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Original article

Association between ratio of fibroblast growth factor 23 (FGF-23) to Klotho and phosphate level in chronic kidney disease patient

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Abstract. *Background and aim:* Chronic Kidney Disease (CKD) is a condition in which there is structural and functional damage to the kidneys that lasts more than 3 months, and when there is a decrease in glomerular filtration rate (GFR), phosphate homeostasis is disrupted. FGF-23 acts as a phosphaturic hormone that increases phosphate excretion when there is an increase in serum phosphate levels. The physiological effects of FGF-23 can occur when it binds to Klotho as a co-receptor, where Klotho is mainly produced in the kidneys. Decreased kidney function triggers a decrease in kidney mass, so that Klotho production will decrease. This study aimed to determine the association between the ratio of FGF-23 to Klotho and phosphate levels in patients with CKD. *Methods:* This was an observational study involving 60 patients with stage 3, 4, and 5 non-dialysis CKD. Serum of FGF-23, Klotho, phosphate, and creatinine were measured using the ELISA method. Statistical analysis was performed using Mann-Whitney, Chi-Square, and Spearman Correlation Tests. *Results:* The median value of FGF23-Klotho ratio was 0.17. A significant correlation was found between the FGF23-Klotho ratio and phosphate levels (P<0.001, r= 0.581). There was a significant relationship between the female sex and the incidence of hyperphosphatemia (P=0.0038). *Conclusion:* The higher the FGF23-Klotho ratio, the higher the phosphate levels in CKD patients and it can be a risk factor, protective factor, and for diagnostic, therapeutic and prognostic purposes in assessing phosphate levels and hyperphosphatemia conditions in CKD patients. (www.actabiomedica.it)

Key words: FGF-23, Klotho, FGF23-Klotho ratio, phosphate, Chronic Kidney Disease

Introduction

Chronic Kidney Disease (CKD) is a condition where there is structural or functional damage to the kidneys that lasts ≥ 3 months and has implications for health. The progression of CKD is categorized into five stages based on the decrease in glomerular filtration rate (GFR). These stages are: stage 1 (GFR > 90 ml/minute/1.73m^2), stage 2 (GFR 60-89 ml/minute/1.73m^2), stage 3 (GFR 30-59 ml/minute/1.73m^2), stage 4 (GFR 15-29 ml/minute/1.73m^2), and stage 5 (GFR < 15 ml/minute/1.73m^2) (1).

When GFR decreases, it disrupts phosphate homeostasis. As kidney function decreases, phosphate excretion will decrease, which in turn causes an increase in phosphate levels in the blood. However, in reality, during mild decline in kidney function (stages 2 and 3), phosphate levels remain within normal limits. This is caused by the body's compensation mechanism where there will be an increase in a hormone called phosphotonin such as Fibroblast Growth Factor-23 (FGF-23) (2).

Fibroblast Growth Factor-23 is synthesized by bone cells, specifically osteoclasts, with a smaller

contribution from osteoblasts. FGF-23 is also called a phosphaturic hormone because of its function in increasing phosphate excretion when there is an increase in phosphate levels, to maintain phosphate levels within the normal range. The phosphaturic effect of FGF-23 is mediated by reducing the expression of NaPi-IIa and NaPi-IIc co-transporters in the kidney. This leads to a decrease in phosphate reabsorption in the tubules, resulting in increased phosphate excretion. FGF-23 also decreases the expression of the NaPi-IIb co-transporter in the small intestine, leading to reduced phosphate absorption in this organ (3).

Various studies (4-7) has reported that there was an increase in phosphate levels as GFR decreased. Hyperphosphatemia conditions will cause vascular and heart valve calcification which will increase cardiovascular (CV) morbidity and mortality (8). Also, hyperphosphatemia conditions together with calcium metabolism disorders, decreased vitamin D synthesis, increased PTH will cause chronic kidney disease - mineral and bone disorder (CKD-MBD) (9).

The regulation of phosphate serum is influenced by various factors that are not yet fully understood. Diet, gender, age, and genetics are known to influence phosphate serum levels (10). Additionally, FGF-23 levels increase with old age, obesity, hypertension, and diabetes mellitus (DM) (11-12).

The physiological effects of FGF-23 occur when this hormone binds to Klotho as a co-receptor. Klotho is primarily produced in the kidneys. Therefore, when kidney function decreases, resulting in a decrease in kidney mass, Klotho production also decreases. Additionally, the levels of Klotho levels is also influenced by oxidative stress and inflammation that occur in CKD. Klotho as a co-receptor for FGF-23 is in the form of membranous Klotho (mKlotho) which is found in the membranes of renal tubule cells which plays a role in phosphate homeostasis. Klotho is also found in the form of soluble Klotho (sKlotho) which circulates in the circulation and is independent of FGF-23 and acts as a cytoprotector, anti-fibrosis, anti-inflammatory and angiogenesis (5,13). Decreased kidney function will result in a decrease in phosphate excretion. The body will respond by increasing the synthesis and secretion of FGF-23 from bones whose function is to increase phosphate excretion (4,6). In stage 3B CKD, phosphate concentrations begin to increase, indicating that compensatory mechanisms are no longer sufficient to maintain phosphate balance and prevent hyperphosphatemia (7).

It is known that in CKD there is an increase in FGF-23 levels and a decrease in Klotho levels, in other words in CKD there is an increase in the ratio of FGF-23 to Klotho. Changes in this ratio can be one of the parameters of the severity of CKD progression and reflect the high and low blood phosphate levels of CKD patients (5). This study aimed to analyze the association between the ratio of FGF-23 to Klotho and phosphate levels in CKD patients.

Material and methods

Ethics committee approval

This analytic observational study using a cross-sectional design was approved by the Ethics Committee of Biomedical Research on Humans, Faculty of Medicine, Hasanuddin University, Makassar, South Sulawesi, Indonesia. Based on recommendation letter Number: 597/UN4.6.4.5.31/PP36/2023, August 28, 2023 and duration of the study approval from 28 August 2023 to 28 August 2024 with protocol number: UH23080580.

Patient population

The population of this study were stage 3,4 and non-dialysis stage 5 CKD patients who received outpatient or inpatient treatment at Wahidin Sudirohusodo Hospital, Makassar, South Sulawesi, Indonesia which is a tertiary referral center starting from September 01st to October 15th, 2023. The research sample was selected from a population that met the criteria for the study. The study included a sample size of 60 subjects who met the specified criteria.

Inclusion and exclusion criteria

The inclusion criteria for this study were diagnosed with chronic kidney disease stages 3-5, aged 18-65 years, not currently taking phosphate, vitamin D, and calcium binding drugs, and willing to participate

in the research by signing an informed consent form. The exclusion criteria were CKD patients undergoing hemodialysis.

Clinical data and sample collection

Demographic and clinical data extracted taken from patient medical records. Sampling for the study of FGF-23, Klotho, and phosphate examination was performed when the patient was diagnosed with stage 3,4, and 5 non-dialysis CKD and met the study criteria and was willing to become a research sample by signing an informed consent form. Sample testing was performed at Prodia Widyahusada Research Laboratory Unit. This study used FGF-23, Klotho and phosphate ELISA kits (Immutopics). Serum was measured using the ELISA method with a Thermo Scientific Multiskan FC microplate spectrophotometer with units of measurement FGF23 (rU/ml), Klotho (pg/ml), and phosphate (mg/dl). The FGF23-Klotho ratio is obtained by dividing the FGF-23 value by the Klotho value. We also classify patients based on phosphate levels into normophosphatemia, and hyperphosphatemia with a cutoff value of 4.5 mg/dl.

Statistic analysis

The data analysis was conducted using SPSS version 26. The method of analysis consisted of a descriptive method that aimed to describe the characteristics of the research sample by calculating the mean, standard deviation, median, minimum, and maximum values. Kolmogorov-Smirnov test to assess data normality, and Mann-Whitney, Chi-Square, and Spearman's correlation test is a statistical analysis for data that is not normally distributed. Statistical test results are significant if the P-value <0.05.

Results

Study population

In this study, the sample size consisted of 60 subjects, with 31 males (51.7%) and 29 females (48.3%). The subjects had an age range of 18-65 years (48.27 ± 12.62 years), and 22 subjects (29.7%) were found to have hyperphosphatemia. The characteristics of the research variables are shown in Table 1. In this study, the FGF-23 level range was 8.40 - 2,013.20 rU/ml with a median value of 100 rU/ml. Klotho levels ranged from 214.50 - 6981.50 pg/ml, with a median of 571 pg/ml. The ratio of FGF-23 levels to Klotho levels ranges from 0.02 – 7.23 with a median value of 0.17. Analysis of the characteristics of these subjects can be seen in Table 2. In this study, stages of CKD stratified in stages 3, 4 and 5 non-dialysis with research variables such as FGF-23 levels, Klotho levels, FGF-23 to Klotho ratio, and the incidence of hyperphosphatemia. These variables were presented in mean, median, frequency and percentage. The stratification results showed that there was an increase in FGF-23 levels, the ratio of FGF-23 to Klotho,

Table 1. Characteristics of research subjects.

Variable	N	%
Gender		
Male	31	51,7
Female	29	48,3
Age		
≥ 60 years	14	23,3
< 60 years	46	76,7
BMI		
Obesity	16	26,7
Non-obesity	44	73,3
Phosphate Level		
Hyperphosphatemia	22	29,7
Normophosphatemia	38	51,3
CKD		
Stage 3	20	33,3
Stage 4	20	33,3
Stage 5 (non-dialysis)	20	33,3

Abbreviations: BMI: body mass index; CKD: chronic kidney disease.

Table 2. Characteristics of study population.

Variables	Results (N=60) Median (min - max)
Phosphate (mg/dl)	4.15 (2.20 – 13.50)
eGFR (ml/minute per 1,73m2)	24.60 (1.10 – 56.30)
FGF-23 (rU/ml)	100 (8.40 - 2.013.20)
Klotho (pg/ml)	571 (214.50 – 6981.50)
FGF23-Klotho Ratio (rU/pg)	0.17 (0.02 – 7.03)

Abbreviations: eGFR: glomerular filtration rate; FGF-23: fibroblast growth factor 23.

Table 3. Stratification of CKD stage with research variable.

CKD stage	(N)	FGF- 23 Median (min-max)	Klotho Median (min-max)	FGF23 –Klotho Ratio Median (min-max)	Hyperphosphatemia N (%)
3	20	61.70 (12.30-1223)	628.50 (275.60-1329.10)	0.1 (0.02-3.11)	3 (15%)
4	20	80.35 (8.40-710)	586.80 (406.10-6981.50)	0.13 (0.02-0.91)	4 (20%)
5	20	298.10 (80.40–2013.20)	538.50 (214.50-956.60)	0.66 (0.17-7.23)	15 (75%)

Table 4. Association between ratio of FGF23-Klotho and phosphate levels.

Ratio of FGF23-Klotho	Phosphate Median (mg/dl)	Min-max	P	Ratio of FGF23-Klotho	Normophosphatemia (n)	Hyperphosphatemia (n)	P
> 0,17	4,8	2,7-13,5	< 0,001*	> 0,17	12	17	<0,001**
≤ 0,17	3,9	2,2-7,9		≤ 0,17	26	5	

Note: **Mann-Whitney Test; **Chi-Square Test.*

and the percentage of hyperphosphatemia incidents as the CKD stage increased or kidney function decreased, whereas there was a decrease in Klotho levels as the CKD stage increased or kidney function decreased (Table 3).

Association between ratio of FGF23-Klotho and phosphate level

In Table 4, the median value of the FGF23-Klotho ratio is 0.17 so it is divided into 2 groups, namely the group with values above the median (> 0.17) and below or equal the median (≤ 0.17). In the above the median group, the median phosphate was 4.8 mg/dl, 12 subjects were normophosphatemia, and 17 subjects were hyperphosphatemia. Meanwhile, in the below or equal the median group, the median phosphate was 3.9 mg/dl, 26 subjects were normophosphatemia, and 5 subjects were hyperphosphatemia. These results are statistically significant with P: < 0.001 and < 0.001, respectively.

Correlation between ratio of FGF23-Klotho and phosphate level

Correlation test was carried out between the FGF23-Klotho ratio and phosphate levels. In the Spearman correlation test, a significant positive correlation was found with P: < 0.001. These results show that the higher the FGF23-Klotho ratio value, the higher the phosphate level with a correlation coefficient of 0.581 (Figure 1).

A correlation test was carried out between FGF23-Klotho ratio and phosphate levels by stratifying them based on CKD stage (stages 3, 4 and 5). In CKD stages 3 and 5, the Spearman correlation test found a significant positive correlation with P: 0.028 and < 0.001 respectively. These results show that the higher the FGF23-Klotho ratio value, the higher the phosphate levels with correlation coefficients of 0.49 and 0.73 respectively. In CKD stage 4, the Spearman correlation test did not find a significant positive correlation with P: 0.142 (Figure 2).

Association of confounding variable and phosphate level

In this study, the association between the confounding variables (age, gender, BMI) and phosphate levels was also assessed where a significant association was found between female gender and the incidence of hyperphosphatemia (P 0.0038) (Table 5).

Discussion

In this study, the median value of FGF23-Klotho ratio was 0.17. It was found that median

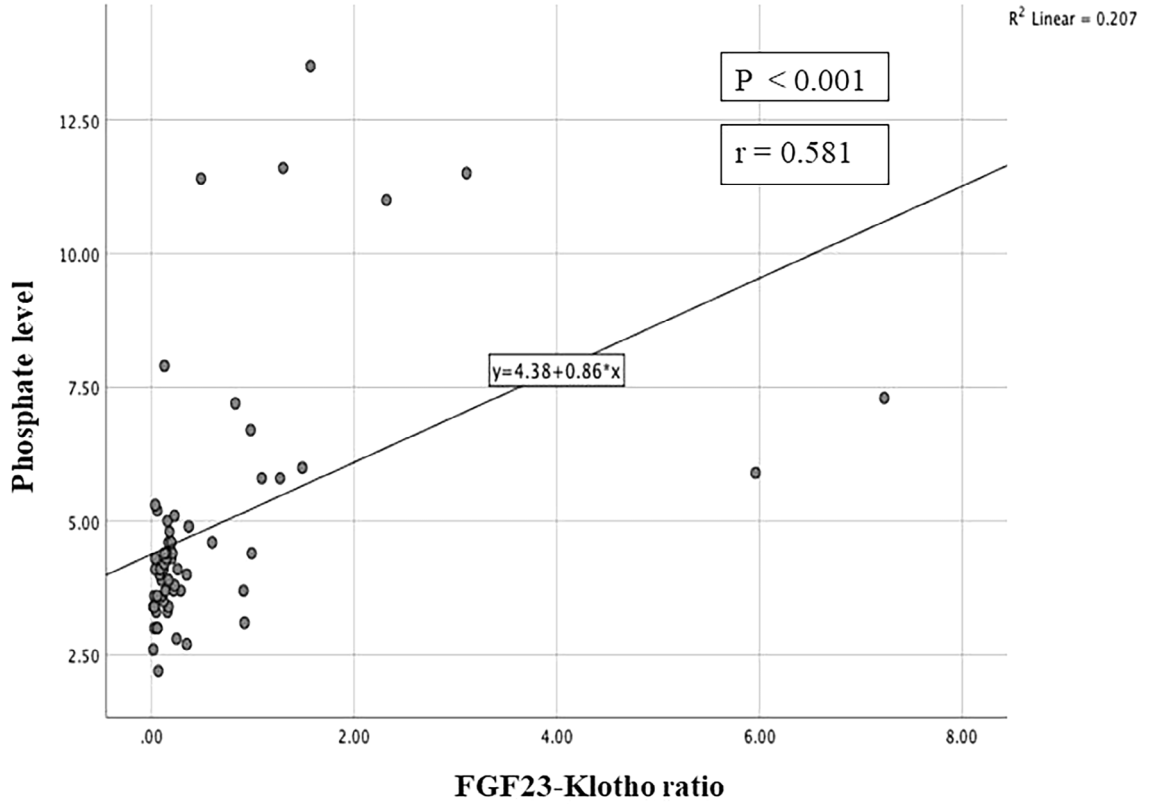

Figure 1. Correlation between ratio of FGF23-Klotho and phosphate level (Spearman's correlation test).

phosphate was higher in the above the median group (> 0.17) compared to the below or equal median group (≤ 0.17), namely 4.8 mg/dl and 3.9 mg/dl respectively (p<0.001). In addition, a greater proportion of normophosphatemia was found in the below or equal the median group (p<0.001). In this study, it was also found that as the CKD stage increases, the median of FGF-23, the FGF23-Klotho ratio, and the proportion of hyperphosphatemia will increase, while the median Klotho will decrease.

In line with this study, a study by Liu Z et al. (5) involving 152 patients showed that in CKD stages 3-5, creatinine, iFGF23 levels, and cFGF23-Klotho ratio were higher (P: < 0.01), higher phosphate levels (P: < 0.05), and lower Klotho levels (P: <0.01) compared to controls. cFGF23 levels were higher in CKD stages 4-5 (P: < 0.01). In CKD stage 4-5, creatinine, iFGF23, phosphate levels, and the cFGF23-Klotho ratio were higher (P: < 0.01), cFGF23 levels were higher (P: < 0.05), and Klotho levels were lower (P: <0.05) compared with CKD stage 3. Phosphate levels were positively correlated with the cFGF23-Klotho ratio (r=0.235, P: < 0.01).

A recent study by Rotondi et al. (14) supports the finding of this study, by showing that increment of FGF23-Klotho ratio was correlated with increment of CKD stage and phosphate level. This study in 68 CKD subjects, found an increase in FGF23, presentation of hyperphosphatemia, a decrease in klotho levels as the stage of CKD increased or kidney function decreased.

Phosphate levels in CKD patients are mainly regulated by diet, FGF-23, Klotho, PTH, and 1,25(OH)2D3. FGF-23 in blood consists of three forms: intact FGF-23 (iFGF-23), amino-terminal peptide segment (nFGF-23), and carboxyl-terminal peptide segment (cFGF-23). As CKD progresses, increasing phosphate levels stimulate the secretion of iFGF-23, which induces urinary phosphate excretion by binding to the FGF-23 receptor (FGFR)–Klotho complex thereby regulating phosphate metabolism by inhibiting 1,25(OH)2D3 synthesis and PTH secretion. However, cFGF-23 competitively binds to

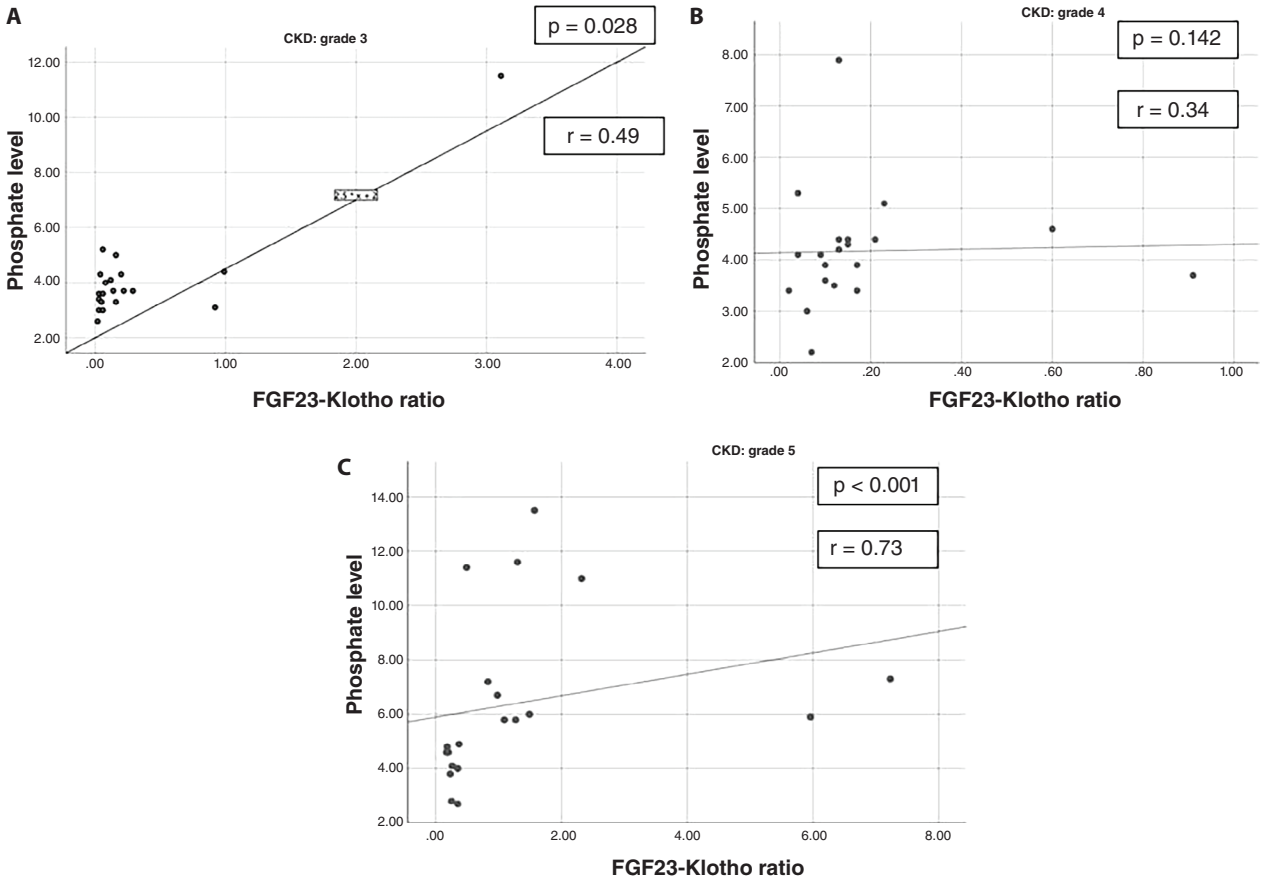

Figure 2. Correlation between ratio of FGF23-Klotho and phosphate level (a. stage 3, b. stage 4, c. stage 5) (Spearman's correlation test).

Table 5. Distribution of phosphate level according to age, gender and BMI, and statistical analysis using Chi-Square.

Variable			Phosphate level		p
			Hyperphosphatemia	Normophosphatemia	
Age	≥60 year	n	3	11	0.177
		%	21,40	78,57	
	<60 year	N	19	27	
		%	41,30	58,69	
Gender	Male	N	7	24	**0.0038**
		%	22,58	77,42	
	Female	N	15	14	
		%	51,72	48,27	
BMI	Obesity	N	6	10	0.936
		%	37,50	62,50	
	Non-obesity	N	16	28	
		%	36,36	63,63	

the FGFR–Klotho binary complex to inhibit the iFGF23–Klotho signaling axis and urinary phosphate excretion. There are no reports regarding the effect of cFGF-23 on phosphate metabolism of CKD patients. As CKD progresses, iFGF-23 and cFGF-23 levels increase; iFGF-23 and cFGF-23 regulate phosphate metabolism via Klotho (5).

This study found a significant positive correlation between FGF23-Klotho ratio and phosphate levels (P: < 0.001). It shows that the higher the FGF23-Klotho ratio value, the higher the phosphate level with a correlation coefficient of 0.581. After stratification based on CKD stage, a significant positive correlation was found in CKD stages 3 and 5 (P=0.028, P:<0.001 respectively). Meanwhile, no significant positive correlation was found in CKD stage 4 (P=0.142).

The possibility of not finding a correlation between the FGF23-Klotho ratio and phosphate level in CKD stage 4 in our study is due to the lack of samples representing CKD stage 4, or patients having a process of improvement in CKD, or compensatory mechanisms for hyperphosphatemia conditions are still occurring, and or several factors such as nutrition that can improve kidney function.

These results reflect those of Caravaca et al. (15) who also found linear correlation between the level of change in eGFR and mean serum phosphate levels, where high serum phosphate levels were strongly and independently associated with a greater decline in kidney function in 184 subjects with stages 3, 4 and 5 non-dialysis CKD.

In line with this study, a study by Kritmetapak et al. (4), involving 85 subjects with various eGFR, showed a progressive increase in serum concentrations of iFGF23, PTH, phosphate and a decrease in serum 1,25(OH)2D concentrations as eGFR decreased. At eGFR ≥ 60 mL/min/1.73 m^2 phosphate levels are 3.3 ± 0.4 mg/dl; In CKD stage 3A the phosphate level is 3.3 ± 0.7 mg/dl; In CKD stage 3B the phosphate level is 3.5 ± 0.6 mg/dl; In CKD stage 4 the phosphate level is 4.0 ± 0.6 mg/dl and in CKD stage 5 the phosphate level is 4.7 ± 0.3 mg/dl.

In accordance with the present results, previous studies by Canziani et al. (16) research on 96 CKD subjects found association between FGF-23 and hyperphosphatemia (P=0.034) with OR 1.01 (95% CI 1.001-1.022). Research conducted by Faul et al. (17) with a cohort sample of 3,070 CKD subjects, showed an association between FGF-23 and hyperphosphatemia (P: <0.001) with an OR of 1.5 (95% CI 1.3-1.9). Research by Mirza et al. (18) on the correlation between FGF-23 and hyperphosphatemia in 795 CKD subjects showed that increased serum FGF-23 was associated with increased phosphate levels in the elderly with OR 1.28 (95% CI 1.09–1.51). Shibata et al. (19) research on 70 CKD subjects showed that increased FGF-23 levels were positively correlated with increased phosphate levels with ORs of 13.46 and 2.87, respectively. One of the issues that emerges from these finding is none of these studies used FGF23-Klotho ratio parameter and only used CKD stage (GFR), FGF-23, and phosphate parameter separately. Our study is one of a limited number of studies that consider this ratio.

Gutierrez et al. (6) divided subjects into 4 groups: (1) GFR > 60, (2) 45-60, (3) 30-45, (4) < 30 ml/minute. Obtained FGF-23 levels were 86.2 ± 61.4, 136.2 ± 69.1, 224.6 ± 200.1 and 436.0 ± 493.8 rU/ml, respectively. Marsell et al.'s research on 3016 subjects aged 70-80 years with GFR > 60 ml/minute/1.73m^2, found a negative relationship between FGF-23 levels and GFR (20). Fliser et al. (21) studied 227 non-diabetic CKD subjects who were divided into 4 groups, namely (1) GFR > 90, (2) 60-89, (3) 30-59, (4) < 30 ml/minute. Obtained FGF23 levels were 57 ± 43, 81 ± 52, 187 ± 194 and 456 ± 475 rU/ml, respectively.

On the other hand, a decrease in GFR caused by reduced kidney mass will be followed by a decrease in Klotho levels because Klotho mRNA expression is highest in the renal tubules (13). Pavik et al. (22) found that every decrease of 1 ml/min/1.73m^2 GFR would be followed by a decrease of 3.2 pg/mL Klotho. Kim et al. (23) studied 243 CKD patients, finding that serum Klotho levels were positively correlated with decreased kidney function (P: < 0.001). Shimamura et al. (24) studied 292 CKD patients, finding that Klotho was positively correlated with GFR (r = 0.441; P: <0.0001) and negatively correlated with serum creatinine levels (r = 0.181; P: <0.001). Seiler et al. (25) studied prospectively 321 CKD patients, whose Klotho levels were at stage 2, 3A, 3B, and 4 were 554 (472-757) pg/mL, 550 (462-665) pg/mL, 536 (443-661) pg/mL, and 530 (429-678) pg/mL respectively.

Due to the decrease in GFR, FGF-23 levels increase accompanied by decreased Klotho levels, this will cause an increase in the ratio of FGF-23 levels to Klotho levels. One of the issues that emerges from these finding is none of these studies used FGF23-Klotho ratio parameter and only used CKD stage (GFR), FGF-23, and Klotho parameter separately.

In the early stages of CKD, phosphate metabolism is already impaired, but serum phosphate levels are usually maintained within the normal range due to compensatory increases in FGF-23 and PTH until the final stages of kidney disease (26,27). In advanced stages of CKD (stage 3B), when GFR drops to <45 mL/minute/1.73 m^2, the kidneys are no longer able to adequately compensate for the phosphate load and an increase in serum phosphate levels occurs (28). As GFR decreases, the cFGF23-Klotho ratio increases and the inhibitory effect of cFGF23 on the iFGF23–Klotho signaling axis increases thereby reducing urinary phosphate excretion. It was found that the regulatory effect of iFGF-23 on phosphate was gradually reduced when the cFGF23-Klotho ratio was greater than 2.88. Maintaining the cFGF23-Klotho ratio in the appropriate range by increasing Klotho expression as well as inhibiting iFGF-23 cleavage can improve phosphate metabolism in CKD patients (5).

This outcome is contrary to that of Abdallah et al. (29) who did not find an association between Klotho and hyperphosphatemia (P=0.080) in 88 CKD subjects, Branislav et al. (30) study on 142 CKD patients, did not find association between Klotho levels and hyperphosphatemia (P=0.079), Tanaka et al. (31) from 234 CKD patients did not find association between Klotho levels and hyperphosphatemia (P=0.230). However, our results are consistent with data obtained in several studies that show correlation between Klotho levels and hyperphosphatemia. Semba et al. (32) conducting research on 1023 Italian, it was found that high Klotho levels were independently associated with low phosphate levels. Likewise, Seifert et al. (33) in 38 CKD patients who were followed for 1 year found that independently a decrease in Klotho levels was associated with an increase in hyperphosphatemia. Yang et al. (34) in a study involving 86 CKD patients and experimental animals concluded that increasing phosphate levels was negatively related to Klotho levels.

Klotho works through at least three mechanisms, namely: as a phosphaturic hormone, preventing a decrease in GFR and direct effects on soft tissue including vascular smooth muscle. Circulating Klotho increases nitric oxide synthesis in endothelial cells. In experimental animals that are deficient in Klotho, they show impaired vascular vasodilation and increased vascular calcification, while in conditions overexpressing Klotho, there is improvement in endothelial dysfunction, increased production of nitric oxide and reduced blood pressure (35,36).

There was a significant association between female gender and the incidence of hyperphosphatemia in CKD patients in our study (P=0.0038). In line with research by Wojcicki et al. (37), which involved 19,380 subjects without CKD, it showed that women were more likely to have high serum phosphate (17.66% versus 12.73%, P: <0.01, (OR 1.61, 95% CI 1.39-1.87)), the same condition was also found in younger participants (42.34 ± 0.49 versus 45.91 ± 0.65 years, P: < 0.01).

Research by Rabbani et al. (38), which involved 80 stage 5 CKD patients who had undergone dialysis for more than 6 months, found that hyperphosphatemia was positively correlated with female gender (OR 2.94, 95% CI 1.05-8.23).

Research by Bellasi et al. (39), involving 1,716 CKD stage 3-5 subjects, found that older age and male gender were associated with lower phosphate levels. Female gender is another risk factor known to contribute to hyperphosphatemia, although the mechanism is unclear. One study showed that estrogen has a direct effect on suppressing sodium-dependent phosphate absorption in the proximal tubule of the kidney which induces phosphaturia and reduces serum phosphate; post-menopausal women and estrogen deficiency increase the risk of hyperphosphatemia (37). In cases where bone resorption is accelerated, such as in menopause, phosphate enters the circulation along with calcium from the bones. In contrast, administration of estrogen to women at menopause causes a decrease in bone resorption, thereby suppressing the flux of calcium and phosphate into the circulation and ultimately reducing circulating phosphate levels (40). Thus the relationship between hyperphosphatemia and female sex may be the result of estrogen-mediated regulation

of renal phosphate reabsorption (38). A limitation of this study, we did not assess the phosphate intake of the study subjects.

Conclusion

A significant correlation was found between the FGF23-Klotho ratio and phosphate levels in CKD patients. As the FGF23-Klotho ratio increases, there will be a subsequent rise in both mean phosphate levels and the prevalence of hyperphosphatemia. As chronic kidney disease (CKD) progresses, there is a corresponding increase the mean FGF23, FGF23-Klotho ratio, proportion of hyperphosphatemia. Conversely, the mean level of Klotho decreases. Females with chronic kidney disease (CKD) tend to have higher phosphate levels.

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Conflict of Interest: Each author declares that he or she has no commercial associations (e.g. consultancies, stock ownership, equity interest, patent/licensing arrangement etc.) that might pose a conflict of interest in connection with the submitted article

Author's Contribution: IK (Concept, Design, Resources, Materials, Data Collection and Processing, Analysis and Interpretation, Literature Search, Writing Manuscript). HK (Concept, Design, Supervision, Analysis and Interpretation, Literature Search). SB (Concept, Design, Supervision, Analysis and Interpretation, Literature Search). AML (Concept, Design, Critical Review). HU (Concept, Design, Critical Review). AS (Concept, Design, Analysis and Interpretation, Critical Review).

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