Sleep disordered breathing and fibroblast growth factor 23 in the Hispanic Community Health Study/Study of Latinos

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ABSTRACT

Preclinical data suggest that hypoxia stimulates fibroblast growth factor 23 (FGF23) transcription and cleavage in osteocytes, resulting in elevated circulating c-terminal (cFGF23) levels but normal intact FGF23 (iFGF23) levels. We conducted a case-control study within the Hispanic Community Health Study/Study of Latinos to investigate whether sleep disordered breathing, as a model of hypoxemia, is independently associated with elevated cFGF23 levels in the general population and with elevated cFGF23 and iFGF23 levels in patients with chronic kidney disease (CKD), in whom FGF23 cleavage may be impaired. Cases ($n = 602$) had severe sleep disordered breathing defined as an apnea/hypopnea index (AHI) of ≥ 30 . Controls without severe sleep disordered breathing ($n = 602$) were matched for sex and CKD stage. The median AHI in the cases was 45.8 (IQR 35.5–62.5) compared to 2.6 (IQR 0.6–8.2) in the controls. Cases had higher cFGF23 levels than controls (66.2 RU/mL, IQR 52.8–98.4 vs. 61.2 RU/mL, IQR 49.5–80.1, p value < 0.001). There were no differences in iFGF23 levels between cases and controls. In adjusted linear regression and multinomial regression analyses, body mass index attenuated the relationship between severe sleep disordered breathing and cFGF23 levels. No significant relationships were seen in analyses of severe sleep disordered breathing and iFGF23 levels or in analyses of iFGF23 and cFGF23 stratified by CKD status. Additional studies using other models of intermittent and chronic hypoxia are needed to confirm whether hypoxia stimulates FGF23 transcription in humans and to determine the impact on iFGF23 levels in CKD.

1. Introduction

Fibroblast growth factor 23 (FGF23) is an osteocyte-derived hormone that regulates systemic phosphate and vitamin D homeostasis [1]. Chronic kidney disease (CKD) represents the most common setting of

secondary FGF23 excess. FGF23 levels rise early in CKD and FGF23 levels may be thousands-fold higher than in CKD by the time end stage renal disease occurs [2]. Preclinical data demonstrate direct toxic effects of FGF23 on cardiac myocytes through activation of fibroblast growth factor receptor 4 [3, 4]. Results from observational studies are

Abbreviations: FGF23, fibroblast growth factor 23; cFGF23, c-terminal fibroblast growth factor 23; iFGF23, intact fibroblast growth factor 23; CKD, chronic kidney disease; HCHS/SOL, Hispanic Community Health Study/Study of Latinos; AHI, apnea/hypopnea index; UACR, urinary albumin-to-creatinine ratio; eGFR, estimated glomerular filtration rate; CRP, C-reactive protein; OR, odds ratio

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corroborative: higher FGF23 levels are strongly associated with left ventricular hypertrophy, cardiovascular events and mortality in patients with CKD [5–12]. Collectively, these findings have spurred investigations to identify modifiable factors that regulate circulating FGF23 levels.

Studies in animal models demonstrate that the balance between FGF23 transcription and post-translational cleavage of newly synthesized FGF23 protein determines levels of circulating bioactive FGF23 [13]. FGF23 transcription is upregulated in response to known stimuli and in health, this is matched by an increase in post-translational FGF23 cleavage [13, 14]. Thus, circulating levels of biologically-active hormone, which are measured by intact assays (iFGF23), stay within the normal range, but C-terminal inactive fragments are elevated, which are measured by assays that detect both the biologically-active intact hormone and C-terminal fragments (cFGF23) [13–15]. Iron deficiency and inflammation are two recently discovered stimuli of FGF23 transcription that are accompanied by parallel increases in FGF23 cleavage, resulting in elevations of cFGF23 but not iFGF23 levels. In CKD, FGF23 cleavage may be impaired such that stimuli that upregulate FGF23 transcription in osteocytes produce elevations in both cFGF23 and iFGF23 [1]. Preclinical data demonstrate that hypoxia may be a novel stimulus of FGF23 transcription [16]. However, few clinical studies have investigated the effect of hypoxemia on circulating FGF23 levels in health and in CKD.

Sleep disordered breathing is a common cause of nocturnal hypoxemia that is strongly associated with increased risk of cardiovascular disease in the general population and in patients with CKD [17–25]. We conducted a nested case-control study within a large prospective cohort of individuals with and without CKD to test the hypothesis that individuals with severe sleep disordered breathing would have higher baseline cFGF23 levels compared to individuals without severe sleep disordered breathing. Given that post-translational cleavage of FGF23 may be impaired in CKD [15, 26], in exploratory analyses, we tested the hypothesis that individuals with sleep disordered breathing and CKD, defined as an estimated glomerular filtration rate $< 60 \text{ mL/min/1.73 m}^2$, would have elevations in both cFGF23 and iFGF23 levels.

2. Methods

2.1. Study population and design

The Hispanic Community Health Study/Study of Latinos (HCHS/SOL) is an ongoing multi-center prospective cohort study of risk factors for and the prevalence of cardiovascular, pulmonary and chronic diseases in the Hispanic/Latino population [27, 28]. The HCHS/SOL study population included 16,415 participants aged 18–74 years who were recruited between 2008 and 2011 from four field centers across the United States: Chicago, IL; Bronx, NY; Miami, FL; San Diego, CA. The sampling design and implementation of the HCHS/SOL were previously described [27, 28]. All participants gave written informed consent, and the study was approved by the Institutional Review Board at each participating field center [27, 28].

Of the total HCHS/SOL study population, 14,440 individuals underwent a sleep study at their baseline visit with sufficient somnography data for analyses [29]. For this case-control study [30], we identified 602 individuals who met the severe sleep disordered breathing case definition and who had plasma available for additional measurements. We then selected 602 controls, frequency-matched for sex and CKD stage, who did not have severe sleep disordered breathing. Our total population included 1204 individuals in whom we measured baseline plasma cFGF23, iFGF23 and phosphate (Fig. 1).

2.2. Primary exposure

The primary exposure was the presence of severe sleep disordered breathing. Participants were instructed on use of a Type 3 sleep apnea

monitor for overnight recording at their baseline clinic visit, when participants also had their blood drawn [29]. This monitor measures airflow via nasal cannula, pulse rate, snoring levels, and hemoglobin oxygen saturation via transcutaneous oximetry [29]. All sleep records were scored at a central sleep reading center by a certified polysomnologist, who manually edited artifacts, identified periods of sleep, and interpreted the oxyhemoglobin desaturation associated with each respiratory event [29]. Respiratory events were defined as a 50% or greater reduction in airflow that lasted $\geq 10 \text{ s}$ [29]. Apnea/hypopnea index (AHI) ≥ 30 was defined as > 30 respiratory events per estimated sleep hour with 3% desaturation. Within and between scorer reliability was high for the AHI (intra-class and inter-class correlation coefficients > 0.95) [29]. Severe sleep disordered breathing was defined as AHI ≥ 30 on overnight sleep testing.

2.3. Outcomes

Our primary outcomes were plasma cFGF23 and iFGF23 levels. We measured FGF23 using Quidel's (formerly known as Immutopics) cFGF23 and iFGF23 assays (San Clemente, California, Quidel) at the HCHS/SOL Central Laboratory at the University of Minnesota after a single thaw of frozen samples. To quantify precision, we measured in a blinded manner a random selection of 10% of samples in duplicate. The mean intra-assay coefficients of variation for cFGF23 and iFGF23 were 4.4% and 6.6%. The inter-assay coefficients of variation ranged from 4.8–8.3% for cFGF23 and 6.1–6.4% for iFGF23.

2.4. Covariates

Information on participant demographics, medication use and clinical data was collected at the baseline visit [27, 28]. Resting blood pressure was measured via standardized protocols. Diabetes was defined as one of the following: fasting plasma glucose level $\geq 126 \text{ mg/dL}$, a 2-hour post-load glucose level $\geq 200 \text{ mg/dL}$, hemoglobin A1C level $\geq 6.5\%$, or documented use of hypoglycemic agents on their medication inventory [31, 32]. The centralized laboratory measured serum creatinine and calcium, and urinary albumin-to-creatinine ratio (UACR) via standard assays. Phosphate was measured in plasma on a Roche Cobas 6000 Chemistry Analyzer (Roche Diagnostics Corporation) using a colorimetric method. For plasma phosphate, the laboratory intra-assay CV was 0.8% at a concentration of 6.1 mg/dL and the inter-assay CV was 1.9% at a concentration of 3.7 mg/dL . CKD was defined as an estimated glomerular filtration rate (eGFR) $< 60 \text{ mL/min/1.73 m}^2$ using the creatinine-based Chronic Kidney Disease Epidemiology Collaboration Cystatin C equation [33]. The amount of missing data for covariates was minimal and less than $< 0.5\%$.

2.5. Statistical analysis

We used standard descriptive statistics to summarize and compare baseline characteristics of our study population according to presence of severe sleep disordered breathing at study enrollment.

To test the association of sleep disordered breathing with FGF23 levels, we fit separate models for cFGF23 and iFGF23. First, we used multinomial logistic regression analyses with cFGF23 and iFGF23 expressed in ascending quartiles, as previously done [7, 34]. We then examined FGF23 as a continuous outcome variable in linear regression models, for which we log transformed cFGF23 and iFGF23 levels because they were not normally distributed. We hierarchically adjusted all models for potential confounding factors, including demographics (age and sex), CKD risk factors (eGFR, albuminuria and plasma phosphate), comorbidities (blood pressure, smoking, diabetes, and coronary heart disease), and inflammation (C-reactive protein [CRP]). In our final model, we further adjusted for body mass index.

Given that FGF23 cleavage may be impaired in CKD [15, 26], we performed pre-planned exploratory subgroup analyses in those with

Fig. 1. Case control study design

Controls are frequency matched for sex and CKD stage.

Abbreviations: HCHS/SOL, Hispanic Community Health Study/Study of Latinos; N, number; SDB, sleep disordered breathing; CKD, chronic kidney disease.

and without CKD. In analyses restricted to those with or without CKD, we compared those with severe sleep disordered breathing to controls using standard descriptive statistics. We used linear regression to investigate the associations of severe sleep disordered breathing and cFGF23 and iFGF23 expressed as a continuous variable. We used a similar hierarchical modeling approach to that used in our total population.

In sensitivity analyses, we replaced body mass index with waist circumference, which may be a more accurate measure of adiposity, in our multinomial and linear regression models.

Analyses were performed using SAS version 9.4 (Cary, NC, USA). Two-sided *p* values < 0.05 were considered statistically significant.

3. Results

3.1. Associations of sleep disordered breathing with FGF23 levels

Table 1 presents clinical and demographic characteristics of the 1204 HCHS/SOL participants according to presence of severe sleep disordered breathing. The median AHI in individuals with severe sleep disordered breathing was 45.8 (IQR 35.5–62.5) compared to 2.6 (IQR 0.6–8.2) in the control group. Compared to controls, individuals with severe sleep disordered breathing were older and had higher prevalence of diabetes. Individuals with severe sleep disordered breathing also had higher levels of inflammation and were more obese compared to controls (CRP 3.4 mg/L, IQR 1.8–6.9 vs. 2.0 mg/L, IQR 0.9–4.8, *p* value < 0.001; body mass index 35.2 ± 6.6 kg/m² vs. 29.9 ± 5.8 kg/m², *p* value < 0.001, **Table 1**). Levels of cFGF23 were slightly higher in individuals with severe sleep disordered breathing compared to controls (66.2 RU/mL, IQR 52.8–98.4 vs. 61.2 RU/mL, IQR 49.5–80.1, *p* value < 0.001, **Table 1**). iFGF23 levels did not differ between individuals with severe sleep disordered breathing and those without (**Table 1**).

We investigated the associations of sleep disordered breathing with ascending FGF23 quartiles using multinomial logistic regression analyses. In unadjusted analyses, individuals with severe sleep disordered breathing had higher odds of belonging to cFGF23 Quartile 4 versus

Table 1

Characteristics of total population according to sleep disordered breathing status.

	Severe SDB	Severe SDB	<i>p</i> value
	Yes N = 602	No N = 602	
Age, years	52.8 ± 11.2	48.7 ± 14.5	< 0.001
Female, %	211 (35.1)	210 (34.9)	1.0
BMI, kg/m ²	35.2 ± 6.6	29.9 ± 5.8	< 0.001
SBP, mm Hg	129.5 ± 18.5	125.8 ± 18.0	< 0.001
Hypertension, N (%)	300 (49.8)	219 (36.4)	< 0.001
Diabetes, N (%)	232 (38.5)	139 (23.1)	< 0.001
CHD, N (%)	44 (7.3)	30 (5.0)	0.09
eGFR, mL/min/1.73 m ²	93.7 ± 22.8	97.2 ± 25.2	0.01
Albuminuria, N (%)	126 (20.9)	77 (12.8)	< 0.001
Albumin/creatinine ratio, mg/g	8.6 (5.2–21.6)	6.7 (4.4–13.1)	< 0.001
Phosphate, mg/dL	3.3 ± 0.5	3.3 ± 0.5	0.4
CRP, (mg/L)	3.4 (1.8–6.9)	2.0 (0.9–4.8)	< 0.001
C-terminal FGF23, RU/mL	66.2 (52.8–98.4)	61.2 (49.5–80.1)	< 0.001
Intact FGF23, pg/mL	53.2 (42.8–67.7)	53.2 (42.8–71.2)	0.8
Apnea/hypopnea index	45.8 (35.5–62.5)	2.6 (0.6–8.2)	< 0.001

Results are reported as means ± standard deviation, proportions or medians with interquartile ranges.

Abbreviations: N, number; SDB, sleep disordered breathing; BMI, body mass index; SBP, systolic blood pressure; CHD, cardiovascular heart disease; eGFR, estimated glomerular filtration rate; CRP, C-reactive protein FGF23, fibroblast growth factor 23.

Quartile 1 (cFGF23 Quartile 4 vs. Quartile 1 Odds Ratio [OR] 2.02, CI 1.46–2.80 **Table 2**). After adjustment for demographics, CKD-associated risk factors, comorbidities, and CRP, individuals with severe sleep disordered breathing continued to have higher odds of belonging to cFGF23 Quartile 4 versus Quartile 1 (cFGF23 Quartile 4 vs. Quartile 1 OR 1.93, CI 1.31–2.86 **Table 2**). After adjustment for body mass index, the association was attenuated but remained significant (cFGF23 Quartile 4 vs. Quartile 1 OR 1.70, CI 1.12–2.59, **Table 2**). We did not appreciate a significant relationship between severe sleep disordered

Table 2

Adjusted associations of severe sleep disordered breathing presence with cFGF23 expressed as quartiles.

		cFGF23 quartiles (RU/mL)			
AHI ≥ 30 vs. AHI < 30	Total N	Quartile 1 < 50.5 N = 302	Quartile 2 50.5–63.15 N = 300	Quartile 3 63.16–87.95 N = 301	Quartile 4 > 87.96 N = 301
Unadjusted	1204	REF	1.22 (0.89–1.69)	1.32 (0.95–1.81)	2.02 (1.46–2.80)
Model 1	1204	REF	1.21 (0.87–1.68)	1.29 (0.93–1.79)	1.98 (1.41–2.77)
Model 2	1203	REF	1.27 (0.91–1.77)	1.43 (1.01–2.01)	2.23 (1.53–3.24)
Model 3	1200	REF	1.24 (0.89–1.74)	1.37 (0.96–1.93)	2.10 (1.43–3.08)
Model 4	1200	REF	1.19 (0.84–1.69)	1.41 (0.99–2.02)	1.93 (1.31–2.86)
Model 5	1197	REF	1.08 (0.75–1.57)	1.39 (0.95–2.03)	1.70 (1.12–2.59)

Model 1 adjust for age and gender.

Model 2 adjusts for covariates in Model 1 plus plasma phosphate, eGFR and UACR.

Model 3 adjusts for covariates in Model 2 plus hypertension, diabetes presence, cardiovascular disease, cigarette use.

Model 4 adjusts for covariates in Model 3 plus C-reactive protein.

Model 5 adjusts for covariates in Model 4 plus body mass index.

Abbreviations: FGF23, fibroblast growth factor 23; N, number; eGFR, estimated glomerular filtration rate; UACR, urinary albumin-to-creatinine ratio.

breathing and iFGF23 Quartiles (iFGF23 Quartile 4 vs. Quartile 1 OR 0.76, CI 0.52–1.11).

In the unadjusted linear regression analyses, severe sleep disordered breathing was associated with higher cFGF23 levels expressed as a continuous variable (β -estimate 0.12, CI 0.04–0.19, p value 0.002, Table 3). The relationship remained significant after adjustment for demographics, CKD risk factors, comorbidities, and CRP (β estimate 0.08, CI 0.02–0.15, p value 0.02). However, the association became non-significant after the addition of body mass index (β estimate 0.07, CI –0.005–0.14, p value 0.07, Table 3). We did not identify a significant relationship between severe sleep disordered breathing and iFGF23 levels (Table 3).

3.2. Subgroup analyses by CKD status

Of the 1204 individuals in the total population, 106 met criteria for CKD. Clinical characteristics of individuals by severe sleep disordered breathing status in individuals with and without CKD are presented in Supplemental Tables 1 and 2. The mean eGFR in those with CKD and

severe sleep disordered breathing was 46.3 ± 13.3 mL/min/1.73 m² and in those with CKD without severe sleep disordered breathing was 45.0 ± 13.5 mL/min/1.73 m² (Supplemental Table 1). Individuals with CKD and severe sleep disordered breathing had significantly higher body mass index and CRP than those with CKD but without severe sleep disordered breathing (body mass index 35.4 ± 6.7 kg/m² vs. 30.0 ± 6.8 kg/m², p value < 0.001; CRP 6.1 mg/L IQR 2.7–11.6 vs. 3.0 mg/L IQR 1.4–5.8, p value 0.001, Supplemental Table 1). cFGF23 and iFGF23 levels were also higher in those with severe sleep disordered breathing than in those without severe sleep disordered breathing, however this did not reach significance (Supplemental Table 1). In multivariable analyses that were restricted to individuals with CKD, severe sleep disordered breathing was not associated with higher cFGF23 levels or iFGF23 levels (Supplemental Table 3).

3.3. Sensitivity analyses

Because waist circumference may be a more accurate measurement of adiposity compared to BMI, we replaced body mass index with waist circumference in our multinomial regression and linear regression models. Our results were qualitatively similar. In our adjusted multinomial regression analyses, the association between severe sleep disordered breathing and cFGF23 quartiles remained significant but was attenuated after adjustment for waist circumference (cFGF23 Quartile 4 vs. Quartile 1 OR 1.62, CI 1.07–2.46). In our linear regression analyses investigating the associations between severe sleep disordered breathing and higher cFGF23 levels, similar to our models with body mass index, the association became non-significant after the addition of waist circumference (β estimate 0.06, CI –0.01–0.13, p value 0.11).

4. Discussion

In a sample drawn from a large cohort of Hispanic/Latino patients with and without CKD, we demonstrated that individuals with severe sleep disordered breathing had slightly higher levels of cFGF23 than those without severe sleep disordered breathing, but no difference in iFGF23 levels. Body mass index or waist circumference were the main confounders that attenuated the sleep disordered breathing-cFGF23 relationship in both linear and multinomial logistic regression analyses. We did not appreciate any relationship with severe sleep disordered breathing presence and iFGF23 in the total population or in the subgroup with CKD. These results are broadly consistent with our hypotheses and suggest that in addition to iron deficiency and inflammation, sleep disordered breathing may be another clinical entity in which we see upregulation of FGF23 transcription in osteocytes and an elevation in cFGF23 but not iFGF23 levels.

In vivo and in vitro studies demonstrate that hypoxia regulates

Table 3

Adjusted associations of severe sleep disordered breathing with cFGF23 and iFGF23 levels expressed as continuous variables.

		c-terminal FGF23			iFGF23		
AHI ≥ 30 vs. AHI < 30	Total N	Parameter Estimate	95% CI	p-Value	Parameter estimate	95% CI	p-Value
Unadjusted	1204	0.12	0.04–0.19	0.002	–0.02	–0.08–0.03	0.4
Model 1	1204	0.10	0.03–0.17	0.006	–0.03	–0.09–0.02	0.3
Model 2	1203	0.09	0.03–0.16	0.006	–0.03	–0.09–0.02	0.2
Model 3	1200	0.08	0.02–0.15	0.01	–0.03	–0.08–0.02	0.2
Model 4	1200	0.08	0.02–0.15	0.02	–0.03	–0.08–0.03	0.4
Model 5	1197	0.07	–0.005–0.14	0.07	–0.03	–0.09–0.03	0.3

Model 1 adjust for age and gender.

Model 2 adjusts for covariates in Model 1 plus plasma phosphate, eGFR and UACR.

Model 3 adjusts for covariates in Model 2 plus hypertension, diabetes presence, cardiovascular disease, cigarette use.

Model 4 adjusts for covariates in Model 3 plus C-reactive protein.

Model 5 adjusts for covariates in Model 4 plus body mass index.

Abbreviations: FGF23, fibroblast growth factor 23; N, number; CI, confidence interval; eGFR, estimated glomerular filtration rate; UACR, urinary albumin-to-creatinine ratio.

FGF23 transcription [16]. When UMR-106 cells are grown in an oxygen deprived environment, expression of FGF23 mRNA increases. This parallels increases in HIF-1 α protein expression, making it plausible that the appreciable increase in FGF23 mRNA expression is in response to cell hypoxia [16]. In vivo models support these findings. When animals are exposed to a hypobaric atmosphere, cFGF23 increases by six fold, but iFGF23 remains unchanged [16]. This indicates that hypoxic environments may upregulate FGF23 transcription, which is represented by an increase in cFGF23, along with a commensurate increase in FGF23 cleavage resulting in normal bioactive iFGF23 levels [35]. Observational human studies also support a hypoxia-FGF23 relationship. A single center study of 32 pediatric patients demonstrated that preoperative oxygen saturation level negatively correlated with cFGF23 levels, suggesting that hypoxemia may also regulate FGF23 transcription in humans [36]. We now demonstrate that individuals with severe sleep disordered breathing are more likely to have elevated cFGF23 levels, but not iFGF23 levels.

Severe sleep disordered breathing is characterized by multiple overnight respiratory events associated with oxygen desaturation [37]. Prior reports of experiments conducted in cell cultures and animal models demonstrate that episodic hypoxia can directly impact the bone by increasing expression of HIF-1 α and stimulating FGF23 transcription [38, 39]. Based on these data, we propose that intermittent nocturnal hypoxemia in sleep disordered breathing may directly trigger FGF23 transcription in the bone resulting in elevated cFGF23 levels. We further speculate that increases in FGF23 transcription induced by nocturnal hypoxemia in the absence of CKD is coupled to parallel increases in FGF23 cleavage, resulting in normal iFGF23 levels.

There are additional possible explanations for our findings. The elevation of cFGF23 in the severe sleep disordered breathing group may be a result of coexisting risk factors, such as elevated body mass index, inflammation, and hematologic indices in individuals with severe sleep disordered breathing. Prior investigations led by our group and others demonstrated significant associations between elevated body mass index and elevated FGF23 levels [40–45]. Indeed, adjustment for body mass index or waist circumference attenuated our results in both our multinomial logistic regression and linear regression analyses, and these covariates were the major confounders that rendered the relationship in the linear regression models non-significant. The mechanisms underlying the associations of elevated body mass index and FGF23 levels remain unknown, but an overlap between metabolic and inflammatory signals is known to exist, which may contribute to an inflammatory milieu in obesity [46]. Both acute and chronic inflammation are known stimuli of FGF23 transcription [47]. In all comparisons, including our subgroups of CKD and non-CKD, those with severe sleep disordered breathing had significantly higher CRP values than those without severe sleep disordered breathing. Although we adjusted for CRP in our models, inflammation remains a biologically plausible mechanism that may link sleep disordered breathing and elevated body mass index with FGF23 elevation.

Prior investigations also demonstrate that erythropoietin levels may be elevated in individuals with sleep disordered breathing [48] and data suggest that erythropoietin may directly regulate FGF23 production [16]. In addition, iron deficiency upregulates FGF23 transcription and may also be related to sleep related diseases [47, 49]. Although iron stores and erythropoietin are not available for our analyses, disturbances in hematologic parameters represent another mechanism by which cFGF23 levels may be elevated in severe sleep disordered breathing. Finally, prior investigations in humans demonstrated a relationship between obstructive sleep apnea and aberrant mineral metabolism [50–52]. By triggering alterations in the mineral metabolism axis, intermittent hypoxia may indirectly lead to FGF23 elevation through alterations in vitamin D and parathyroid hormone.

We acknowledge additional limitations in our study. Sleep disordered breathing exemplifies a good model of intermittent hypoxemia, but differs from models of chronic hypoxemia. Each model of

hypoxemia associates with different biological responses [53], and the relationship of each of these to FGF23 homeostasis is unknown. We demonstrate small and likely not clinically meaningful elevations in cFGF23 levels in individuals with intermittent hypoxia. Whether chronic hypoxia models would demonstrate larger differences in cFGF23 levels is unknown. Human models of chronic hypoxia however, also have limitations. For example, clinical scenarios of severe lung disease are characterized as high inflammatory states, and a major etiology of lung disease includes smoking, another known risk factor associated with elevated FGF23 levels [54]. Due to the small sample size of the CKD subgroup, we could not elucidate the impact of severe sleep disordered breathing on FGF23 transcription or cleavage in CKD. Although we were able to adjust for important confounders such as body mass index and CRP in our cross-sectional analyses, we were unable to adjust for iron indices and do not have longitudinal FGF23 measurements available. Finally, our study only included Hispanic/Latino populations. Given that prior studies have demonstrated racial variation in FGF23 levels [55], the generalizability of our findings may be limited.

In settings of increased FGF23 transcription but normal FGF23 cleavage, which result in isolated cFGF23 elevation, we do not yet understand what role c-terminal FGF23 fragments have given that the biologic activity and maladaptive cardiovascular effects of FGF23 are believed to be mediated by elevation in iFGF23 levels. However, elevated levels of cFGF23 and iFGF23 both demonstrate associations with morbidity and mortality in the general population and in those with CKD [7, 9, 10]. It is critical to continue investigations that help elucidate factors that regulate FGF23 production and cleavage. The current findings demonstrate that, similar to iron deficiency and inflammation, severe sleep disordered breathing may be another clinical state characterized by cFGF23 elevation in the absence of iFGF23 elevation. Further human studies in alternative models of hypoxemia and in larger CKD populations are needed to confirm our findings and ascertain the impact of CKD on cFGF23 and iFGF23 levels in sleep disordered breathing.

Conflict of interest statement

All authors have read the journal's policy on disclosure of potential conflicts of interest. RM has interest in Abbot Laboratories, AbbVie, Inc. and Teva Pharmaceuticals Industries Ltd. TI has received honoraria from Bayer and grant support from Shire. MW has received research support, honoraria or consultant fees from Amgen, Ardelyx, DiaSorin, Keryx, Lilly, Pfizer, Shire and Ultragenyx.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.bone.2018.06.024>.

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