Association of Disorders in Mineral Metabolism with Progression of Chronic Kidney Disease

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Abnormalities of mineral metabolism are associated with increased mortality in patients with ESRD, but their effects in predialysis chronic kidney disease (CKD) are less well characterized. In this study, the associations between levels of serum phosphorus, calcium, and calcium-phosphorus product and progression of CKD were examined. Historical data were collected on 985 male US veterans (age 67.4 ± 10.9; 23.9% black) with CKD stages 1 through 5. Unadjusted and multivariable-adjusted relative risks for progressive CKD (defined as the composite of ESRD or doubling of serum creatinine) were calculated for categories of serum phosphorus, calcium, and calcium-phosphorus product using Cox proportional hazards models. Higher phosphorus was associated with a higher risk for the composite end point (adjusted hazard ratio [HR] [95% confidence interval (CI)] for phosphorus levels 3.3 to 3.8, 3.81 to 4.3, and >4.3 *versus* <3.3 mg/dl 0.83 [0.54 to 1.27], 1.24 [0.82 to 1.88], and 1.60 [1.06 to 2.41]; $P = 0.001$ for trend). A 1-mg/dl higher phosphorus level was associated with an adjusted HR (95% CI) of 1.29 (1.12 to 1.48; $P < 0.001$). Higher calcium-phosphorus product also was associated with higher risk for progressive CKD (adjusted HR [95% CI] for calcium-phosphorus products 30 to 35, 36 to 40, and >40 *versus* <30 mg^2/dl^2 0.58 [0.36 to 0.94], 0.87 [0.57 to 1.34], and 1.37 [0.91 to 2.07]; $P = 0.002$ for trend). A 10-mg^2/dl^2 higher calcium-phosphorus product was associated with an adjusted HR (95% CI) of 1.29 (1.11 to 1.51; $P = 0.001$). Lower serum calcium showed a trend toward higher risk for progressive CKD but without statistical significance. Higher serum phosphorus and higher calcium-phosphorus product are associated with progression of CKD.

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Abnormalities of bone and mineral metabolism are associated with higher mortality in patients who have ESRD and are on dialysis (1). Whereas abnormalities in calcium and phosphorus metabolism are present already in patients who have chronic kidney disease (CKD) and are not yet on dialysis (2), their impact on outcomes in this patient population is less well described. Dietary protein restriction has been associated with slower progression of CKD (3). This benefit has been attributed in part to the dietary phosphorus restriction that occurs as a result of the lower protein intake (4–8), but the underlying mechanisms of action still are not fully understood. It also is unclear whether actual serum phosphorus levels and other markers of disordered bone and mineral metabolism (*e.g.*, serum calcium, calcium-phosphorus product) are associated with progression of kidney disease in patients with CKD. We examined the association of baseline levels of serum phosphorus, calcium, and calcium-phosphorus product with renal functional outcomes in a well-characterized cohort of US veterans who had CKD stages 1 through 5 and were not yet on dialysis.

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Materials and Methods

Study Population and Outcomes

We collected data in a historical prospective cohort of patients who were referred to a single outpatient nephrology clinic between January 1, 1990, and June 30, 2005, at Salem Veterans Affairs Medical Center (VAMC). After exclusion of patients with a kidney transplant, patients who were on renal replacement therapy (RRT), and patients referred for problems other than CKD, 1012 patients with CKD stages 1 through 5 were identified. Of these, 16 (1.6%) patients had no serum phosphorus measurement available and were excluded from further analyses. Because there were only 11 female patients in the cohort, they also were excluded from further analyses. The final analysis included 985 patients.

Patients were followed until death or until August 31, 2005, with the recording of death from all causes, the initiation of RRT, and the doubling of baseline serum creatinine level. Patients were categorized as lost to follow-up when they had no contact with the medical center for >6 mo. Deaths were recorded from the VA computerized patient record system and cross-checked with death certificate–based data that were obtained from the National Death Index. RRT, defined as initiation of hemodialysis or peritoneal dialysis, was identified from medical records at Salem VAMC, including Medicare Form 2728.

Data Collection

Baseline data were collected from paper charts and electronic medical records at Salem VAMC and included demographic and anthropometric information, comorbidities, smoking status, baseline BP measurement, and laboratory measures. All of the laboratory studies were

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performed in a single clinical laboratory at Salem VAMC. The baseline use of phosphate binders, angiotensin-converting enzyme inhibitors (ACEI), and angiotensin II receptor blockers (ARB) was collected from computerized patient records and paper charts. Review of phosphate binder medications revealed that only two (0.2%) patients were using sevelamer hydrochloride at baseline; therefore, only data on calcium-containing binders was included in analyses. Diabetes was defined as the presence of a fasting glucose level >126 mg/dl or antidiabetic therapy. Atherosclerotic cardiovascular disease was defined as a history of cardiovascular or peripheral vascular disease. Serum creatinine values were recorded longitudinally until a major end point (death or dialysis) ensued, with exclusion of values that were measured during episodes of acute renal failure (defined as an increase of 20% from baseline over a period of 24 h). GFR was estimated from baseline creatinine levels using the abbreviated equation developed for the Modification of Diet in Renal Disease Study (9), and patients were categorized according to the Kidney Disease Outcomes Quality Initiative Clinical Practice Guidelines for Chronic Kidney Disease: Evaluation, Classification, and Stratification (2).

Statistical Analyses

Continuous variables were expressed as mean ± SD or geometric mean (95% confidence interval [CI]), and categorical variables were expressed as proportions. Continuous variables with skewed distribution (estimated GFR [eGFR], blood cholesterol, and 24-h urine protein) were log-transformed. Missing data points for serum albumin (1.4% missing), blood cholesterol (1.2% missing), and 24-h urine protein (2.9% missing) were imputed using linear regression with all other patient characteristics serving as independent variables. To address missing data points for smoking status (6.1% missing), we analyzed this variable by adding a third, "missing" category (active smokers, nonsmokers, and "missing smoking status"). Serum calcium concentration was corrected for serum albumin concentration using the following formula: Corrected calcium = measured calcium + 0.8(4 − serum albumin) (10).

Survival Modeling

The starting time for survival analysis was the date of the initial encounter, and the outcome measure was the composite of ESRD (defined as initiation of RRT) or doubling of baseline serum creatinine level. Patients who died were censored at the date of death, and patients who were lost to follow-up were censored at the date of the last documented contact with the medical center. Event rates were calculated using the person-years approach. We analyzed the unadjusted and multivariable-adjusted association of baseline serum phosphorus, serum calcium, and calcium-phosphorus product with the main outcome measure in Cox proportional hazards models. Multivariable models were *a priori* adjusted for all variables with which baseline phosphorus or calcium levels correlated and also for variables that were expected to be associated independently with progression of CKD. Nonlinear associations were explored by inclusion of quadratic terms. Effect modification was explored by performing subgroup analyses and by inclusion of interaction terms. The proportionality assumption was tested using plots of log (−log [survival rate]) against log (survival time) and by comparing predicted with actual survival curves. All analyses were repeated after the exclusion of patients with a baseline eGFR of <15 ml/min per 1.73 m^2. $P < 0.05$ was considered significant. Statistical analyses were performed using STATA statistical software version 8 (STATA Corp., College Station, TX). The study protocol was approved by the Research and Development Committee at the Salem VAMC.

Results

Baseline characteristics are presented in Table 1 for patient groups divided by quartiles of serum phosphorus and in Table 2 for patient groups divided by quartiles of serum calcium. Patients with serum phosphorus levels >4.3 mg/dl were younger; more frequently had diabetes and were active smokers; and had lower eGFR, hemoglobin, albumin, calcium, and bicarbonate and higher proteinuria. Patients with serum calcium levels <9.1 mg/dl were more likely to be white and had higher levels of systolic BP, serum albumin, blood urea nitrogen (BUN), and proteinuria and lower levels of GFR, hemoglobin, and bicarbonate. The baseline characteristics of the 24 patients who were lost to follow-up (2.4% of total) were not significantly different (data not shown). A total of 258 patients reached the composite end point of ESRD or doubling of serum creatinine (210 patients reached ESRD, and 48 patients had a doubling of serum creatinine), for an overall incidence rate (95% CI) of 88.6/1000 patient-years (78.4 to 100.1). Table 3 shows the number (%) of outcomes overall and by subgroups of serum phosphorus, calcium, and calcium-phosphorus product, indicating more events in the subgroups with higher phosphorus, lower calcium, and higher calcium-phosphorus product. Median duration of follow-up was 2.1 yr, and total time at risk was 2911 patient-years.

Serum Phosphorus

Figure 1 shows the hazard ratio (HR) of the composite end point, by quartiles of serum phosphorus, in the unadjusted Cox model and after adjustment for age, race, systolic BP, diastolic BP, diabetes, smoking status, eGFR, serum albumin, calcium, bicarbonate, BUN, hemoglobin, proteinuria, and use of calcium-containing phosphate binders and ACEI/ARB. A baseline serum phosphorus level of >4.3 mg/dl was associated with the highest HR for the composite end point in both the unadjusted and the adjusted models (adjusted HR [95% CI] for serum phosphorus levels 3.3 to 3.8, 3.81 to 4.3, and >4.3 *versus* <3.3 mg/dl 0.83 [0.54 to 1.27], 1.24 [0.82 to 1.88], and 1.60 [1.06 to 2.41]; $P = 0.001$ for trend). The group with serum phosphorus of 3.3 to 3.8 mg/dl had the lowest risk for progression, but inclusion of a quadratic term for serum phosphorus did not confirm a significant nonlinear association ($P = 0.3$ for quadratic term). When serum phosphorus was included as a continuous variable, a 1-mg/dl higher serum phosphorus level was associated with an adjusted HR (95% CI) of 1.29 (1.12 to 1.48; $P < 0.001$). The association between serum phosphorus level and progression of CKD was stronger in patients with diabetes (adjusted HR [95% CI] associated with a 1-mg/dl higher phosphorus level 1.47 [1.17 to 1.84] in those with diabetes *versus* 1.06 [0.85 to 1.33] in those without diabetes), but the interaction was not statistically significant ($P = 0.1$ for the interaction term). A quantitative interaction was present between serum phosphorus and serum calcium, with a higher HR in the subgroup with elevated calcium level (adjusted HR [95% CI] associated with a 1-mg/dl higher phosphorus level 1.18 [0.98 to 1.41] in patients with serum calcium <9.2 mg/dl *versus* 1.48 [1.12 to 1.96] in patients with serum calcium ≥9.2 mg/dl; $P = 0.001$ for the interaction term). There was no significant

Table 1. Baseline characteristics of individuals stratified by quartiles of serum phosphorus level[a]

	Serum Phosphorus (mg/dl)				*P*
	<3.3	3.3 to 3.8	3.81 to 4.3	>4.3	
n (%)	255 (25.9)	287 (29.1)	223 (22.6)	220 (22.3)	
Age (yr)	68.0 ± 10.4	68.6 ± 10.7	67.9 ± 11.2	64.5 ± 11.1	0.0001
Black race	63 (24.7)	63 (21.9)	54 (24.2)	55 (25.0)	0.8
Diabetes	118 (46.3)	157 (54.7)	121 (54.3)	142 (64.6)	0.001
ASCVD	151 (59.2)	170 (59.2)	133 (59.6)	136 (61.8)	0.9
Active smokers	54 (22.7)	69 (25.1)	58 (28.3)	77 (37.4)	0.004
SBP (mmHg)	150.9 ± 26.0	152.5 ± 25.1	150.8 ± 29.2	154.3 ± 26.5	0.4
DBP (mmHg)	75.7 ± 15.8	76.1 ± 15.1	74.8 ± 15.8	76.1 ± 16.1	0.8
Use of calcium	16 (6.3)	11 (3.8)	8 (3.6)	21 (9.5)	0.019
Use of ACEI/ARB	152 (59.6)	165 (60.0)	132 (59.2)	131 (59.5)	0.9
GFR (ml/min per 1.73 m^2)	39.0 (37.3 to 41.0)	36.8 (35.1 to 38.5)	32.3 (30.9 to 34.7)	25.1 (23.4 to 26.9)	<0.0001
CKD stages					
1	7 (2.7)	5 (1.7)	5 (2.2)	1 (0.4)	<0.001
2	27 (10.6)	25 (8.7)	12 (5.4)	5 (2.3)	
3	168 (65.9)	184 (64.1)	112 (50.2)	76 (34.5)	
4	48 (18.8)	67 (23.3)	90 (40.4)	106 (85.4)	
5	5 (1.9)	6 (2.1)	4 (1.8)	32 (14.5)	
BUN (mg/dl)	25.9 ± 10.3	30.8 ± 12.7	34.3 ± 14.2	43.1 ± 19.7	<0.0001
Hemoglobin (g/dl)	13.1 ± 1.8	13.0 ± 1.8	12.4 ± 1.9	11.8 ± 1.9	<0.0001
Cholesterol (mg/dl)	180.7 (175.1 to 186.4)	185.4 (179.4 to 191.7)	189.2 (182.7 to 196.0)	187.7 (180.7 to 194.9)	0.26
Albumin (g/dl)	3.7 ± 0.4	3.7 ± 0.5	3.6 ± 0.5	3.5 ± 0.5	<0.0001
Calcium (mg/dl)	9.2 ± 0.5	9.2 ± 0.5	9.1 ± 0.6	9.0 ± 0.7	<0.0001
Bicarbonate (mmol/L)	25.9 ± 3.2	25.7 ± 3.4	25.4 ± 3.4	24.6 ± 4.1	0.0002
Urinary protein (mg/24 h)	521.4 (435.8 to 623.7)	618.3 (524.4 to 729.0)	770.4 (628.1 to 945.0)	1806.3 (1497.4 to 2178.9)	<0.0001

[a]Data are means ± SD, *n* (% of total), or geometric mean (95% CI). ACEI, angiotensin-converting enzyme inhibitor; ARB, angiotensin II receptor blocker; ASCVD, atherosclerotic cardiovascular disease; BUN, blood urea nitrogen; CI, confidence interval; CKD, chronic kidney disease; DBP, diastolic BP; SBP, systolic BP. Comparisons were made by ANOVA, Fisher exact test, or χ^2 test.

Table 2. Baseline characteristics of individuals stratified by quartiles of serum calcium level[a]

	Serum Calcium (mg/dl)				*P*
	<9.1	9.1 to 9.4	9.41 to 9.7	>9.7	
n (%)	235 (23.8)	218 (22.1)	252 (25.5)	280 (28.4)	
Age (yr)	68.5 ± 10.6	68.0 ± 10.9	66.7 ± 10.3	66.3 ± 11.5	0.07
Black race	45 (19.1)	52 (23.8)	56 (22.2)	89 (29.2)	0.05
Diabetes	116 (49.3)	119 (54.5)	145 (57.5)	158 (56.4)	0.27
ASCVD	147 (62.5)	131 (60.0)	138 (54.7)	174 (62.1)	0.25
Active smokers	59 (27.0)	45 (22.3)	78 (32.3)	76 (28.7)	0.13
SBP (mmHg)	154.3 ± 25.3	152.4 ± 24.2	153.8 ± 28.4	148.3 ± 27.5	0.03
DBP (mmHg)	75.6 ± 15.7	76.1 ± 14.8	76.5 ± 15.2	74.6 ± 16.5	0.5
Use of calcium	16 (6.8)	15 (6.9)	11 (4.4)	14 (5.0)	0.5
Use of ACEI/ARB	132 (56.2)	135(61.9)	156 (61.9)	167 (59.6)	0.5
GFR (ml/min per 1.73 m^2)	29.2 (27.4 to 31.2)	33.2 (31.4 to 35.1)	35.5 (33.6 to 37.4)	35.4 (33.5 to 37.5)	<0.0001
CKD stage					
1	3 (1.3)	2 (0.9)	7 (2.8)	6 (2.1)	0.014
2	15 (6.4)	12 (5.5)	16 (6.3)	26 (9.3)	
3	107 (45.5)	126 (57.8)	144 (57.1)	163 (58.2)	
4	92 (39.1)	70 (32.1)	78 (30.9)	71 (25.4)	
5	18 (7.7)	8 (3.7)	7 (2.8)	14 (5.0)	
BUN (mg/dl)	35.7 ± 16.1	32.7 ± 15.3	30.7 ± 13.1	33.3 ± 17.1	0.006
Hemoglobin (g/dl)	12.2 ± 1.9	12.4 ± 1.8	12.8 ± 1.7	12.8 ± 2.0	0.0009
Cholesterol (mg/dl)	184.4 (178.7 to 190.3)	183.2 (176.5 to 190.1)	185.5 (179.6 to 191.7)	188.2 (181.7 to 194.9)	0.7
Albumin (g/dl)	3.8 ± 0.4	3.6 ± 0.4	3.5 ± 0.5	3.4 ± 0.5	<0.0001
Phosphorus (mg/dl)	3.9 ± 0.9	3.8 ± 0.7	3.7 ± 0.7	3.9 ± 0.8	0.07
Bicarbonate (mmol/L)	24.5 ± 3.9	25.4 ± 3.1	25.8 ± 3.2	25.9 ± 3.5	0.0001
Urinary protein (mg/24 h)	905.0 (743.4 to 1101.8)	614.8 (507.9 to 744.2)	857.9 (702.6 to 1048.0)	799.8 (670.1 to 954.6)	0.03

[a]Data are means ± SD, *n* (% of total), or geometric mean (95% CI). Comparisons were made by ANOVA, Fisher exact test, or χ^2 test.

Table 3. Distribution of events, by quartiles of serum phosphorus, calcium, and calcium-phosphorus product[a]

	Composite End Point	ESRD	Doubling of Serum Creatinine
All	258 (26.1)	210 (21.3)	48 (4.8)
PO_4 (mg/dl)			
<3.3	40 (15.6)	31 (12.1)	9 (3.5)
3.3–3.8	54 (18.8)	40 (13.9)	14 (4.8)
3.81 to 4.3	60 (26.9)	50 (22.4)	10 (4.4)
>4.3	104 (47.2)	89 (40.4)	15 (6.8)
Ca (mg/dl)			
<9.1	82 (34.8)	71 (30.2)	11 (4.6)
9.1 to 9.4	51 (23.3)	40 (18.3)	11 (5.0)
9.41 to 9.7	62 (24.6)	51 (20.2)	11 (4.3)
>9.7	63 (22.5)	48 (17.1)	15 (5.3)
Ca × PO_4 (mg^2/dl^2)			
<30	35 (18.0)	30 (15.4)	5 (2.5)
30 to 35	39 (15.6)	28 (11.2)	11 (4.4)
36 to 40	68 (25.0)	53 (19.4)	15 (5.5)
>40	116 (42.9)	99 (36.6)	17 (6.3)

[a]Data are *n* (% of total).

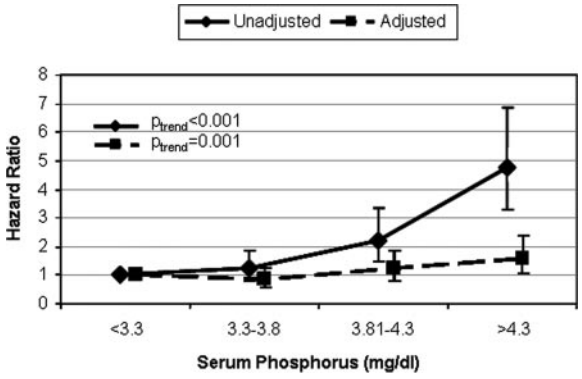

Figure 1. Hazard ratio (HR) (95% confidence interval [CI]) of the composite outcome of ESRD and doubling of serum creatinine associated with various quartiles of serum phosphorus, unadjusted and after adjustment for age, race, systolic (SBP) and diastolic BP (DBP), diabetes, smoking status, estimated GFR (eGFR), serum albumin, calcium, bicarbonate, blood urea nitrogen (BUN), hemoglobin, 24-h urine protein, and use of calcium-containing phosphate binders and angiotensin-converting enzyme inhibitors (ACEI)/angiotensin II receptor blockers (ARB). The group with serum phosphorus <3.3 mg/dl served as reference.

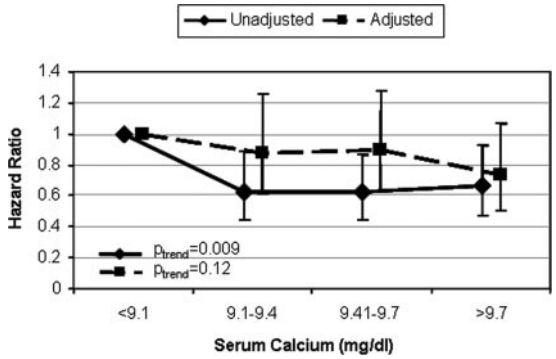

Figure 2. HR (95% CI) of the composite outcome of ESRD and doubling of serum creatinine associated with various quartiles of serum calcium, unadjusted and after adjustment for age, race, SBP and DBP, diabetes, smoking status, eGFR, serum albumin, phosphorus, bicarbonate, BUN, hemoglobin, 24-h urine protein, and use of calcium-containing phosphate binders and ACEI/ARB. The group with serum calcium <9.1 mg/dl served as reference.

interaction with age, race, or level of GFR. Results were not significantly different after the exclusion of patients with a baseline eGFR of <15 ml/min per 1.73 m^2 (adjusted HR [95% CI] for serum phosphorus level >4.3 mg/dl *versus* <3.3 mg/dl 1.74 [1.14 to 2.64]; $P = 0.01$).

Serum Calcium

Figure 2 shows the hazard of the composite outcome associated with various levels of serum calcium level in the unadjusted Cox model and after adjustment for age, race, systolic BP, diastolic BP, diabetes, smoking status, eGFR, serum albumin, calcium, bicarbonate, BUN, hemoglobin, proteinuria, and use of calcium-containing phosphate binders and ACEI/ARB. Higher serum calcium levels showed a trend toward lower risk for progression (adjusted HR [95% CI] for serum calcium levels of 9.1 to 9.4, 9.41 to 9.7, and >9.7 *versus* <9.1 mg/dl 0.88 [0.61 to 1.25], 0.89 [0.62 to 1.28], and 0.73 [0.51 to 1.06]; $P = 0.12$ for trend) but without statistical significance in the adjusted model. When serum calcium was included as a continuous variable, a 1-mg/dl higher serum calcium level was associated with an adjusted HR (95% CI) of 0.80 (0.63 to 1.02; $P = 0.07$). No significant interactions were detected. The baseline use of calcium-containing phosphate binders showed no significant as-

sociation with progression of CKD (adjusted HR [95% CI] for users *versus* nonusers of calcium-containing phosphate binders 0.77 [0.49 to 1.21]; $P = 0.26$).

Calcium-Phosphorus Product

Figure 3 shows the unadjusted and multivariable adjusted HR for the composite outcome by quartiles of the calcium-phosphorus product. A calcium-phosphorus product of >40 mg^2/dl^2 was associated with the highest HR for the composite outcome (adjusted HR [95% CI] for serum calcium-phosphorus product of 30 to 35, 36 to 40, and >40 *versus* <30 mg^2/dl^2 0.58 [0.36 to 0.94], 0.87 [0.57 to 1.35], and 1.37 [0.91 to 2.07]; $P = 0.002$ for trend). The group with a calcium-phosphorus product of 30 to 35 mg^2/dl^2 had the lowest risk for progression, but inclusion of a quadratic term did not confirm a significant nonlinear association ($P = 0.8$ for quadratic term). In a model with calcium-phosphorus product included as a continuous variable, a 10-mg^2/dl^2 higher level was associated with an adjusted HR (95% CI) of 1.29 (1.10 to 1.51; $P = 0.001$).

Discussion

Abnormalities in bone and mineral metabolism are associated with higher mortality in patients with ESRD (1). Even though these abnormalities start developing in earlier stages of CKD (2), their impact on major outcomes in this latter patient population is less well described. The impact of phosphorus level on mortality in patients who have CKD and are not yet on dialysis has been examined, with two studies showing different results (11,12), but the effect of bone-mineral abnormalities on kidney function is unclear. We examined the association of serum phosphorus, calcium, and calcium-phosphorus product levels with the composite outcome of ESRD or doubling of serum creatinine to characterize their impact on progression of CKD.

Higher serum phosphorus was associated with significantly higher risk for progression of CKD, even after adjustment for multiple potential confounders. This finding complements earlier studies that showed a beneficial effect of dietary phosphorus restriction on progression of CKD in experimental animals (5,6,8) and in humans (4,7). The mechanism underlying this association is unclear but may be related to increased nephrocalcinosis, hyperparathyroidism, alterations in cellular energy metabolism, or altered renal hemodynamics (6,13). Our other findings of a higher calcium-phosphorus product being associated with more progressive CKD and of a significant quantitative interaction between serum phosphorus and calcium levels (the association of higher serum phosphorus with progressive CKD was more accentuated in patients with higher serum calcium levels) supports the hypothesis that tissue calcification (nephrocalcinosis) may be the reason behind the observed associations. This process, once thought to be a passive one dependent on secondary hyperparathyroidism and an elevated calcium-phosphate product, seems now to involve active steps at the cellular and subcellular levels (14–18), with hyperphosphatemia shown to be associated with increased expression of osteoblast-specific proteins in vascular smooth muscle cells, a process that depends directly on the phosphate co-transporter Pit-1 (16). The effect of ambient serum calcium concentration on phosphorus-mediated vascular smooth muscle calcification was explored by Reynolds *et al.* (19), who showed that higher ambient serum calcium level led to more significant phosphorus-driven calcification *in vitro*. A similar mechanism could explain why we observed a more pronounced risk for progressive CKD associated with higher phosphorus levels in patients with higher serum calcium level. The association of serum phosphorus level with progression of CKD in our study also seemed to be more pronounced in patients with diabetes, although the interaction did not reach statistical significance. Diabetes affected the association of serum phosphorus with mortality in patients with ESRD in the study by Block *et al.* (1) (with the relative risk for mortality being higher in those with diabetes), and serum phosphorus was not associated with mortality in a study that was performed in patients who had CKD and were not yet on dialysis, who almost exclusively did not have diabetes (12). The mechanism of action behind the observed effect modification rendered by diabetes is unclear and warrants further research.

We also found an association between lower calcium levels and progressive CKD in unadjusted analyses, but the association was NS after adjustments, especially after adjustment for eGFR, suggesting that lower serum calcium level may be a surrogate marker of lower GFR rather than an independent risk factor for progression of CKD. Besides being associated with lower eGFR, low calcium may be associated with higher parathyroid hormone levels (20), which were not available to us for analyses; therefore, additional residual confounding is possible. The baseline use of calcium-containing phosphate binders also was not associated with progressive CKD, even though the use of such medications is associated with cardiovascular calcification in patients with ESRD (21,22). In a rat model of CKD, the administration of calcium-containing phosphate binders resulted in nephrocalcinosis that was comparable to control animals and significantly more severe compared with a group that was treated with sevelamer hydrochloride (23). Given the very

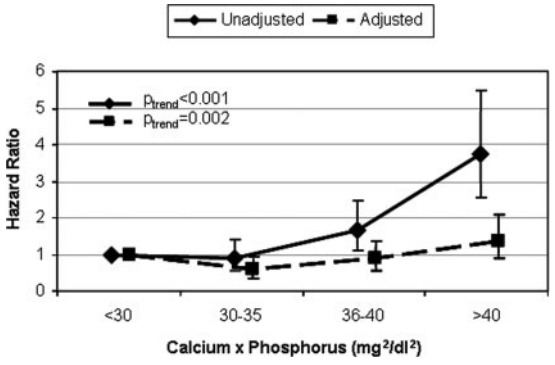

Figure 3. HR (95% CI) of the composite outcome of ESRD and doubling of serum creatinine associated with various quartiles of calcium-phosphorus product, unadjusted and after adjustment for age, race, SBP and DBP, diabetes, smoking status, eGFR, serum albumin, bicarbonate, BUN, hemoglobin, 24-h urine protein, and use of calcium-containing phosphate binders and ACEI/ARB. The group with calcium-phosphorus product <30 mg^2/dl^2 served as reference.

small proportion of patients who were using these medications in our study (56 patients [5.7%] using calcium-containing medications and two [0.2%] patients using sevelamer hydrochloride) and the possibility for the introduction of such medications during the time between our baseline assessment and the main outcome measure, we are unable to draw any valid conclusions regarding the impact of these therapies on progression of CKD.

Several shortcomings of our study have to be acknowledged. Our study population consisted exclusively of male US veterans who were drawn from a limited geographic area; therefore, our results may not apply to women or to patients from other geographic locations. Given the retrospective nature of our study, we can determine only the presence of associations but cannot establish causality. For the same reason, although we tried to correct for the effect of the major confounding variables that are known to affect progression of CKD, the effect of residual confounding cannot be ruled out. The impact of three potentially important variables has to be stressed in this regard. First, we could not measure the amount of protein intake in our patients. Protein restriction is associated with more favorable renal outcomes, and higher phosphorus levels may be surrogate markers for higher protein intake in our patient population. We used proxy markers of protein intake (eGFR-adjusted BUN and serum bicarbonate levels) to address this issue. Furthermore, the renoprotective mechanism of protein restriction is mediated at least in part by phosphorus restriction (8) and consequent lower serum phosphorus levels; therefore, we argue that serum phosphorus level could be regarded as an independent risk factor rather than a mere surrogate marker of protein intake, especially given the plausible pathophysiologic mechanisms underlying the observed associations. Second, we could not account for the confounding effect of parathyroid hormone (PTH) levels, which were not available to us. Higher PTH levels could be associated with both lower calcium and higher phosphorus levels; therefore, we cannot rule out a residual confounding effect related to hyperparathyroidism. Third, although we adjusted for eGFR, true assessment of GFR in a historical study is not possible; therefore, the association between serum phosphorus and progressive CKD may have been affected by residual confounding stemming from differences between eGFR and true GFR. The association of serum phosphorus with progressive CKD, however, showed no interaction with the level of eGFR (the HR were similar in patients with higher and lower eGFR), making it more likely to be independent from it. Other limitations are conferred by the use of baseline data to predict future outcomes and the nonconcurrent historical cohort design. By using baseline data, we cannot account for the longitudinal changes of the variables studied and of the therapeutic interventions applied (phosphate binders and ACEI/ARB). The nonconcurrent enrollment makes it even more difficult to account for the effect of therapeutic interventions, because standards of therapy might have changed during the study period (especially true for ACEI/ARB). Exclusion of calcium-containing binders and ACEI/ARB from our analyses, however did, not significantly alter the studied outcomes (data not shown).

Conclusion

Our study shows an association between higher levels of serum phosphorus and calcium-phosphorus product with an unfavorable renal outcome in male US veterans who had stages 1 through 5 CKD and were not yet on dialysis. This association was present even after adjustment for several variables that are known to affect progression of CKD. Although such an association seems plausible on the basis of animal studies and *in vitro* data, future clinical trials will have to be conducted to clarify the role of secondary hyperparathyroidism and to assess whether this association is causal and whether therapeutic interventions that target abnormalities in bone and mineral metabolism would be helpful in retarding the progression of CKD.

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