwide health challenge due to its high prevalence and strong independent associations with kidney failure, CVD, and early death.[1] However, the underlying mechanisms by which CKD contributes to these adverse outcomes are not fully understood. This limitation in our understanding is partly due to the reliance on currently available clinical measures of kidney health, urine albumin and serum creatinine, because they do not fully capture kidney health status and primarily reflect the low-energy, circulation-driven filtration process and ignore the many homeostatic controls of the kidney tubules. A major goal of modern kidney research is to identify biomarkers that reflect the underlying biological processes in the tubules, thereby potentially improving

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aspects of CKD diagnosis, prognosis, and ultimately treatment.

Previous research has established associations of urine biomarkers of kidney tubular health with kidney and CVD outcomes.[2-6] For example, higher concentrations of biomarkers reflecting tubular injury, dysfunction, fibrosis, and impaired secretion are all shown to be associated with adverse CKD outcomes independent of eGFR and albuminuria.[7-10] Another biomarker, epidermal growth factor (EGF), has been shown in animal studies to be important for maintaining the normal functions of glomeruli, proximal, and distal convoluted tubules, as well as a promising indicator of cellular growth and tissue repair following ischemic injury.[11-13] Building on these findings, human studies have demonstrated an association between higher uEGF levels and slower kidney function loss.[14,15] Further, a study using kidney biopsy tissue transcriptomics has shown that higher intrarenal EGF mRNA and uEGF concentrations are associated with a lower risk of CKD progression.[16] However, it is unknown whether uEGF provides insight into the etiology of increased risk of CVD and mortality among persons with CKD.

In this study, we investigated the associations of uEGF concentrations with the risk of CKD progression, AKI, CVD, and all-cause mortality among SPRINT participants with established CKD. We hypothesized that lower concentrations of uEGF, indicating impaired kidney tubule health, would be associated with a higher risk of each outcome independent of eGFR and albuminuria.

METHODS

Study Population and Design

The SPRINT was a randomized, controlled, open-label study conducted at 102 clinical sites throughout the United States, including Puerto Rico. The design and results of the parent study have been published previously.[17] Inclusion criteria were age 50 to 85 years, systolic blood pressure (SBP) of 130 to 180 mm Hg, and high CVD risk. Exclusion criteria included diabetes mellitus, polycystic kidney disease, proteinuria $>$ 1g/d, previous stroke, recent heart failure exacerbation, and left ventricular ejection fraction $<$ 35%. The 9361 eligible participants were randomized to "standard" (target SBP $<$ 140 mm Hg) or "intensive" (target SBP $<$ 120 mm Hg) treatment groups. Initially, participants had monthly visits for the first 3 months, followed by every 3 months thereafter. Institutional Review Boards of all participating institutions approved the study.

In this ancillary study, we included participants with prevalent CKD, defined as eGFR $<$ 60 ml/min per 1.73m^2 by the 2012 CKD-Epidemiology Collaboration equation for creatinine and cystatin C ($N = 2514$) at the baseline visit.[18] We excluded 168 participants without available urine samples from the baseline visit, leaving 2346 (93%) participants for analysis.

Urine EGF

Urine specimens collected at the baseline visit were stored at −80 °C until biomarker measurement without any prior thawing. Urine EGF was measured at the Kidney Health Research Collaborative Laboratory using the Meso Scale Discovery platform (Rockville, MD) by personnel blinded to clinical information. The average interassay and intraassay coefficients of variation were 6.4 and 1.1%, respectively. Duplicate measurements of uEGF concentration were taken for each urine sample, and the results were averaged to enhance precision.

Outcomes

Outcomes of this study included annualized percentage change in eGFR, binary ≥30% decline in eGFR, ESKD, AKI, CVD, and all-cause mortality. Annualized percent change in eGFR was calculated using the 2012 CKD-Epidemiology Collaboration equation for creatinine to be consistent with the SPRINT trial results.[18] Serum creatinine measurements were conducted at baseline, month 1, month 3, and then at intervals of every 3 to 6 months during the study period. The median number of creatinine measurements per participant was 9 (interquartile range = 8–11). ESKD was defined as kidney failure requiring dialysis or transplantation. Participants were diagnosed with AKI if the hospital discharge summary listed AKI as one of the top 3 reasons for admission or continued hospitalization, with a few cases of AKI noted in the emergency department if the participant presented for one of the other conditions of interest.[19] The primary CVD composite end point included myocardial infarction, acute coronary syndrome not resulting in myocardial infarction, stroke, acute decompensated heart failure, or death from cardiovascular causes.

Covariates

We included the following confounders: age, sex, race/ethnicity (White, Black, Hispanic, Other), randomized treatment assignment, smoking status (never, former, current), history of CVD, baseline SBP and diastolic blood pressure, total cholesterol, high-density lipoprotein, antihypertensive medications at baseline (angiotensin-converting enzyme inhibitor, angiotensin II receptor blocker, beta blocker, calcium channel blocker, and diuretics), urine albumin, urine creatinine, and baseline eGFR.

Information on age, sex, race/ethnicity, medical history, medications, and smoking status was gathered through a questionnaire. Blood pressure was measured per SPRINT study protocol. Fasting serum total cholesterol, high-density lipoprotein, urine albumin, and creatinine were measured at the SPRINT Central Laboratory. Urine creatinine and albumin measurements were performed by an enzymatic procedure (Roche, Indianapolis, IN) and by a nephelometric method (Siemens, Tarrytown, NY), respectively.

Statistical Analysis

We described baseline demographics and clinical characteristics across quartiles of uEGF concentrations. Using Spearman's correlation test, we calculated correlations of uEGF/Cr with eGFR and UACR.

Prior to analyses, uEGF, urine creatinine, and urine albumin were log-base-2 transformed due to their skewed distributions. Urine EGF was modeled as a log-linear, continuous predictor and categorized into quartiles. To investigate the relationship between annual percent change in eGFR and uEGF, we used restricted cubic splines with the specified sequence of interior knots placed at the quartiles of the distributions of uEGF. We used linear mixed-effect models to evaluate associations of uEGF concentrations with the annualized percentage change in eGFR. These models incorporated random intercepts and slopes to account for intraindividual correlations. Given our hypotheses that lower uEGF would be associated with higher risk of outcomes, the highest quartile was assigned as the reference group.

We evaluated associations of uEGF concentration with ≥30% eGFR decline, CVD composite outcome, and all-cause mortality using Cox proportional hazards regression models. We evaluated uEGF associations with AKI and ESKD using the Fine and Gray method to account for the competing risk of death and reported subdistribution hazard ratios.[20] Potential confounders were adjusted in the following nested models: model 1 adjusted for age, sex, race, randomized treatment arm, and urine creatinine; model 2 additionally adjusted for smoking status, history of CVD, SBP, diastolic blood pressure, total cholesterol, high-density lipoprotein, angiotensin-converting enzyme inhibitor, angiotensin II receptor blocker, beta blocker, calcium channel blocker, and diuretics; and model 3 additionally adjusted for baseline urine albumin and eGFR. In our primary models, rather than indexing for urine creatinine, we adjusted the models for urine creatinine; we believe that the ratio variable of uEGF/Cr has greater potential for spurious associations. Using a ratio variable in statistical modeling can lead to misleading interpretations and oversimplify the relationship between variables. By independently assessing urine creatinine and uEGF, we aimed to provide a more accurate understanding of their contributions to the study outcomes.[21] However, studies examining the effects of indexing versus adjusting for urine creatinine on associations of urine biomarkers with CVD events, mortality, and kidney outcomes have usually shown similar findings between models indexed and adjusted for urine creatinine.[22,23] We conducted models with the uEGF/Cr ratio to compare our results. In addition, we used likelihood ratio tests to evaluate the interaction between uEGF and randomized treatment arm for each outcome.

All analyses were conducted using Stata version 18.0 (College Station, TX) and SPSS (IBM SPSS Statistics for Windows, Version 28.0, IBM Corp, Armonk, NY).

RESULTS

Study Population and Baseline Characteristics

Among the 2346 SPRINT participants with baseline CKD included in the present analysis, the mean age was 73 ± 9 years, 41% were female, and 25% were Black. The mean eGFR was 46 ± 11 ml/min per 1.73 m^2 and the median UACR was 15 mg/g (interquartile range: 7–49). Baseline characteristics of the study population across quartiles of uEGF are shown in Table 1. Compared to participants in higher uEGF quartiles, participants in the lowest quartile were more likely to be Black and to have prevalent CVD. In addition, there was a progressive decrease in the mean eGFR and an increase in the median UACR across descending uEGF quartiles.

Associations of uEGF With Kidney Outcomes

Urine EGF/Cr had moderately strong correlations with eGFR (r = +0.54) and UACR (r = −0.29). The annual percentage decline in eGFR was approximately 1% in Q2 to Q4 of uEGF and 3.33% for Q1 (Figure 1). The associations of uEGF concentrations with kidney outcomes are summarized in Table 2 and Figure 2. The results of the regression models indexed for urine creatinine are depicted in Supplementary Table S1. The magnitude of associations of uEGF with all kidney outcomes gradually increased with the descending uEGF quartiles across the sequence of models when uEGF was modeled in quartiles.

In models adjusting for demographics, randomized treatment arm, and clinical characteristics, each 50% lower concentration of uEGF was associated with 1.41% (95% CI: 1.06–1.76) faster decline in eGFR annually. However, upon adding baseline urine albumin and eGFR to the model, the estimate reduced by almost half, with each 50% lower concentration of uEGF being associated with 0.74% (95% CI: 0.29–1.19)

Table 1. Summary of demographic and clinical characteristics of the SPRINT participants with prevalent CKD, stratified by quartile of uEGF

Range (pg/ml) *N*	uEGF Q1 (≤ 1725) 586	uEGF Q2 (1726–3174) 587	uEGF Q3 (3175–5401) 587	uEGF Q4 (> 5401) 586	Total 2346
Age, yr	73 (10)	74 (9)	74 (9)	73 (8)	73 (9)
Female	252 (43)	232 (40)	227 (39)	253 (43)	964 (41)
Race					
White	358 (61)	397 (68)	400 (68)	408 (70)	1563 (67)
Black	164 (28)	146 (25)	136 (23)	137 (23)	583 (25)
Hispanic	53 (9)	36 (6)	39 (7)	35 (6)	163 (7)
Other	11 (2)	8 (1)	12 (2)	6 (1)	37 (2)
Randomized treatment arm					
Standard arm	274 (47)	296 (50)	296 (50)	285 (49)	1151 (49)
Intensive arm	312 (53)	291 (50)	291 (50)	301 (51)	1195 (51)
Smoking status					
Never	249 (42)	269 (46)	277 (47)	282 (48)	1077 (46)
Former	276 (47)	269 (46)	266 (45)	257 (44)	1068 (46)
Current	61 (10)	49 (8)	44 (8)	47 (8)	201 (9)
BMI, kg/m^2	29.2 (5.7)	29.3 (6.0)	29.7 (6.0)	30.0 (5.6)	29.5 (5.8)
Prevalent CVD	156 (27)	141 (24)	151 (26)	140 (24)	588 (25)
SBP	142 (17)	140 (17)	138 (16)	139 (16)	140 (16)
DBP	74 (12)	74 (12)	74 (12)	76 (12)	74 (12)
Number of antihypertensive meds	2 [2,3]	2 [1,3]	2 [1,3]	2 [1,3]	2 [1,3]
Anti-hypertensive meds					
ACEi/ARB	364 (62)	385 (66)	362 (62)	349 (60)	1460 (62)
BB	321 (53)	253 (43)	283 (48)	247 (42)	1095 (47)
CCB	269 (46)	234 (40)	220 (38)	222 (39)	945 (40)
Diuretic	355 (61)	315 (54)	311 (53)	298 (51)	1279 (55)
Total cholesterol, mg/dl	183 (40)	184 (41)	185 (40)	183 (42)	184 (41)
HDL, mg/dl	52 (15)	54 (15)	52 (14)	51 (13)	52 (14)
eGFR, ml/min per 1.73 m^2	38 (11)	45 (9)	48 (9)	51 (7)	46 (11)
UACR, mg/g	40 [14,193]	16 [7, 51]	11 [6, 27]	10 [6, 22]	15 [7, 49]

ACEi, angiotensin converting enzyme inhibitors; ARB, angiotensin receptor blockers; BB, beta blockers; BMI, body mass index; CCB, calcium channel blockers; CKD, chronic kidney disease; CVD, cardiovascular disease; DBP, diastolic blood pressure; eGFR, estimated glomerular filtration rate; HDL, high density lipoprotein; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; SBP, systolic blood pressure; SPRINT, systolic blood pressure intervention trial; UACR, urine albumin creatinine ratio; uEGF, urine epidermal growth factor.

per year faster decline in eGFR. The association of uEGF with annual percent change in eGFR was visualized using a spline function, revealing a directionally consistent association between lower baseline uEGF concentrations and faster declines in eGFR, although the relationship seemed to plateau somewhat at higher uEGF concentrations (Figure 3). When uEGF was examined in quartiles, the first quartile was associated with almost 2.5% faster annual eGFR decline than the highest quartile in models 1 and 2. This association attenuated and did not reach statistical significance in model 3.

During a median of 3.1 years of follow-up, 418 (17.8%) had a ≥30% decline in eGFR. Lower baseline uEGF had a borderline significant association with a higher risk of ≥30% eGFR decline in multivariable models adjusting for urine albumin and eGFR (Table 2 and Figure 2). There were 73 participants (3.1%) who developed ESKD. Each 50% lower uEGF concentration was associated with 4.14-fold (95% CI: 3.06–5.61) increased risk of ESKD in models adjusting for demographics, randomized treatment arm, and clinical characteristics; however, the association attenuated and was no longer statistically significant after additionally adjusting for baseline urine albumin and eGFR (hazard ratio: 1.24 [95% CI: 0.81–1.89]) (Table 2 and Figure 2).

There were 183 participants (7.8%) who experienced an AKI event during the follow-up period. As with the ESKD outcome, lower uEGF was strongly associated with increased risk of AKI in models 1 and 2, but not after urine albumin and eGFR adjustment (Table 2 and Figure 2).

We found no significant interactions of uEGF with the randomized treatment arm for the kidney outcomes except for ≥30% eGFR decline; associations of uEGF with ≥30% eGFR decline appeared stronger in the standard treatment arm of the trial. In model 3, each 50% lower uEGF concentration was associated with 1.23 times (95% CI: 0.93–1.64) higher risk of ≥30% eGFR decline in the standard treatment arm and 1.09 times (95% CI: 0.90–1.31) higher risk of ≥30% eGFR decline in the intensive treatment arm (*P* for interaction: 0.001) (Supplementary Table S2).

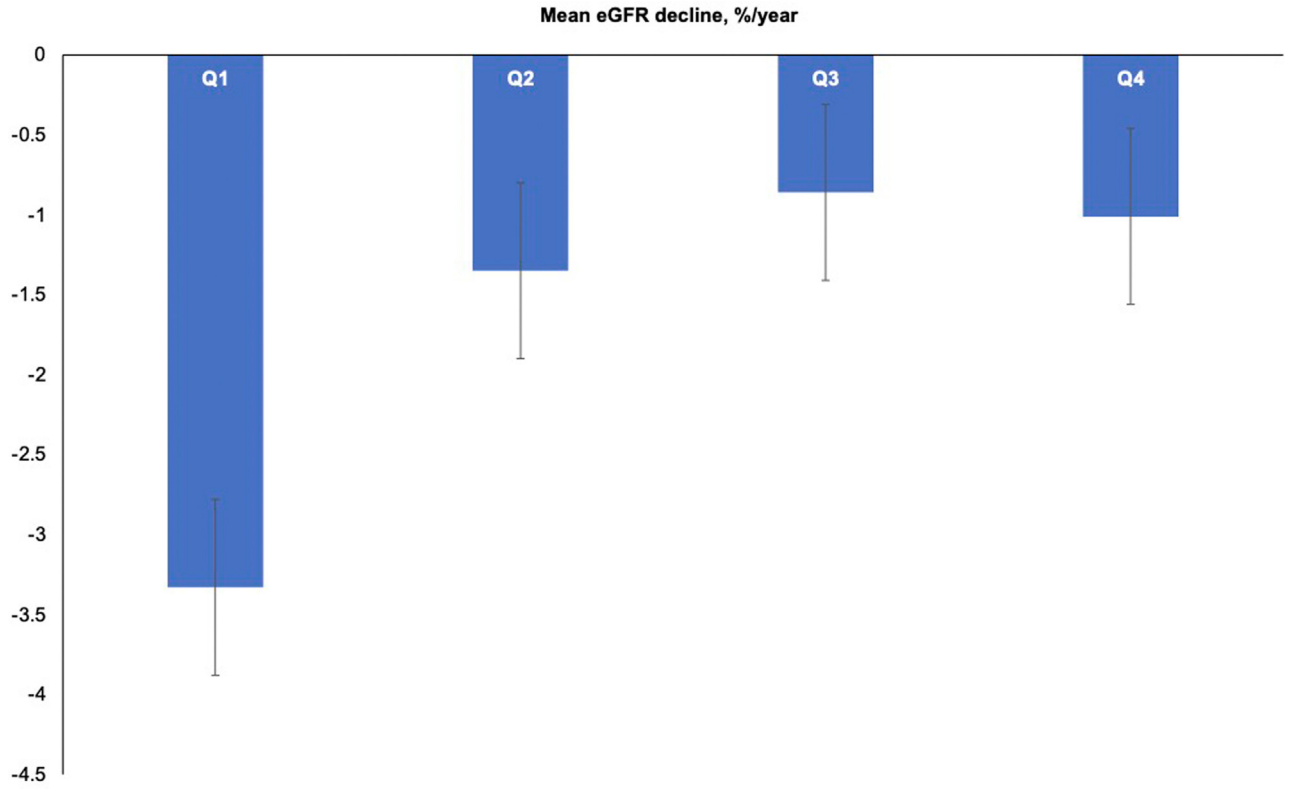

Figure 1. Unadjusted mean eGFR decline during the study period across uEGF quartiles. Mean eGFR decline was defined as percent change per year. eGFR, estimated glomerular filtration rate; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; uEGF, urine epidermal growth factor.

Table 2. Associations of uEGF concentrations with kidney outcomes among SPRINT participants with prevalent CKD

	uEGF Q1	uEGF Q2	uEGF Q3	uEGF Q4	Per 50% lower uEGF concentration
Outcome = Annualized percent change in eGFR					
Mean eGFR decline, % /yr	−3.33 (−3.88 to −2.78)	−1.35 (−1.91 to −0.80)	−0.86 (−1.41 to −0.31)	−1.01 (−1.56 to −0.47)	−1.80 (−2.42 to −1.17)
	β (95% CI)	β (95% CI)	β (95% CI)	β (95% CI)	β (95% CI)
Model 1[a]	−2.47 (−3.56 to −1.39)	−0.45 (−1.35 to 0.45)	0.17 (−0.71 to 0.94)	0 (ref)	−1.43 (−1.79 to −1.08)
Model 2[b]	−2.39 (−3.46 to −1.31)	−0.40 (−1.29 to 0.49)	0.03 (−0.79 to 0.84)	0 (ref)	−1.41 (−1.76 to −1.06)
Model 3[c]	−0.05 (−1.31 to 1.21)	0.58 (−0.36 to 1.52)	0.52 (−0.30 to 1.33)	0 (ref)	−0.74 (−1.19 to −0.29)
Outcome = ≥30% eGFR Decline					
Incident events, *n*	151	101	85	81	418
Incidence rate, %/yr	9.4	5.9	4.7	4.6	6.1
	HR (95% CI)	**HR (95% CI)**	**HR (95% CI)**	**HR (95% CI)**	**HR (95% CI)**
Model 1[a]	2.05 (1.42–2.96)	1.34 (0.96–1.88)	1.03 (0.75–1.42)	1.00 (ref)	1.43 (1.27–1.61)
Model 2[b]	1.89 (1.31–2.73)	1.27 (0.91–1.78)	1.02 (0.74–1.41)	1.00 (ref)	1.39 (1.23–1.56)
Model 3[c]	1.09 (0.70–1.69)	1.01 (0.70–1.43)	0.93 (0.68–1.29)	1.00 (ref)	1.17 (1.00–1.36)
Outcome = AKI					
Incident events, *n*	75	47	30	31	183
Incidence rate (%/yr)	4.42	2.67	1.61	1.7	2.56
	SHR (95% CI)	SHR (95% CI)	SHR (95% CI)	SHR (95% CI)	SHR (95% CI)
Model 1[a]	3.67 (2.07–6.49)	1.91 (1.12–3.25)	1.05 (0.62–1.77)	1.00 (ref)	1.66 (1.38–2.01)
Model 2[b]	3.44 (1.93–6.13)	1.83 (1.07–3.12)	1.01 (0.60–1.71)	1.00 (ref)	1.63 (1.35–1.98)
Model 3[c]	1.38 (0.68–2.80)	1.16 (0.65–2.05)	0.79 (0.46–1.37)	1.00 (ref)	1.11 (0.85–1.45)

AKI, acute kidney injury; CI, confidence interval; CKD, chronic kidney disease; eGFR, estimated glomerular filtration rate; HR, hazard ratio; *n*, number; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; SHR, subdistribution HR; SPRINT, systolic blood pressure intervention trial; uEGF, urine epidermal growth factor.
[a]Adjusted for age, sex, race, randomized treatment arm, and urine creatinine.
[b]Additionally adjusted for smoking status, body mass index, history of cardiovascular disease, systolic blood pressure, diastolic blood pressure, total cholesterol, high density lipoprotein, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta blocker, calcium channel blockers, and diuretics.
[c]Additionally adjusted for baseline urine albumin and eGFR.

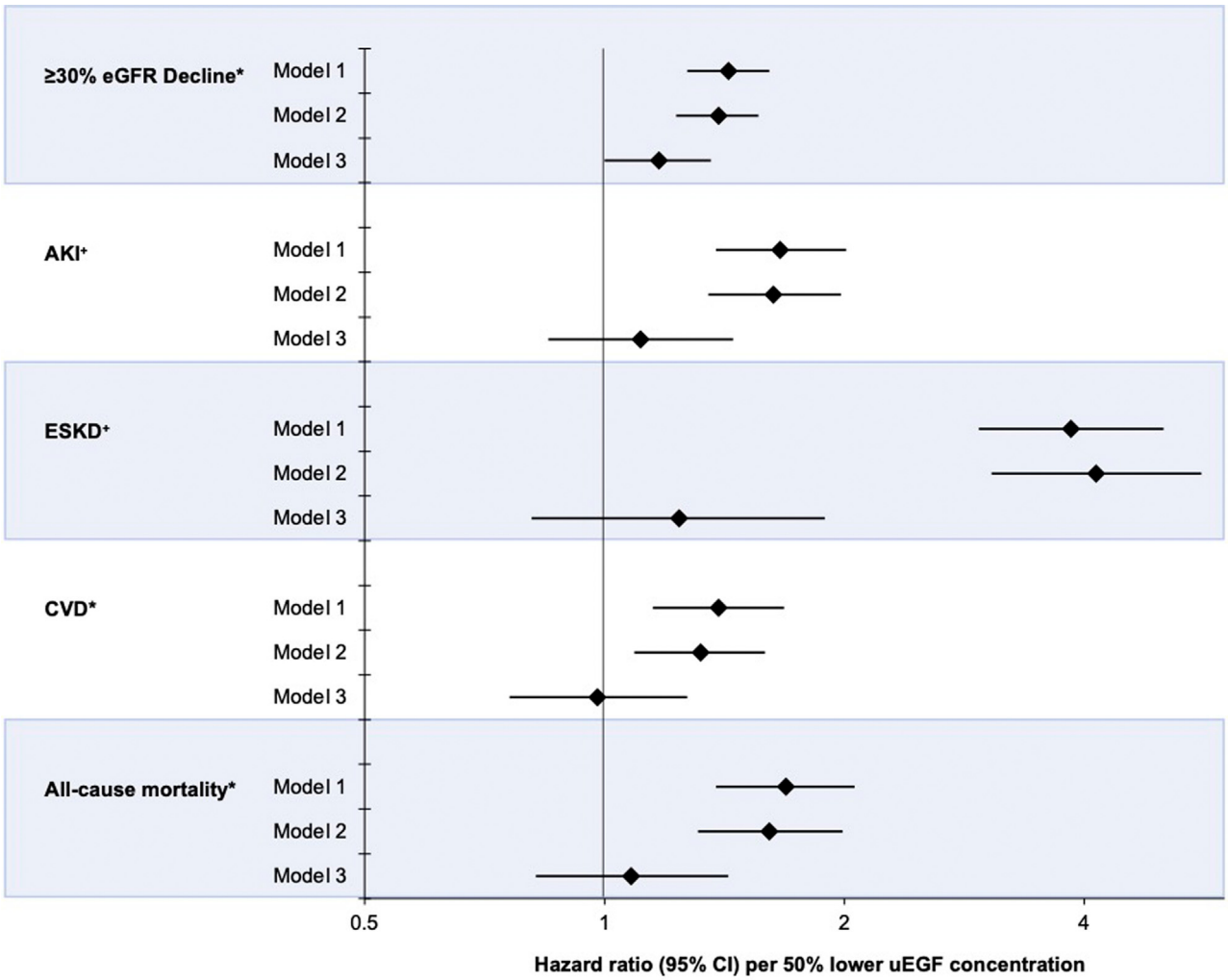

Figure 2. Forest plot showing associations between uEGF and kidney, cardiovascular, and mortality outcomes and their respective adjusted hazard ratios (95% confidence interval). Hazard ratios (per 50% lower uEGF concentration) with 95% confidence intervals were obtained from multivariable models. Model 1: adjusted for age, sex, race, randomized treatment arm, and urine creatinine. Model 2: additionally adjusted for smoking status, body mass index, history of cardiovascular disease, systolic blood pressure, diastolic blood pressure, total cholesterol, high density lipoprotein, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta blocker, calcium channel blockers, and diuretics. Model 3: additionally adjusted for baseline urine albumin and eGFR. *Calculated via Cox proportional hazards regression model. The measure of outcome is the hazard ratio. $^{+}$Calculated via Fine and Gray model to account for the competing risk of death. The measure of outcome is the sub-distribution hazard ratio. AKI, acute kidney injury; CVD, cardiovascular disease; eGFR, estimated glomerular filtration rate; ESKD, end-stage kidney disease; uEGF, urine epidermal growth factor.

Associations of uEGF With Cardiovascular Outcomes and All-Cause Mortality

The study sample experienced 180 CVD events (7.7%) and 158 deaths (6.7%) during follow-up. Each 50% lower uEGF concentration was associated with greater than 1.3-fold higher risk of developing CVD in model 2, but the association was no longer significant in model 3 (Table 3 and Figure 2).

Higher uEGF concentration was strongly and inversely associated with mortality risk in models 1 and 2; however, the association was attenuated after adjustment for baseline urine albumin and eGFR (Table 3 and Figure 2). For both the CVD and mortality outcomes, the magnitude of associations increased gradually with descending uEGF quartiles when uEGF was modeled in quartiles. The results of the regression models indexed for urine creatinine are depicted in Supplementary Table S3.

There were no statistically significant interactions by the randomized treatment arm for the associations of uEGF with incident CVD and all-cause mortality.

DISCUSSION

In this study of hypertensive persons with CKD, we found that lower uEGF concentrations at baseline were associated with faster rates of kidney function decline independent of demographics, clinical characteristics, baseline urine albumin and eGFR. Lower uEGF was also associated with higher risks of ESKD, AKI, CVD, and

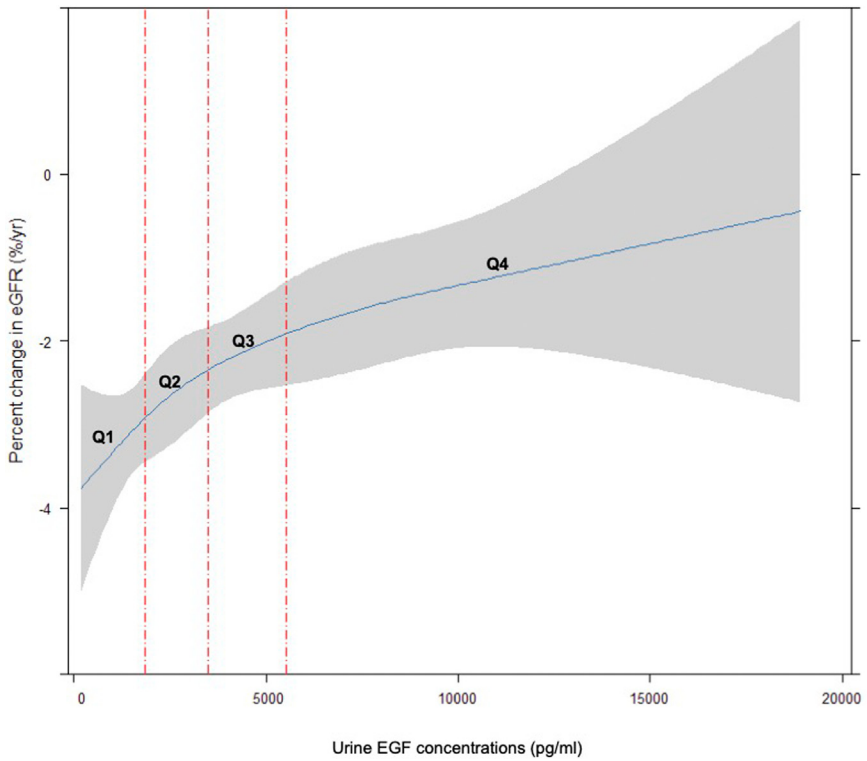

Figure 3. Relationship of uEGF and annualized eGFR change among SPRINT participants with prevalent CKD. Restricted cubic spline function demonstrating the association of uEGF with the annualized percent change in eGFR. The blue line represents the adjusted point estimates, gray shaded area represents the 95% confidence interval, and the red lines represent uEGF quartile boundaries. The spline function was adjusted for age, sex, race, randomized treatment arm, urine creatinine, smoking status, body mass index, history of cardiovascular disease, systolic blood pressure, diastolic blood pressure, total cholesterol, high density lipoprotein, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta blocker, calcium channel blockers, diuretics, urine albumin, and eGFR. CKD, chronic kidney disease; eGFR, estimated glomerular filtration rate; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; SPRINT, systolic blood pressure intervention trial; uEGF, urine epidermal growth factor.

all-cause mortality in models adjusting for demographics and clinical characteristics; however, these associations were attenuated and rendered no longer statistically significant after additional adjustment for urine albumin and eGFR.

EGF serves as a critical regulator of normal cell growth and differentiation.[24] It is primarily synthesized within the thick ascending limb of the loop of Henle and the distal convoluted tubule of the kidneys in adults.[25] Preclinical studies have shown the reparative potential of EGF in kidney diseases. In one study of rats, exogenous administration of EGF accelerated the kidney repair process after an ischemic injury.[12] Another study showed that EGF deficiency in mice was associated with the development of interstitial fibrosis and inflammation in the kidneys.[13]

Clinical studies suggest that lower uEGF levels are associated with adverse kidney outcomes.[16,26,27] In the Translational Research Investigating Biomarker Endpoints in AKI study, lower uEGF levels following cardiac surgery were subsequently associated with higher risk of incident CKD and CKD progression.[15] In the Assessment, Serial Evaluation, and Subsequent Sequela of Acute Kidney Injury study, lower uEGF levels measured after AKI episodes during hospitalization were associated with increased risk of major adverse kidney events, including CKD incidence, progression, and development of kidney failure.[28] A study investigating 2 European cohorts comprising community-living adults without preexisting CKD or diabetes mellitus showed that lower uEGF concentrations were associated with higher risk of rapid eGFR decline and incident CKD.[14] In addition, among middle-aged adults with preserved kidney function in the Coronary Artery Risk Development in Young Adults study, our group recently demonstrated that in an analysis of 7 kidney tubule biomarkers, only uEGF had strong and independent associations with subsequent eGFR decline. Further, its association with incident reduced eGFR (eGFR $<$ 60 ml/min per 1.73 m^2) persisted even after adjustment for baseline urine albumin and eGFR.[29]

Building on previous research that has shown the associations between uEGF, a marker of renal repair, and both kidney fibrosis and recovery from AKI events,[13,28,30] we examined its impact on the prognosis of CKD by evaluating uEGF associations with kidney

Table 3. Associations of uEGF concentrations with CVD and all-cause mortality among SPRINT participants with prevalent CKD

	uEGF Q1	uEGF Q2	uEGF Q3	uEGF Q4	Per 50% lower uEGF concentration
Outcome = CVD[S]					
Incident events, *n*	55	57	32	36	180
Incidence rate, %/yr	3.6	2.8	1.4	1.6	2.8
	HR (95% CI)	HR (95% CI)	HR (95% CI)	HR (95% CI)	HR (95% CI)
Model 1[a]	1.85 (1.06–3.21)	1.61 (1.01–2.58)	0.87 (0.53–1.42)	1.00 (ref)	1.39 (1.15–1.68)
Model 2[b]	1.69 (0.97–2.95)	1.60 (0.99–2.60)	0.86 (0.52–1.42)	1.00 (ref)	1.32 (1.09–1.59)
Model 3[c]	0.80 (0.40–1.60)	1.17 (0.69–1.98)	0.75 (0.45–1.24)	1.00 (ref)	0.98 (0.76–1.27)
Outcome = All-cause mortality					
Incident events, *n*	56	45	33	24	158
Incidence rate, %/yr	3	2.4	1.7	1.2	2.1
	HR (95% CI)	HR (95% CI)	HR (95% CI)	HR (95% CI)	HR (95% CI)
Model 1[a]	3.69 (2.01–6.78)	2.30 (1.32–3.99)	1.48 (0.86–2.55)	1.00 (ref)	1.69 (1.38–2.06)
Model 2[b]	3.30 (1.76–6.16)	2.10 (1.20–3.66)	1.44 (0.83–2.50)	1.00 (ref)	1.61 (1.31–1.99)
Model 3[c]	1.18 (0.56–2.47)	1.32 (0.74–2.36)	1.14 (0.66–1.99)	1.00 (ref)	1.08 (0.82–1.43)

CI, confidence interval; CKD, chronic kidney disease; CVD, cardiovascular disease; HR; hazard ratio; *n*, number; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; SPRINT, systolic blood pressure intervention trial; uEGF, urine epidermal growth factor.
[a]Adjusted for age, sex, race, randomized treatment arm, and urine creatinine.
[b]Additionally adjusted for smoking status, body mass index, history of cardiovascular disease, systolic blood pressure, diastolic blood pressure, total cholesterol, high density lipoprotein, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta blocker, calcium channel blockers, and diuretics.
[c]Additionally adjusted for baseline urine albumin and eGFR.
CVD composite outcome includes myocardial infarction, acute coronary syndrome not resulting in myocardial infarction, stroke, acute decompensated heart failure, or death from cardiovascular causes.

outcomes among community-living hypertensive persons with CKD. Similar to our findings in the Coronary Artery Risk Development in Young Adults study, we observed that lower uEGF is associated with a faster decline in eGFR. Lower uEGF was also associated with an increased risk of ≥ 30% eGFR decline overall, and we observed a significant interaction between uEGF and randomization arm for this outcome (*P* for interaction: 0.001). The association appeared stronger among standard arm participants than the treatment arm; however, these associations were not significant after adjusting the model for urine albumin and eGFR. Our findings of interaction may be explained by the effects of blood pressure lowering on uEGF and kidney function decline. In SPRINT, we have previously found that randomization to intensive blood pressure lowering may influence associations of kidney tubule biomarkers with kidney function changes. Jotwani *et al*. found that urine uromodulin and β2-microglobulin were significantly associated with annual percent eGFR decline in the standard arm, whereas the associations were weaker and did not reach statistical significance in the treatment arm. Nonetheless, our finding of effect modification by treatment arm requires cautious interpretation and replication, because true biological interactions are uncommon.[2]

In addition, lower uEGF was associated with increased risk for AKI and ESKD, but these associations attenuated after accounting for baseline urine albumin and eGFR. Our findings in the prior Coronary Artery Risk Development in Young Adults study and this current SPRINT analysis suggest that the correlation between uEGF and eGFR becomes stronger with more advanced kidney disease, such that uEGF provides less incremental information about kidney health. Therefore, it may offer greater diagnostic ability in the early stages of kidney dysfunction.

To our knowledge, this is the first study investigating the associations of uEGF with incident CVD and mortality. Jotwani *et al*. demonstrated that urine biomarkers of kidney tubule damage in SPRINT participants with CKD are independently associated with mortality but not with CVD risk.[31] In another study among SPRINT participants with CKD, higher baseline urine alpha-1 microglobulin and lower uromodulin levels were associated with an increased risk of CVD, whereas a higher baseline alpha-1 microglobulin level was independently associated with an increased risk of mortality in multivariable analysis including eGFR and albuminuria.[32] Although our study did not reveal a statistically significant association with either of the outcomes after adjusting for baseline urine albumin and eGFR, and though both end points were relatively well powered, further studies are required to corroborate our results and advance our knowledge in this field.

Our study had several strengths, including a diverse population from a large multicenter trial, protocol-based eGFR assessments during follow-up, rigorously adjudicated CVD events, and limited loss to follow-up.

The study has important limitations. First, uEGF measurements were only conducted at baseline, precluding the assessment of longitudinal uEGF changes. Second, due to the design of SPRINT, the findings may not be generalizable to persons with CKD who have diabetes mellitus, severe proteinuria, polycystic kidney disease, heart failure, or prior stroke. Finally, there was a low incidence of ESKD due to the relatively short follow-up duration and few SPRINT participants with advanced CKD at baseline.

In summary, among hypertensive individuals with CKD, lower uEGF is independently associated with a faster decline of eGFR and a greater likelihood of ≥30% eGFR decline. Although lower uEGF concentrations appeared strongly associated with AKI, ESKD, CVD, and all-cause mortality in initial models, these associations were attenuated and rendered no longer statistically significant after adjustment for baseline urine albumin and eGFR. Further studies will be needed to determine the associations of longitudinal uEGF trajectories with these clinical outcomes and whether CKD treatments influence uEGF levels as a potential biological intermediary of CKD.

DISCLOSURE

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AUTHOR CONTRIBUTIONS

MP, RK, JHI, and MGS were primarily responsible for study conception and design. RK carried out statistical analysis. MP, RK, JHI, and MGS were primarily responsible for interpretation of the data. MP, JHI, and MGS drafted the article. All listed authors contributed to revisions and provided intellectual content of critical importance to the work and approved the final version of the manuscript to be submitted for publication.

SUPPLEMENTARY MATERIAL

Supplementary File (PDF)

Table S1. Associations of uEGF/UCr concentrations with kidney outcomes among SPRINT participants with prevalent CKD.

Table S2. Associations of uEGF concentrations with 30% kidney function decline among SPRINT participants with prevalent CKD, stratified by randomized treatment arm.

Table S3. Associations of uEGF/UCr concentrations with CVD and all-cause mortality among SPRINT participants with prevalent CKD.

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