Check for updates

Racial Variations in the Markers of Mineral Bone Disorders in CKD Patients in South Africa

Bala Waziri[1], Raquel Duarte[1], Caroline Dickens[1], Therese Dix Peek[1], Jaya George[2], Vakhtang Rekhviashvili[3], Graham Paget[1] and Saraladevi Naicker[1]

[1]Department of Internal Medicine, Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, South Africa; [2]Department of Chemical Pathology, National Health Laboratory Services, University of the Witwatersrand, Johannesburg, South Africa; and [3]Renal Unit, Donald Gordon Medical Center, University of the Witwatersrand, Johannesburg, South Africa

Introduction: Several studies showed that serum intact parathyroid hormone (PTH), phosphate, and vitamin D levels differ across races. These comparative studies were largely carried out between Caucasians and black Americans. However, little is known of the existence of these associations in an African population with chronic kidney disease (CKD).

Methods: This cross-sectional multicenter study involved 293 CKD patients from 3 renal units in Johannesburg, South Africa.

Results: The 293 CKD patients (208 blacks, 85 whites) had an overall mean age of 51.1 ± 13.6 years, and black patients were significantly younger than the white patients (48.4 ± 13.6 years vs. 57.1 ± 15.5 years; $P < 0.001$). Compared with whites, blacks had higher median intact PTH (498 [range: 37–1084] pg/ml vs. 274 [range: 131–595] pg/ml; $P = 0.03$), alkaline phosphatase (122 [range: 89–192] U/L vs. 103 [range: 74–144] U/L; p = 0.03), and mean 25 OH vitamin D_3 (26.8 ± 12.7 ng/ml vs. 22.7 ± 12.2 ng/ml, $P = 0.01$) levels, whereas their median fibroblast growth factor (FGF) level was 23 (100 [range: 34–639] pg/ml vs. 233 [range: 80–1370] pg/ml; $P = 0.002$), and their mean serum phosphate (1.3 ± 0.5 vs. 1.5 ± 0.5; $P = 0.001$) levels were significantly lower. In multivariable analyses, black race was independently associated with increased log PTH ($\beta = 0.488$, $P = 0.01$) and decreased log FGF-23 ($\beta = -0.636$, $P = 0.02$). Similarly, blacks had a 3.08 times higher likelihood (95% confidence interval: 1.51–6.30; $P = 0.002$) of developing severe hyperparathyroidism than whites.

Conclusion: This study highlighted the existence of racial differences in the circulating markers of mineral bone disorders in an African CKD population.

Modifiable abnormalities of markers of mineral bone disease (MBD) have been consistently associated with adverse clinical outcomes in patients with chronic kidney disease (CKD).[1–3] Addressing these adverse outcomes has led to recommendations by various global and regional societies to assist physicians in the management of CKD MBD.[4–6] However, the consequences of these biochemical abnormalities have been shown to differ across different races; therefore, there is a need to establish race-specific target values for these markers of

MBD.[7] For example, in the MultiEthnic Study of Atherosclerosis (MESA), which involved 6436 participants, 25-hydroxyvitamin D (25-[OH] vitamin D) deficiency was associated with an increased risk of coronary heart disease in white participants, but not in black Americans.[8] A similar trend was found in the National Health and Nutrition Examination Survey, in which low 25 (OH) vitamin D was associated with a higher risk of all-cause mortality in whites compared with black participants.[9] Furthermore, fibroblast growth factor (FGF)-23, which is now being considered to be the principal mediator of secondary hyperparathyroidism, was also shown to differ across races.[10,11] In general, these comparative studies, largely from American populations, reported that compared with whites, blacks had lower levels of 25 (OH) vitamin D and FGF-23, with higher parathyroid hormone (PTH) and alkaline phosphate levels, whereas

Correspondence: Bala Waziri, Department of Internal Medicine, Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, Gauteng 2050, South Africa. E-mail: balawaziri@gmail.com

the results related to phosphate levels were inconsistent.[7,10–12]

The existence of these differences in a heterogeneous African CKD population is largely unknown. Therefore, the aim of this study was to examine racial differences in the levels of FGF-23 and traditional markers of mineral bone metabolism in a South African CKD population.

MATERIALS AND METHODS

This was a cross-sectional multicenter study in 293 CKD patients from 3 renal units in Johannesburg, South Africa. Enrolled patients were aged 18 years or older, with established CKD. We excluded patients with active malignancy, acute kidney injury, and a history of parathyroidectomy. In addition, we excluded patients of Indian and mixed race origin due to their negligible numbers.

A structured questionnaire was used to obtain the demographic characteristics, blood pressure measurements, comorbid disease, and medication history of the patients as related to CKD- MBD. Determination of race was based on self-report by the participants.

Blood Collection and Preparation

Fasting whole blood samples were collected into plain separator vacutainer tubes (for serum), and ethylenediaminetetraacetic tubes (for plasma). Samples were left to clot, and then centrifuged at 5000 rpm at 4°C for 10 minutes. Both serum and plasma were aliquoted into 1.5-ml microcentrifuge tubes and stored at −80°C. Measurements for FGF-23 and 25-(OH) vitamin D samples were done at the end of the study, whereas other biochemical parameters were measured at collection.

Blood sample collection for patients on hemodialysis was generally collected predialysis at midweek, with the exception of the postdialysis serum urea for kinetic modeling. Blood samples were collected mainly during the summer season.

Laboratory Measurements

Plasma intact PTH was measured by an electrochemiluminescence immunoassay run on a Cobas 6000 auto analyzer (Roche Diagnostics, Mannheim, Germany). This is a 2-step sandwich immunoassay that uses a capture antibody against the N-terminus and a single antibody against different parts of the C-terminus. In this study, an additional third-generation assay was not carried out to allow an assessment of the 1−84-PTH/intact-PTH ratio.

Plasma intact FGF-23 was measured using an enzyme-linked immunosorbent assay kit from EMD Millipore Corporation (Billerica, Massachusetts); assay lower limit of detection was 3.2 pg/ml.

The intact fibroblast growth factor–23 assay measures only the full-length FGF-23 (∼32 kDa), whereas the C-terminal assay measures both intact fibroblast growth factor–23 and C-terminal fragments. Although studies have reported a good correlation between the 2 assays,[13] cardiovascular risk–related thresholds may be more specific to C-terminal fragments.[14]

Plasma 25(OH) vitamin D was measured using the high-performance liquid chromatography (HPLC) kit (Recipe, Munich, Germany). HPLC was used to selectively measure 25-(OH) vitamin D2 and 25-(OH) vitamin D3 at a wavelength of 264 nm. The intra- and interassay coefficients of variation were <5%. Our institutional laboratory is a participating member in the vitamin D external quality assurance scheme. In this study, 25-(OH) vitamin D3 was used as a marker of vitamin D status to avoid confounding of the results from exogenous vitamin D supplementation.

Serum calcium, phosphate, and alkaline phosphatase were measured using the ADVIA 1800 centaur auto analyzer (Siemens Diagnostics, Tarrytown, New York).

Creatinine was measured by a modified Jaffe reaction, and the estimated glomerular filtration rate (eGFR) was assessed using the 4 variable Modified Diet Renal Disease equation[15]: GFR (in ml/min per 1.73 m^2) = 175 × SCr (exp[−1.154]) × Age (exp[−0.203]) × (0.742 if female) × (1.21 if black), where SCr is the serum creatinine.

Other biochemical parameters were determined as part of standard of care using routine laboratory techniques.

Operative Definitions and Laboratory Reference Values

The reference ranges were 2.12 to 2.50 mmol/l for calcium, 0.79 to 1.45 mmol/l for phosphate, 53 to 128 U/L for total alkaline phosphatase, and 10 to 65 pg/ml for intact PTH; the 25-(OH) vitamin D reference ranges were: <10 ng/ml as severe deficiency; 10 to 29 ng/ml as moderate deficiency; 30 to 100 ng/ml as sufficiency; and >100 ng/ml as toxic.

Based on the preceding reference values and Kidney Disease Improving Global Outcomes recommendations,[5] hyperparathyroidism was defined as PTH >130 pg/ml (2 times the upper limit of normal) and severe hyperparathyroidism as PTH >585 pg/ml (9 times the upper limit of normal).

Hyperphosphatemia and hypocalcemia were defined as serum phosphate >1.45 mmol/l and calcium as <2.12 mmol/l, respectively.

Ethical Approval

All procedures performed in this study were in accordance with the ethical standards of the institutional research committee and with the 1964

Declaration of Helsinki and its later amendments or comparable ethical standards. The research protocol was approved by the Health Research and Ethics committee of the University of the Witwatersrand; clearance certificate number M141016. Written informed consent was obtained from each patient before enrollment into the study.

Statistical Analysis

Continuous variables are presented as means $\pm$ SD or as medians and interquartile ranges (IQRs) as appropriate, whereas categorical data are reported as percentages. An independent t test or Wilcoxon rank-sum test compared continuous variables between blacks and whites, whereas Pearson's or Fisher exact tests were used for proportion comparisons.

Residuals diagnostics were conducted to evaluate the assumptions of using multiple linear regression analysis. On visualization of the plotted residuals against the fitted values for PTH and FGF-23, there was no clear pattern that showed that the assumption of constant variance for SEs was not violated. However, due to some evidence of non-normality, PTH and FGF-23 data were log-transformed.

Multiple linear regression models were used to determine the effect of independent predictors on log transformed PTH and FGF-23.

Logistic regression analysis was used to determine the predictors of severity of hyperparathyroidism. Variables with $P < 0.10$ on univariable analyses were eligible for inclusion in the multivariable analysis. Spearman correlations were used to determine the correlation between FGF-23, phosphate, PTH, 25-(OH) vitamin D3, and eGFR.

A P value of <0.05 was considered statistically significant at the 95% confidence interval. All analyses were performed using STATA version 12 (StataCorp., College Station, Texas).

RESULTS

Patient Characteristics

The 293 CKD patients included 208 black and 85 white patients with an overall mean age of 51.1 ± 13.6 years. The clinical characteristics of the patients by race are summarized in Table 1. Blacks were significantly younger, had higher blood pressure, median iPTH, alkaline phosphatase, and a mean 25-(OH) vitamin D_3 level, but they also had lower levels of median FGF-23 and serum phosphate than white patients. Diabetes mellitus was significantly more prevalent in whites. The use of CKD-MBD related medications did not differ by race.

Table 1. Characteristics of the study population

Parameters	All (n = 293)	Black (n = 208)	White (n = 85)	P value
Age (yr)	51.1 ± 13.6	48.4 ± 13.6	57.9 ± 15.5	<0.001
Gender				
Male	166 (56.7)	114 (54.8)	52 (61.2)	0.32
Female	127 (43.3)	94 (45.2)	33 (38.8)	
Systolic BP (mm Hg)	143 ± 25	146 ± 26	135 ± 19	0.007
Diastolic BP (mm Hg)	84 ± 20	89 ± 21	71 ± 11	<0.001
Hb (g/dl)	11.3 ± 2.4	11.3 ± 2.5	11.4 ± 2.2	0.83
Albumin (g/dl)	37.0 ± 7.0	37.0 ± 6.9	37.0 ± 6.7	0.55
Calcium (mmol/l)	2.22 ± 0.24	2.20 ± 0.27	2.30 ± 0.19	0.01
iPTH (pg/ml)	353 (133–914)	498 (137–1084)	274 (131–595)	0.03
FGF-23 (pg/ml)	130 (42–970)	100 (34–639)	233 (80–1370)	0.002
Phosphate (mmol/l)	1.4 ± 0.5	1.3 ± 0.5	1.5 ± 0.5	0.001
TAP (U/L)	116 (83–162)	122 (89–192)	103 (74–144)	0.03
25-OHD (ng/ml)	25.6 ± 12.7	26.8 ± 12.7	22.7 ± 12.2	0.01
<30 ng/ml	191 (91.8)	128 (61.5)	63 (74.1)	0.04
<10 ng/ml	18 (6.1)	8 (3.8)	10 (11.8)	0.01
GFR (ml/min)	9 (5–23)	8.5 (4–23)	9 (6–15)	0.19
Causes of renal disease				
HTN	188 (64.2)	141 (67.8)	47 (55.3)	0.002
DM	52 (17.7)	25 (12.0)	27 (31.8)	<0.001
ADPKD	11 (3.8)	5 (2.4)	6 (7.1)	0.06
Obstructive uropathy	6 (2.0)	3 (1.4)	3 (3.5)	0.50
Unknown	36 (12.3)	34 (16.3)	2 (2.4)	0.05
Medications				
Calcium carbonate	120 (41.0)	85 (40.9)	35 (41.2)	0.77
Alfacalcidol	111 (37.9)	78 (37.5)	33 (38.80)	0.68
Calcium carbonate (mg/d)	3429 ± 694	3375 ± 678	3500 ± 750	0.69
Alfacalcidol (μg/wk)	1.63 ± 0.38	1.86 ± 1.09	1.48 ± 0.87	0.43

ADPKD, autosomal dominant polycystic kidney disease; BP, blood pressure; DM, diabetes mellitus; FGF, fibroblast growth factor; Hb, hemglobin; HTN, hypertension; iPTH, intact parathyroid hormone; TAP, total alkaline phosphatase.
Continuous variables are presented as means ± SD or median (interquartile range), and categorical data are presented as number (%).

Comparison of Markers of CKD-MBD Between Black and White Patients

Racial variations in the levels of PTH, calcium, phosphate, and 25-(OH) vitamin D3 according to stages of CKD are shown in Table 2. Median PTH and mean 25-(OH) vitamin D3 levels were significantly higher in blacks than whites in CKD stage 5. Blacks had higher levels of total alkaline phosphatase than those of whites in stage 4 CKD. Median FGF-23 and mean serum phosphate levels increased progressively across stages of CKD and became significantly higher in whites than blacks in CKD stage 5.

Table 3 shows comparisons of markers of CKD-MBD between black and white patients with predialysis CKD and end-stage renal disease. In predialysis CKD patients, whites had significantly higher levels of FGF-23 than blacks (55 [range: 31–81] ng/ml vs. 32 [range: 22–57] ng/ml; $P = 0.01$), and higher levels of calcium (2.33 $\pm$ 0.11 vs. 2.24 $\pm$ 0.14; $P = 0.005$). Other parameters were comparable between the 2 groups. In patients with ESRD, levels of FGF-23 and phosphate were

Table 2. Markers of mineral bone metabolism by race and stages of CKD

	CKD stage 3			CKD stage 4			CKD stage 5		
Variable	Black ($n = 34$)	White ($n = 11$)	P value	Black ($n = 40$)	White ($n = 14$)	P value	Black ($n = 134$)	White ($n = 60$)	P value
IPTH (pg/ml)	120 (92–218)	86 (76–166)	0.21	193 (73–373)	228 (175–329)	0.54	758 (360–1350)	358 (179–814)	0.0004
FGF-23 (pg/ml)	30 (22–44)	42 (31–134)	0.07	35 (22–64)	63 (34–81)	0.07	329 (105–2557)	881 (187–3634)	0.03
Calcium (mmol/l)	2.25 ± 0.15	2.31 ± 0.10	0.22	2.23 ± 0.15	2.34 ± 0.13	0.009	2.18 ± 0.31	2.26 ± 0.21	0.08
Phosphate (mmol/l)	1.04 ± 0.23	1.05 ± 0.14	0.75	1.13 ± 0.25	1.27	0.10	1.44 ± 0.56	1.70 ± 0.48	0.004
25-OHD$_3$ (ng/ml)	24.3 ± 9.3	24.7 ± 11.9	0.91	24.6 ± 11.1	24.3 ± 12.80	0.94	28.1 ± 13.8	22.0 ± 12.2	0.004
TAP (U/L)	98 (77–138)	102 (78–123)	0.76	114 (97–166)	88 (74–111)	0.03	123 (88–209)	128 (73–226)	0.14

25-OHD$_3$, 25-(OH) vitamin D3; FGF, fibroblast growth factor; IPTH, intact parathyroid hormone; TAP, total alkaline phosphatase.
Continuous variables are presented as means ± SDs or median (interquartile range), and categorical data are presented as numbers (%).

significantly higher in whites than in blacks (881 [range: 187–3634] vs. 329 [range: 105–2557]; $P = 0.03$) and (1.70 ± 0.48 vs. 1.44 ± 0.56; $P = 0.004$), respectively. Compared with whites, blacks had higher levels of PTH (758 [range: 360–1350] vs. 358 [range: 179–814]; $P = 0.0004$) and 25-OH vitamin D (28.1 ± 13.8 vs. 22.0 ± 12.2; $P = 0.004$).

Comparisons between blacks and whites in the prevalence of abnormal levels of calcium, PTH, phosphate, and 25-(OH) vitamin D3 across stages of CKD are shown in Figure 1a–d. The proportion of patients with hyperphosphatemia (72.7% vs. 42.9%; $P < 0.001$) and vitamin D deficiency (76.7% vs. 57.5%; $P = 0.01$) was higher in whites than blacks in CKD stage 5.

The prevalence of abnormal levels of other markers of CKD-MBD was similar across the study populations.

Associations Between Race, Clinical Characteristics, and Markers of CKD-MBD

In line with a previous study,[16] in a multiple regression analysis adjusted for age, diabetes status, GFR, serum

Table 3. Markers of CKD-MBD by race in predialysis and end-stage renal disease

Variable	Black	White	P Value
Predialysis	$n = 74$	$n = 25$	
IPTH (pg/ml)	160 (84–280)	174 (94–253)	0.81
FGF-23 (pg/ml)	32 (22–57)	55 (31–81)	0.01
Calcium (mmol/l)	2.24 ± 0.14	2.33 ± 0.11	0.005
Phosphate (mmol/l)	1.09 ± 0.24	1.18 ± 0.27	0.12
25-OHD$_3$ (ng/ml)	24.4 ± 10.3	24.5 ± 12.1	0.99
TAP (U/L)	111 (89–141)	74 (74–114)	0.09
GFR (ml/min per 1.73 m²)	30.9 ± 12.7	30.1 ± 12.5	0.77
End-stage renal disease	$n = 134$	$n = 60$	
IPTH (pg/ml)	758 (360–1350)	358 (179–814)	0.0004
FGF-23 (pg/ml)	329 (105–2557)	881 (187–3634)	0.03
Calcium (mmol/l)	2.18 ± 0.31	2.26 ± 0.21	0.08
Phosphate (mmol/l)	1.44 ± 0.56	1.70 ± 0.48	0.004
25-OHD$_3$ (ng/ml)	28.1 ± 13.8	22.0 ± 12.2	0.004
TAP (U/L)	123 (88–209)	128 (73–226)	0.14
Kt/V	1.41 ± 0.30	1.46 ± 0.30	0.40

Continuous variables are presented as means ± SDs or median (interquartile range).
25-OHD$_3$, 25-(OH) vitamin D3; FGF, fibroblast growth factor; GFR, glomerular filtration rate; IPTH, intact parathyroid hormone; MBD, mineral bone disease; TAP, total alkaline phosphatase.

calcium, phosphate, and alkaline phosphate levels, black race remained significantly associated with increased log PTH ($\beta = 0.488$, $P = 0.01$) and decreased log FGF-23 ($\beta = -0.636$, $P = 0.02$) (Table 4). Similarly, we found a persistent significant association between log FGF-23, calcium, phosphate, and GFR. In unadjusted univariable analysis, white race was significantly associated with decreased 25-(OH) vitamin D3; however, this was attenuated after adjusting for age, diabetes status, calcium, and GFR.

Further exploration of the association between FGF-23 and other markers of CKD-MBD showed that FGF-23 correlated positively with serum phosphate (r = 0.55, $P < 0.001$) and PTH (r = 0.40, $P < 0.001$), and inversely with eGFR (r = −0.61, $P < 0.001$); these overall relationships persisted when stratified by race, with interaction P values of <0.001 (Figures 2 and 3).

Determinants of Secondary Hyperparathyroidism

In logistic regression analysis, the independent predictors of severe hyperparathyroidism were black race (odds ratio: 3.08; 95% confidence interval: 1.51–6.30; $P = 0.002$) and GFR <15 ml/min (odds ratio 10.07; 95% confidence interval: 4.70–21.56; $P < 0.0001$; Table 5).

DISCUSSION

The racial disparities in markers of CKD-MBD have been documented in CKD populations in previous studies from America and Europe.[16,17] A few studies from Africa have documented similar findings in healthy populations but not in CKD patients.[18,19] In this present study, with the exception of 25-(OH) vitamin D3 levels, our findings were consistent with those of previous studies[12,16]; we found that PTH and alkaline phosphatase levels were higher in black patients than those in white patients in CKD stage 5 and CKD stage 4, respectively. After adjusting for the inconsistency between the 2 groups with regard to diabetic status and age, black race still remained significantly associated with higher levels of PTH. Although the mechanisms behind these discrepancies
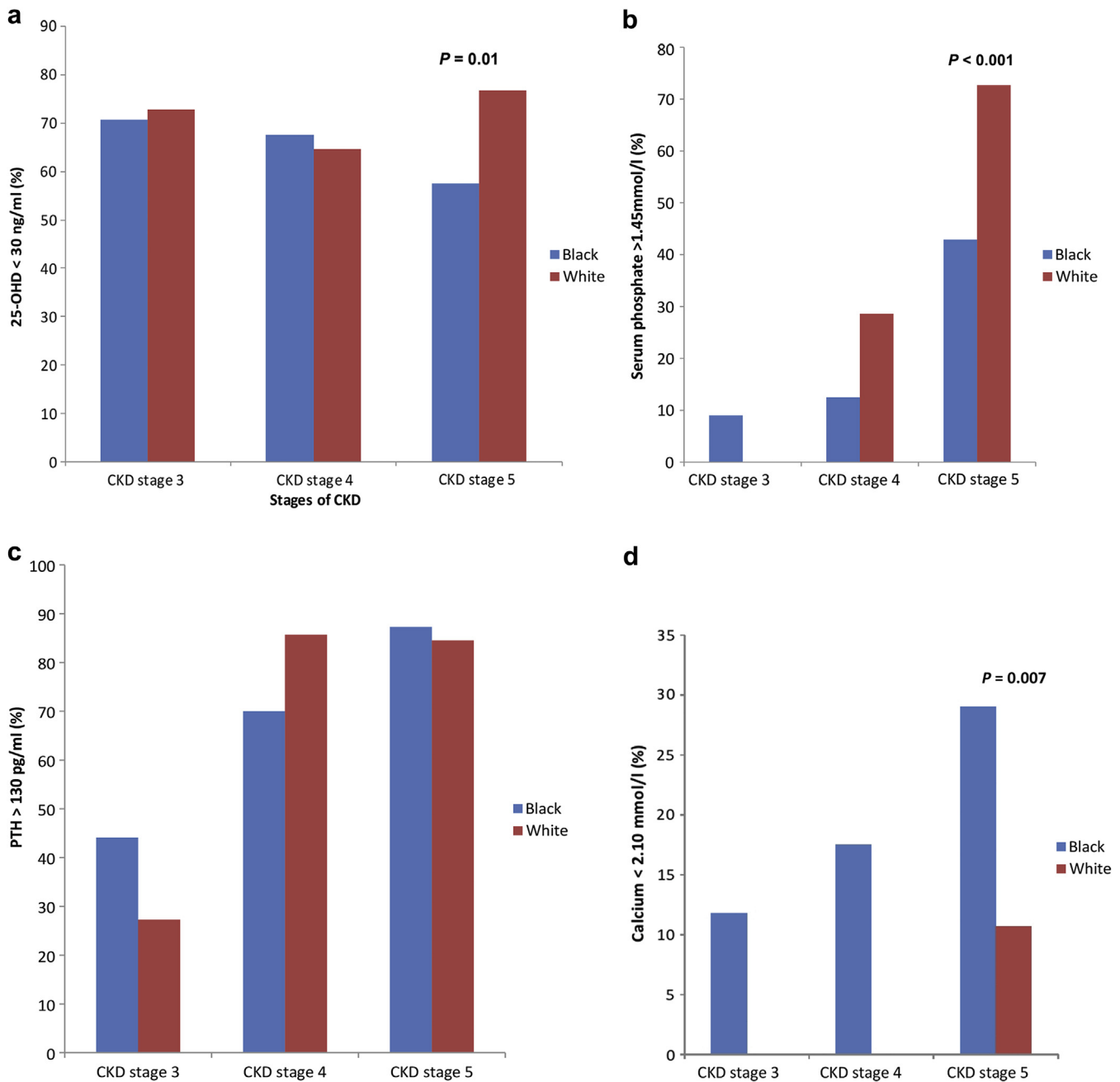
Figure 1. Prevalence of 25 (OH) D3 deficiency by race and stages of chronic kidney disease (CKD) (a). Prevalence of hyperphosphatemia by race and stages of CKD (b). Prevalence of hyperparathyroidism by race and stages of CKD (c). Prevalence of hypocalcemia by race and stages of CKD (d). PTH, parathyroid hormone.

remain largely unclear, several reasons proposed by previous studies included racial differences in skeletal responsiveness to PTH levels, racial variations in sensitivity to the phosphaturic effect of PTH and FGF-23, dietary intake of food rich in phosphate, and underlying genetic differences in bone mineral metabolism.[16,20,21]

An interesting and unexpected finding was the significantly higher levels of 25-(OH) vitamin D3 in CKD stage 5 in blacks compared with whites; this was contrary to most previous studies that reported lower levels of 25-(OH) vitamin D, which was attributed to

skin pigmentation.[16] It is anticipated that increased skin pigmentation in blacks will lead to decreased synthesis of 25-(OH) vitamin D from 7-dehydrocholesterol through exposure to sunlight. One of the limitations of our study was the nonavailability of information related to sun exposure, which could have accounted for these differences; it is possible that our black patients with CKD stage 5 spent more time outdoors (outdoor occupations in a sunny climate), which could have led to more sun exposure compared with white patients. However, some studies showed that blacks and whites have equal capacities to

Table 4. Multivariable analysis of determinant of PTH, FGF-23, and 25-(OH)D₃

Variables	β coefficient	P value	β coefficient	P value
	Unadjusted		Adjusted	
Dependent variable (log PTH)				
Independent variable				
Age	−0.010	0.03	0.013	0.02
Diabetes	−0.390	0.02	−0.112	0.61
Female gender	0.236	0.09	0.247	0.12
Black race	0.413	0.007	0.488	0.01
Calcium	−1.248	<0.001	−1.038	0.001
Phosphate	0.541	<0.001	0.179	0.26
25-(OH)D₃	−0.005	0.35	−0.002	0.77
Alkaline phosphatase	0.002	<0.001	0.002	0.001
GFR	−0.040	<0.001	−0.030	P<0.001
Dependent variable (log FGF-23)				
Independent variable				
Age	−0.033	<0.001	−0.016	0.02
Diabetes	−0.763	0.01	−0.318	0.21
Black race	−0.765	0.003	−0.636	0.02
PTH	0.622	<0.001	0.294	0.001
Calcium	0.921	0.05	1.239	0.004
Phosphate	2.077	<0.001	1.041	<0.001
25-(OH)D₃	0.019	0.04	0.009	0.31
GFR	−1.329	<0.001	−0.752	P<0.001
25-(OH)D₃ (dependent variable)				
Independent variable				
Age	−0.215	<0.001	−0.207	<0.001
Diabetes	−8.745	<0.001	−4.610	0.03
White race	−4.063	0.01	−1.471	0.41
Albumin	0.385	0.001	0.267	0.02
Calcium	9.214	0.002	11.461	<0.001

25-OHD₃, 25-(OH) vitamin D3; FGF, fibroblast growth factor; GFR, glomerular filtration rate; PTH, parathyroid hormone.

synthesize vitamin D postexposure to repeated high doses of ultraviolet B light.[22,23] Brazerol et al.,[23] who compared the skin capacity in blacks and whites exposed to similar doses of ultraviolet B rays (280–315 nm) twice a week for 6 weeks, reported similar response to vitamin D synthesis despite the fact that blacks had lower baseline 25-(OH) vitamin D levels than the white participants. A previous study from South Africa with the aim of assessing vascular calcification in hemodialysis patients found no difference in 25-(OH) vitamin D levels between black and white patients.[24] In addition, the inconsistencies that existed in the relationship between vitamin D levels and clinical outcomes in blacks suggested that the mechanism behind the racial disparities was complex and multifactorial. For example, despite lower levels of 25-(OH) vitamin D demonstrated by some studies, blacks had lower rates of osteoporosis and bone fractures than age- and sex-matched white participants.[25,26] However, the observed lower rates of osteoporosis in blacks might partly be attributable to differences in bone size and numbers of trabeccullae.[27] Furthermore, ethnic

variations in the levels of 25-(OH) vitamin D might partially be explained by differences in vitamin D−binding proteins genotypes and circulating levels of vitamin D−binding protein between black and white populations.[28,29] For instance, a study involving Healthy Aging in Neighborhoods of Diversity across the life Span (HANDLS) cohort, showed that the T allele at rs 7041 was associated decreased levels of 25-(OH) vitamin D among blacks, and that genetic variants independently accounted for 79.4% of the variations in the levels of vitamin D−binding protein.[29] These associations need to be explored in our heterogenous African population.

Studies relating to the prevalence of 25-(OH) vitamin D across stages of CKD revealed conflicting results. Consistent with previous studies, we found no association between vitamin D status and stages of CKD.[12] In contrast to our study, the prevalence of 25-(OH) vitamin D deficiency and/or insufficiency rose slightly across the stages of CKD in the Nephro Test study.[30] However, it is noteworthy that comparisons of vitamin D status across these studies were somewhat hampered by differences in the skin pigmentation of the participants, latitude, cutoff values, and assay methods used among the studies. In addition, it was suggested that vitamin D deficiency was more profound in diabetic patients because of heavy urinary loss of the vitamin D−binding protein.[30,31] Therefore, the higher prevalence of diabetes in our white participants could have accounted for the racial discrepancy in the levels of 25-(OH) vitamin D. This was further supported by the attenuation of the significant association between white race and 25-(OH) vitamin D3 after adjustment for diabetes status in the linear regression model, with 25-(OH) vitamin D3 as the dependent variable.

Consistent with previous studies, phosphate levels increased with worsening of kidney function, and the increase was significantly higher in whites than blacks at CKD stage 5. It was possible that white patients were more likely to consume dairy products, which could have accounted for the higher phosphate levels; this could not to be ascertained in this study because of the nonavailability of dietary history. However, large studies from the United States reported that African Americans had lower consumption of dairy products than whites.[32,33] However, understanding racial differences in serum phosphate levels is intriguing and other factors besides dietary phosphate need to be considered. For example, contrary to our findings, some studies showed surprisingly increased serum phosphate levels in blacks compared with whites, despite increased levels of PTH, and thus attributed these differences to reduced urinary phosphate
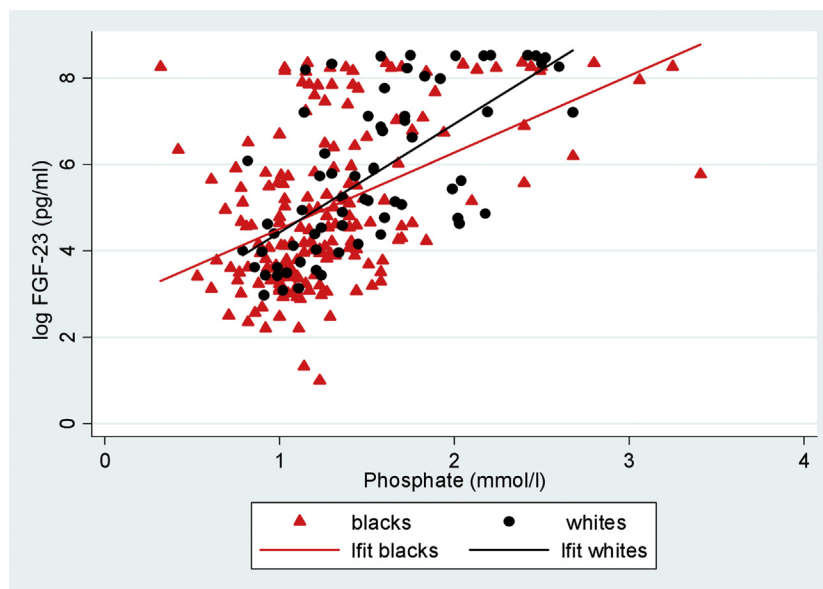
Figure 2. Relationship between fibroblast growth factor (FGF-23) and phosphate by race. lfit, line fit.

excretion and lower FGF-23 in blacks.[10,11] In line with these studies, our black patients had lower levels of FGF-23 compared with whites. It was possible that higher phosphate levels in our white participants could have accounted for the differential levels in FGF-23 as a compensatory mechanism. However, this is in contrast to the explanation offered by a previous study, in which the lower levels of FGF-23 in the black participants, despite higher levels of phosphate, were the result of decreased FGF-23 expression.[10] The complexity in racial disparities with phosphate and FGF-23 levels was further compounded by variations in the use of phosphate binders and alfacalcidol. In this study, there were no differences between blacks and whites in the use of calcium carbonate and alfacalcidol.

Consistent with previous studies,[11,34] FGF-23 correlated positively with phosphate and PTH, and inversely with GFR. The directions of these associations are physiologically plausible, with FGF-23 attempting to mitigate the effect of excess phosphate with worsening renal function. It is also noteworthy that significant positive association with white race persisted after adjusting for other confounding variables. In addition, the significant positive association with white race persisted after adjusting for other confounding variables.

The limitations of our study included the cross-sectional study design; therefore, we could not determine the longitudinal changes in markers of CKD-MBD and the seasonal variation in 25 (OH) vitamin D levels.
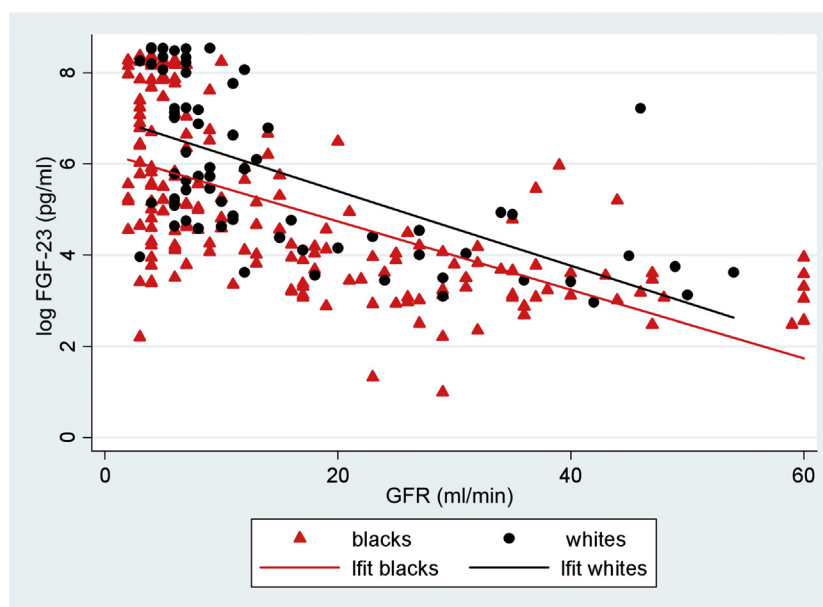

Figure 3. Relationship between fibroblast growth factor (FGF-23) and glomerular filtration rate (GFR). lfit, line fit.

Table 5. Predictors of severe hyperparathyroidism (parathyroid hormone >585 ng/ml)

Variable	OR	95% (CI)	P value
Age <65 yr	0.77	0.32–1.84	0.56
Black race	3.08	1.51–6.30	0.002
Diabetes	0.57	0.25–1.31	0.19
25-(OH)D₃ (<30 ng/ml)	1.68	0.90–3.13	0.10
GFR <15 ml/min	10.07	4.70–21.56	<0.001
Female gender	0.82	0.47–1.43	0.48
Hyperphosphatemia (>1.45 mmol/l)	1.39	0.75–2.58	0.29
Hypocalcemia <2.10 mmol/l	1.46	0.74–2.92	0.28

25-OHD$_3$, 25-(OH) vitamin D3; CI, confidence interval; GFR, glomerular filtration rate; OR, odd ratio.

Information relating to ultraviolet B exposure and dietary phosphate were also lacking.

Due to the nonavailability of data on 24-hour urinary protein, we could not adjust for possible racial differences in proteinuria, which is associated with the urinary loss of the vitamin D—binding protein.

The strengths of this study lie in the heterogeneous nature of our study population (black and white patients) in an African setting, which allowed comparisons of data not only for black Africans with black Americans, but also between whites in Africa and the United States and Europe.

To our knowledge, few studies have compared FGF-23 levels across races in developed countries, and no such studies have been reported from Africa. Therefore, this is the first study in Africa that has given important insights with regard to the associations among FGF-23, traditional markers of CKD-MBD, and race in an African CKD population. Finally, in this study, high-performance liquid chromatography, which is considered to be the gold standard for vitamin D measurement,[35] was used to specifically measure 25-(OH) vitamin D3 and 25-(OH) vitamin D2. In addition, with the exception of high-performance liquid chromatography, several test methodologies were shown to demonstrate a considerable variation of individual 25-(OH) vitamin D values compared with liquid chromatography–mass spectrometry/mass spectrometry—defined target concentrations.[36]

CONCLUSION

There was a racial difference in the markers of CKD-MBD; compared with whites, African blacks had higher levels of PTH, alkaline phosphatase, and lower levels of FGF-23 and serum phosphate. It remains unclear whether the present CKD-MBD management guidelines are appropriate for all races.

DISCLOSURE

SN received research grant support from MRC SA. All the authors declared no competing interests.

REFERENCES

1. Floege J, Kim J, Ireland E, et al. Serum iPTH, calcium and phosphate, and the risk of mortality in a European haemodialysis population. *Nephrol Dial Transplant*. 2011;26:1948–1955.

2. Block GA, Klassen PS, Lazarus JM, Ofsthun N, Lowrie EG, Chertow GM. Mineral metabolism, mortality, and morbidity in maintenance hemodialysis. *J Am Soc Nephrol*. 2004;15: 2208–2218.

3. Tentori F, Blayney MJ, Albert JM, et al. Mortality risk for dialysis patients with different levels of serum calcium, phosphorus, and PTH: the Dialysis Outcomes and Practice Patterns Study (DOPPS). *Am J Kidney Dis*. 2008;52:519–530.

4. Jindal K, Chan CT, Deziel C, et al. Hemodialysis clinical practice guidelines for the Canadian Society of Nephrology. *J Am Soc Nephrol*. 2006;17(3 Suppl 1):S1–S27.

5. Kidney Disease: Improving Global Outcomes (KDIGO) CKD-MBD Work Group. KDIGO clinical practice guideline for the diagnosis, evaluation, prevention, and treatment of Chronic Kidney Disease-Mineral and Bone Disorder (CKD-MBD). *Kidney Int Suppl*. 2009;113:S1–S130.

6. National Kidney Foundation. K/DOQI clinical practice guidelines for bone metabolism and disease in chronic kidney disease. *Am J Kidney Dis*. 2003;42(4 Suppl 3):S1–S201.

7. Scialla JJ, Parekh RS, Eustace JA, et al. Race, mineral homeostasis and mortality in patients with end-stage renal disease on dialysis. *Am J Nephrol*. 2015;42:25–34.

8. Robinson-Cohen C, Hoofnagle AN, Ix JH, et al. Racial differences in the association of serum 25-hydroxyvitamin D concentration with coronary heart disease events. *JAMA*. 2013;310:179–188.

9. Melamed ML, Michos ED, Post W, Astor B. 25-hydroxyvitamin D levels and the risk of mortality in the general population. *Arch Intern Med*. 2008;168:1629–1637.

10. Gutierrez OM, Mannstadt M, Isakova T, et al. Fibroblast growth factor 23 and mortality among patients undergoing hemodialysis. *N Engl J Med*. 2008;359:584–592.

11. Jovanovich A, Chonchol M, Cheung AK, et al. Racial differences in markers of mineral metabolism in advanced chronic kidney disease. *Clin J Am Soc Nephrol*. 2012;7: 640–647.

12. Ennis J, Worcester E, Coe F. Contribution of calcium, phosphorus and 25-hydroxyvitamin D to the excessive severity of secondary hyperparathyroidism in African-Americans with CKD. *Nephrol Dial Transplant*. 2012;27:2847–2853.

13. Heine GH, Seiler S, Fliser D. FGF-23: the rise of a novel cardiovascular risk marker in CKD. *Nephrol Dial Transplant*. 2012;27:3072–3081.

14. Kendrick J, Cheung AK, Kaufman JS, et al. FGF-23 associates with death, cardiovascular events, and initiation of chronic dialysis. *J Am Soc Nephrol*. 2011;22:1913–1922.

15. Levey AS, Coresh J, Greene T, et al. Using standardized serum creatinine values in the modification of diet in renal disease study equation for estimating glomerular filtration rate. *Ann Int Med*. 2006;145:247–254.

16. Gutierrez OM, Isakova T, Andress DL, Levin A, Wolf M. Prevalence and severity of disordered mineral metabolism in blacks with chronic kidney disease. *Kidney Int*. 2008;73:956–962.

17. Malluche HH, Mawad HW, Monier-Faugere MC. Renal osteodystrophy in the first decade of the new millennium: analysis of 630 bone biopsies in black and white patients. *J Bone Miner Res*. 2011;26:1368–1376.

18. George JA, Norris SA, van Deventer HE, Crowther NJ. The association of 25 hydroxyvitamin D and parathyroid hormone with metabolic syndrome in two ethnic groups in South Africa. *PLoS ONE*. 2013;8:e61282.

19. Daniels ED, Pettifor JM, Schnitzler CM, Moodley GP, Zachen D. Differences in mineral homeostasis, volumetric bone mass and femoral neck axis length in black and white South African women. *Osteoporos Int*. 1997;7:105–112.

20. Goodman WG, Quarles LD. Development and progression of secondary hyperparathyroidism in chronic kidney disease: lessons from molecular genetics. *Kidney Int*. 2008;74:276–288.

21. Cosman F, Morgan DC, Nieves JW, et al. Resistance to bone resorbing effects of PTH in black women. *J Bone Miner Res*. 1997;12:958–966.

22. Matsuoka LY, Wortsman J, Chen TC, Holick MF. Compensation for the interracial variance in the cutaneous synthesis of vitamin D. *J Lab Clin Med*. 1995;126:452–457.

23. Brazerol WF, McPhee AJ, Mimouni F, Specker BL, Tsang RC. Serial ultraviolet B exposure and serum 25 hydroxyvitamin D response in young adult American blacks and whites: no racial differences. *J Am Coll Nutr*. 1988;7:111–118.

24. Freercks R, Swanepoel C, Carrara H, Moosa S, Lachman A, Rayner B. Vascular calcification in South African dialysis patients: ethnic variation, prevalence, detection and haemodynamic correlates. *Nephrology*. 2012;17:607–615.

25. Baron JA, Barrett J, Malenka D, et al. Racial differences in fracture risk. *Epidemiology (Cambridge, Mass)*. 1994;5:42–47.

26. Barrett-Connor E, Siris ES, Wehren LE, et al. Osteoporosis and fracture risk in women of different ethnic groups. *J Bone Miner Res*. 2005;20:185–194.

27. Bhudhikanok GS, Wang MC, Eckert K, Matkin C, Marcus R, Bachrach LK. Differences in bone mineral in young Asian and Caucasian Americans may reflect differences in bone size. *J Bone Miner Res*. 1996;11:1545–1556.

28. Yousefzadeh P, Shapses SA, Wang X. Vitamin D binding protein impact on 25-hydroxyvitamin D levels under different physiologic and pathologic conditions. *Int J Endocrinol*. 2014;2014:981581.

29. Powe CE, Evans MK, Wenger J, et al. Vitamin D-binding protein and vitamin D status of black Americans and white Americans. *N Engl J Med*. 2013;369:1991–2000.

30. Urena-Torres P, Metzger M, Haymann JP, et al. Association of kidney function, vitamin D deficiency, and circulating markers of mineral and bone disorders in CKD. *Am J Kidney Dis*. 2011;58:544–553.

31. Fernandez-Juarez G, Luno J, Barrio V, et al. 25 (OH) vitamin D levels and renal disease progression in patients with type 2 diabetic nephropathy and blockade of the renin-angiotensin system. *Clin J Am Soc Nephrol*. 2013;8:1870–1876.

32. Fulgoni V 3rd, Nicholls J, Reed A, et al. Dairy consumption and related nutrient intake in African-American adults and children in the United States: continuing survey of food intakes by individuals 1994-1996, 1998, and the National Health And Nutrition Examination Survey 1999-2000. *J Am Diet Assoc*. 2007;107:256–264.

33. Ranganathan R, Nicklas TA, Yang SJ, Berenson GS. The nutritional impact of dairy product consumption on dietary intakes of adults (1995-1996): the Bogalusa Heart Study. *J Am Diet Assoc*. 2005;105:1391–1400.

34. Isakova T, Wahl P, Vargas GS, et al. Fibroblast growth factor 23 is elevated before parathyroid hormone and phosphate in chronic kidney disease. *Kidney Int*. 2011;79:1370–1378.

35. Binkley N, Krueger D, Cowgill CS, et al. Assay variation confounds the diagnosis of hypovitaminosis D: a call for standardization. *J Clin Endocrinol Metab*. 2004;89:3152–3157.

36. Roth HJ, Schmidt-Gayk H, Weber H, Niederau C. Accuracy and clinical implications of seven 25-hydroxyvitamin D methods compared with liquid chromatography-tandem mass spectrometry as a reference. *Ann Clin Biochem*. 2008;45(Pt 2):153–159.