Short-Term Effects of Very-Low-Phosphate and Low-Phosphate Diets on Fibroblast Growth Factor 23 in Hemodialysis Patients

A Randomized Crossover Trial

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Abstract

Background and objectives The short-term effects of low-phosphate diets on fibroblast growth factor 23 (FGF23) level and the optimal amount of dietary phosphate restriction in patients undergoing hemodialysis remain unknown.

Design setting, participants, & measurements This was a randomized, active-controlled trial with a crossover design that included 35 adults with ESKD undergoing thrice-weekly hemodialysis and with a serum phosphate level >5.5 mg/dl or between 3.5 and 5.5 mg/dl with regular phosphate binder use at a hemodialysis unit of tertiary teaching hospital in Taiwan. Subjects were randomized 1:1 to receive a very-low-phosphate diet, with a phosphate-to-protein ratio of 8 mg/g, or a low-phosphate diet, with a phosphate-to-protein ratio of 10 mg/g for 2 days, each with a 5-day washout during which subjects adhered to their usual diet. The primary outcome measure was mean difference in change-from-baseline intact FGF23 level between intervention groups. Secondary outcomes included difference in change-from-baseline serum phosphate, intact parathyroid hormone (PTH), and C-terminal FGF23 level between intervention groups.

Results There was no significant difference in the mean change-from-baseline in intact FGF23 levels between the two study diets. The very-low-phosphate diet significantly lowered serum phosphate (mean difference, 0.6 mg/dl; 95% confidence interval [95% CI], 0.2 to 1.0; P=0.002). There were no significant differences in change-from-baseline intact PTH and C-terminal FGF23 levels between the two study diets.

Conclusions Over the 2-day period, the FGF23-lowering effect of the very-low-phosphate diet is similar to that of the low-phosphate diet. The very-low-phosphate diet has an additional phosphate-lowering effect compared with the low-phosphate diet.

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Introduction

Dietary phosphate restriction is a recommended way to control serum phosphate levels (1). The effectiveness of nutritional counseling in treatment of hyperphosphatemia in patients with kidney disease is compromised by limited number of dietitians in clinical practice and lack of sustained beneficial effect (2). To limit dietary phosphate intake while ensuring adequate protein intake, the Kidney Disease Outcomes Quality Initiative (KDOQI) clinical practice guidelines recommend a low-phosphate diet with phosphate-to-protein ratio of 10–12 mg/g (3). The inherent relationship between phosphorus and protein content in foods makes it difficult for patients on dialysis to adhere to a low-phosphate diet (4). With phosphate-to-protein ratio as low as 8 mg/g, low-phosphate hospital diets have been recommended for patients on dialysis to control serum phosphate level in both inpatient and outpatient settings (5).

Patients receiving hemodialysis have an extremely higher risk of cardiovascular morbidity and mortality than the general population (6,7). Among several cardiovascular risk factors, an elevated fibroblast growth factor 23 (FGF23) level is strongly associated with left ventricular hypertrophy (8,9), and elevation of FGF23 has been identified as an independent risk factor for congestive heart failure and mortality in patients on hemodialysis(10,11). In addition, animal studies have demonstrated that a low-phosphate diet reduces circulating FGF23 level (12). Acute change in circulating FGF23 can be achieved through dietary intervention (13,14). Clinical trials assessing the effect of dietary phosphate restriction on FGF23 have focused on nondialysis populations (14,15). In contrast,

little is known about the effects of low-phosphate diets on circulating FGF23 in patients on hemodialysis who have high rates of hyperphosphatemia and elevated FGF23 levels. Furthermore, the optimal amount of dietary phosphate restriction has not been studied in a hemodialysis population.

The aims of this study were to evaluate the short-term effects of low-phosphate diets on circulating FGF23 level and to determine the optimal amount of dietary phosphate restriction in patients on hemodialysis. We compared the FGF23- and phosphate-lowering effects of a very-low-phosphate diet (phosphate-to-protein ratio of 8 mg/g) with those of a low-phosphate diet (phosphate-to-protein ratio of 10 mg/g) in patients on hemodialysis.

Materials and Methods

Study Design

Figure 1 illustrates the study design and outcome assessments. Between January 3, 2018 and June 8, 2018 we conducted a randomized, active-controlled trial with a crossover design in which we randomly assigned the participants with an allocation ratio of 1:1 to two interventions. The trial protocol is available in Supplemental Appendix 1. The study was approved by the institutional review board at Far Eastern Memorial Hospital (FEMH-106108-F) and was registered online before study initiation (Clinicaltrials.gov identifier NCT03367338). At the time of study initiation, the difference in change-from-baseline FGF23 levels between the two diets was designated as the primary outcome measure, without specifying the assay type. After the study participants were enrolled, we specified the primary outcome (FGF23) measured with intact assay, removed 1,25-dihydroxyvitamin D_3 from the list of secondary outcomes, and added C-terminal FGF23 as the secondary outcome. The rationale for the changes are that intact FGF23 is biologically active and can more precisely reflect dietary response than C-terminal FGF23, and 1,25-dihydroxyvitamin D_3 measurement was unavailable at our central laboratory.

Study Population

Participants were recruited from a hemodialysis unit of a tertiary teaching hospital and asked to participate if they met the following inclusion criteria: (*1*) aged >20 years, (*2*) having ESKD and having undergone thrice-weekly hemodialysis for >3 months, (*3*) having adequate dialysis (urea reduction ratio ≥65%), (*4*) most recent serum phosphate level >5.5 mg/dl or between 3.5 and 5.5 mg/dl with regular phosphate binder use, and (*5*) serum intact parathyroid hormone (PTH) level <800 pg/ml. In accordance with available study diets restricted to between 42.5 and 67.5 kg body wt, we did not enroll participants with a dry weight outside of this range. The exclusion criteria were serum albumin level <2.5 g/dl, hospitalization within the past 4 weeks, psychiatric disorders, mental retardation, dislike of the study meals, or poor dietary adherence. All participants provided written informed consent.

Randomization

After recruitment, one of the investigators equally stratified the participants by intact PTH level and dialysis shift before randomization. The intact PTH level is used as one of the stratification factors because it determines whether response to a dietary intervention. Participants

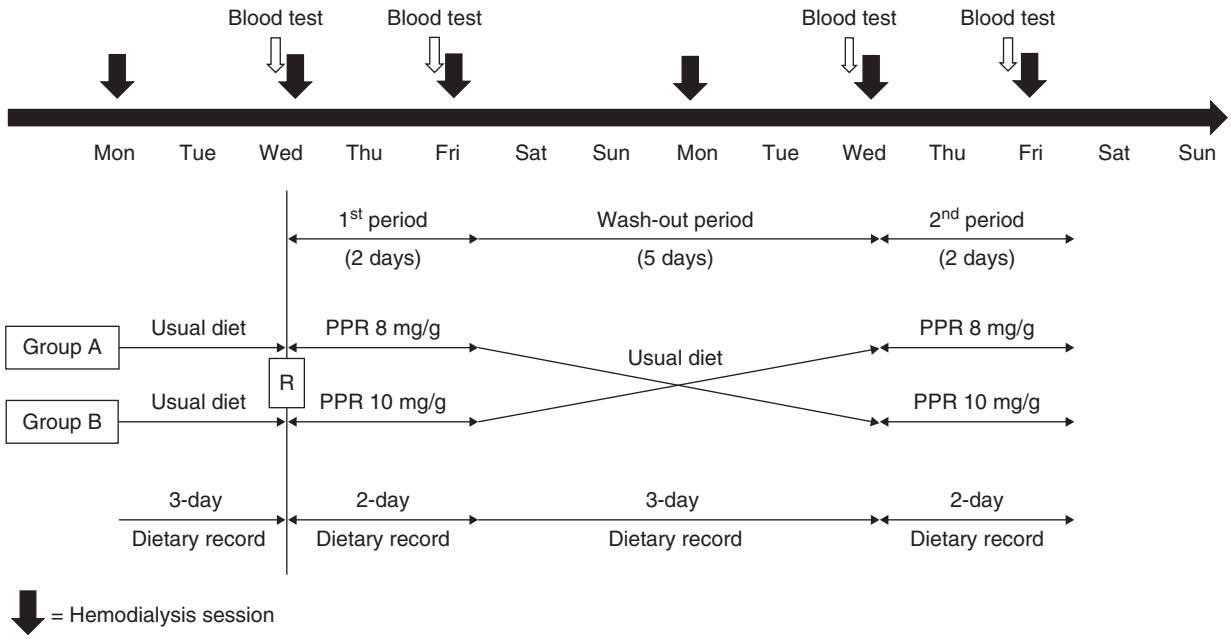

Figure 1. | **Study design and outcome assessments.** Participants in group A received a 2-day diet with a PPR of 8 mg/g, followed by a 5-day washout period, and then received a 2-day diet with a PPR of 10 mg/g. Those in group B received the diets in the opposite order. A total of four repeated measurements for the primary and secondary outcomes were obtained before dialysis sessions. Before each study phase, each participant performed a 3-day dietary record to estimate the nutrient content of his or her usual diet. During the study periods, dietary adherence was assessed by a 2-day dietary record. PPR, phosphate-to-protein ratio; R, randomization.

are stratified by dialysis shift because there is concern about postprandial hypotension, whereby participants with the afternoon shift consume study lunch during hemodialysis and are therefore unable to adhere to the study protocol, leading to an unwanted dropout rate and the subsequent variability in the analyses. Within each stratum, a random allocation sequence was generated by computer-based randomization and used to randomly allocate the participants to the interventions. The investigator sent the random allocation sequence to the dietitian, who prepared individualized study meal boxes for the participants according to assigned group. The study meals were prepared by the same person and with similar natural food materials, appearance, and taste, so that the participants were blinded from their allocation group.

Characteristics of Study Diets

The characteristics and preparation of the study diets are described in Supplemental Appendix 2. In brief, both study diets (5) were made up of natural food sources without any phosphate-containing additives, boiling meats to reduce phosphorus amount (16), and increased proportion of plant-based foods (17). Before patient enrollment, the compositions of the study diets were subjected to chemical analysis; the measurement methods are described in Supplemental Appendix 3. Table 1 lists the nutrient composition of each diet. The two diets were designed to have minimal variation between them in total calorie, protein, and calcium contents, while differing in phosphate and phosphate-to-protein ratio contents. As shown in Supplemental Table 1, the study diet was provided in three meals per day. The nutrient compositions of the study diets categorized according to body weight are provided in Supplemental Table 2.

Table 1. Nutrient compositions of the study diets

Nutrient	Diet of Phosphate-to-Protein Ratio 8 mg/g	Diet of Phosphate-to-Protein Ratio 10 mg/g
Calorie, kcal	1677±175	1671±176
Protein, g	67±8	67±7
Protein per body wt, g/kg	1.2±0.1	1.2±0.1
Plant protein, g	36±5	29±4
Fat, g	39±5	52±7
Carbohydrate, g	264±26	233±24
Calcium, mg	398±38	452±63
Phosphate, mg	549±64	689±76
Phosphate per body wt, mg/kg	10.0±0.4	12.6±0.6
Phosphate-to-protein ratio, mg/g	8.2±0.2	10.3±0.3

Data are given as mean±SD. Both study diets were designed to fulfill the following criteria: (*1*) adequate calories (≥30 kcal/kg per day), (*2*) high protein (≥1.2 g/kg per day), (*3*) low phosphate-to-protein ratio (≤10 mg/g), (*4*) low phosphate content (≤800 mg/d), (*5*) increased protein source of phosphate from plant in origin, and (*6*) meats boiled for 30 minutes before cooking.

Dietary Interventions

As shown in Figure 1, each participant consumed two low-phosphate diets, each for 2 days, and diet order was randomized. Participants in group A consumed a very-low-phosphate diet, with a phosphate-to-protein ratio of 8 mg/g, which is equivalent to 10 mg phosphate per kg of body wt, for 2 days, which was followed by a 5-day washout period in which they adhered to their usual diets. Then, they followed a 2-day low-phosphate diet, with a phosphate-to-protein ratio of 10 mg/g, which is equivalent to 12.6 mg phosphate per kg of body wt. Those in group B consumed the study diets in the opposite order. No additional food was allowed during each study period.

The delivery of study meals was assimilated into the routine hemodialysis schedule. During the intradialytic period, study meal boxes were provided at the hemodialysis unit. During the interdialytic period, the packaged study meal boxes were retrieved at the hospital cafeteria or delivered to the residences *via* a preexisting home delivery service and consumed by the participants as outpatients. Delivery of the meals and consumption of the study diets were verified by telephone.

We made considerable efforts to enhance dietary adherence; these efforts are summarized in Supplemental Appendix 4. To avoid interference with the effects of low-phosphate diets, the following prescriptions were not allowed to change during the entire study period: dialysis duration, dialysis frequency, dialysis shift, dialysate calcium concentration, and dosages of medications, including phosphate binder, vitamin D analogs, and iron agents. Adherence with phosphate binders was assessed before and after each study period.

Dietary Assessment

Before each study period, participants prospectively maintained a dietary record of their daily intake for 3 days, including a dialysis weekday, a nondialysis weekday, and a nondialysis weekend day, allowing us to estimate the nutrient content of their usual diet as well as dietary adherence. During the study periods, the participants maintained a 2-day dietary record of their consumption of portions of the assigned study diets and foods outside of the study diets. The completeness, consistency, and clarity of the food diaries were reviewed by the dietitians.

Data Collection

The following data were recorded: age, sex, dry weight, body mass index, duration of dialysis therapy, history of parathyroidectomy, interdialytic weight gain, dialysis unit BP, type of arteriovenous shunt, dialysate calcium concentration, urea reduction ratio, hemoglobin, ferritin, alkaline phosphatase, albumin, 25-hydroxyvitamin D, glucose, and the amount, frequency, and type of medications, including phosphate-binding agents and vitamin-D analogs. The phosphate-binder doses among study participants were compared by calculating phosphate-binding equivalent dose as described by Daugirdas *et al*. (18).

Laboratory Measurements

Nonfasting venous blood samples were drawn at around 7:30 AM for participants with the morning shift and at

around 12:30 PM for those with the afternoon shift. At the baseline and the beginning of each study period, blood samples were drawn before the second dialysis session of the week. At the end of each study period, blood samples were drawn before the third dialysis session of the week. Standard assays for serum phosphate and calcium were performed using automated analyzers. Intact PTH was analyzed in serum using an immunoradiometric assay (ELSA-PTH; Cisbio Bioassays, Codolet, France). Intact FGF23 was assessed in serum using an ELISA (Kainos Laboratories, Tokyo, Japan). The C-terminal fragments of FGF23 were assessed in EDTA-plasma using a sandwich ELISA (Immutopics, San Clemente, CA) according to the manufacturer's instructions. Each sample was run in duplicate, and mean values are presented.

Outcomes

The primary outcome measure was mean difference in change-from-baseline intact FGF23 level between the two dietary interventions. Secondary outcomes were difference in change-from-baseline serum phosphate, intact PTH, and C-terminal FGF23 levels between the two dietary interventions. We also measured difference in change-from-baseline serum calcium level between the two dietary interventions. As depicted in Figure 1, a total of four repeated measurements for each participant were obtained. The laboratory technicians who performed the outcome measurements were blinded to the allocation sequence.

Sample Size Determination

On the basis of the results from our meta-analysis (1), a total sample size of 29 was required to achieve 90% power and a type 1 error of 0.05, assuming a standardized mean difference in FGF23 levels between two low-phosphate diets of 0.74 and a dropout rate of 25%.

Statistical Analyses

Continuous measures were evaluated as means (±SD) or median (first and third quartiles), and categorical variables as counts and percentages. For non-normally distributed data, Wilcoxon signed-rank tests were performed to evaluate between-group differences. For normally distributed data, paired *t* tests were performed. Effect estimates for the primary and secondary outcomes were presented with Cohen *d* values, which was calculated as the difference between two means divided by a SD for the data. The sign of Cohen *d* indicates the direction of the effect. The Cohen *d* values of 0.2, 0.5, and 0.8 are suggested to correspond to small, medium, and large effects, respectively. We used mixed-effects models to examine the difference in treatment effect of the low-phosphate diets on outcomes and the presence of potential bias resulting from carryover and period effects. In each mixed-effects model, the dependent variable was a primary or secondary outcome, the participant was included as a random effect, and the independent variables were diet, group, and study period. To take account of the stratified randomization, the stratification factors were included as covariates in the model. A two-sided *P* value of <0.05 indicated statistical significance. All analyses were performed with SAS version 9.4 software (SAS Institute, Cary, NC).

Results

Study Flow Diagram

Figure 2 displays a flow diagram of participant enrollment and participation. A total of 143 participants were screened for eligibility, of whom 35 (25%) underwent randomization. Among them, 34 participants completed the first study period; one participant in group B consumed the study diet for 2 days but declined to undergo blood tests and withdrew at the end of the study period. Another five participants withdrew before the beginning of the second study period. As a result, 29 participants completed the second study period.

Baseline Characteristics

Table 2 describes the baseline characteristics of the participants. The mean age of the participants was 64±7 years, and the mean dialysis vintage was 10±7 years. The mean dry weight was 55±7 kg, and interdialytic weight gain was 2.1 (1.7, 2.6) kg. Approximately 90% of participants had arteriovenous fistula, and 60% used low dialysate calcium.

Comparisons of Outcomes between the Two Study Diets

Table 3 presents comparisons of the primary and secondary outcomes between the two study diets. Over the 2-day period, there was no significant difference in the primary outcome, change-from-baseline intact FGF23 level, between the two study diets. Among the three secondary outcomes, the very-low-phosphate diet significantly decreased serum phosphate level compared with the low-phosphate diet, but there were no significant differences in change-from-baseline intact PTH and C-terminal FGF23 levels between the two study diets. Serum phosphate decreased by 1.0 mg/dl (95% confidence interval [95% CI], 0.8 to 1.3) with the very-low-phosphate diet versus 0.4 mg/dl (95% CI, 0.1 to 0.6) with the low-phosphate diet (mean difference, 0.6 mg/dl; 95% CI, 0.2 to 1.0; P=0.002). As the exploratory outcome, serum calcium increased by 0.3 mg/dl (95% CI, 0.2 to 0.5) with the very-low-phosphate diet versus 0.0 mg/dl (95% CI, −0.1 to 0.2) with the low-phosphate diet (mean difference, 0.3 mg/dl; 95% CI, 0.1 to 0.5; P=0.005). The Cohen *d* values for serum phosphate and serum calcium were 0.6 and −0.6, respectively, indicating the greater phosphate-lowering effect and the calcium-increasing effect by the very-low-phosphate diet relative to the low-phosphate diet.

Comparison of Daily Dietary Intake between the Two Study Diets

Table 4 presents a comparison of estimated daily dietary intake between the two study diets. Dietary intake of phosphate and phosphate-to-protein ratio was lower during the very-low-phosphate period than during the low-phosphate period. There were no significant differences in daily calorie intake, protein intake, and calcium intake between the two study diets.

Assessment of Carryover Effects and Period Effects

Supplemental Table 3 demonstrates the results of the mixed-effects models, which assessed carryover and

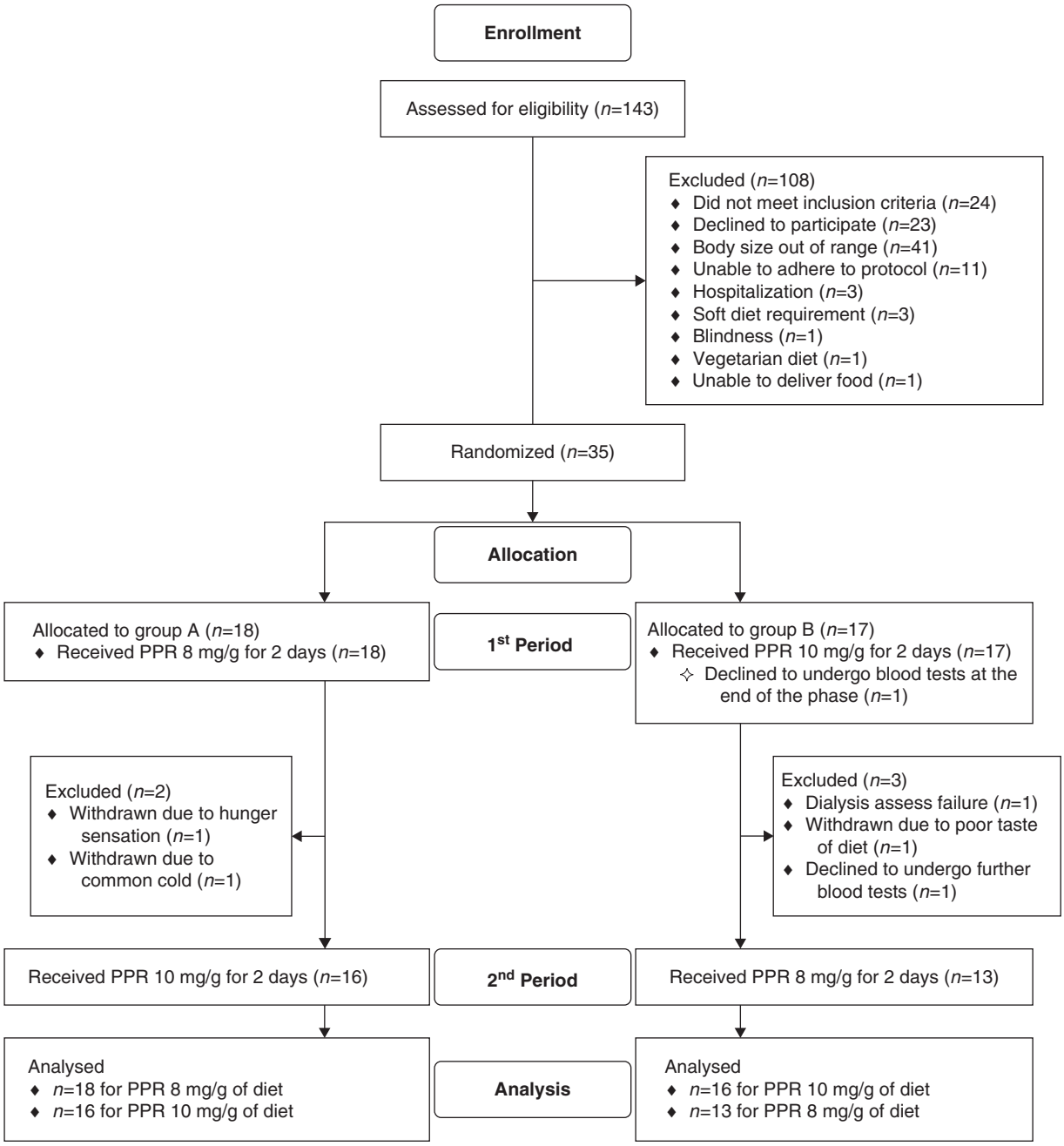

Figure 2. | **Study flow diagram.** PPR, phosphate-to-protein ratio.

period effects. There were no carryover effects for either primary or secondary outcomes between group A participants who consumed the very-low-phosphate diet and the low-phosphate diet in sequence and group B participants who consumed the diets in the opposite sequence. There were no period effects for outcomes including intact FGF23, phosphate, and intact PTH levels, but there was a significant period effect for C-terminal FGF23 level ($P<0.001$). However, there was no significant difference in change in C-terminal FGF-23 levels in group A or group B by Wilcoxon–Mann–Whitney test.

Safety of the Low-Phosphate Diets

Relative to their baseline values, the dry weight, serum albumin, and glucose levels of the participants did not change by the end of the study. One participant who completed the first study period complained of hunger sensation and withdrew before the beginning of the second study period. One participant had mild abdominal cramping pain while consuming breakfast but completed both of the study diets. Four participants exhibited serum phosphate levels below the lower normal limit (2.7 mg/dl), and their phosphate-binder doses were decreased accordingly. We observed no adverse events, including postprandial

Table 2. Participant characteristics at baseline

Characteristic	All Participants (*n*=35)	Group A (*n*=18)[a]	Group B (*n*=17)[a]
Age, yr	64±7	65±7	63±6
Male sex, *n* (%)	14 (40)	9 (50)	5 (30)
Vintage, yr	10±7	10±6	10±7
Dry weight, kg	55±7	56±8	54±6
Body mass index, kg/m^2	22±2	22±2	22±3
Interdialytic weight gain, kg	2.1 (1.7, 2.6)	2.0 (1.7, 2.4)	2.2 (1.7, 2.8)
Systolic BP, mm Hg	133±30	136±31	131±27
Diastolic BP, mm Hg	71±15	73±14	69±16
Urea reduction ratio (%)	74±4	75±4	73±3
Use of AVF, *n* (%)	31 (89)	16 (89)	15 (88)
Low dialysate calcium, *n* (%)[b]	21 (60)	11 (61)	10 (59)
Use of iron agent, *n* (%)	11 (31)	6 (33)	5 (29)
Ferritin, ng/ml	362 (218, 531)	380 (313, 533)	345 (129, 520)
Hemoglobin, g/dl	11.1 (10.8, 11.9)	11.1 (10.7, 11.9)	11.3 (10.8, 11.8)
Parathyroidectomy, *n* (%)	15 (43)	6 (33)	9 (53)
Phosphate-binding equivalent dose, g/d	3.0 (2.0, 4.8)	3.6 (2.1, 5.2)	2.9 (2.0, 4.0)
Vitamin D analogs, *n* (%)	15 (43)	8 (44)	7 (41)
Phosphate, mg/dl	5.0 (4.4, 6.1)	4.6 (4.1, 6.0)	5.1 (4.4, 6.0)
Calcium, mg/dl	9.4 (8.9, 9.8)	9.5 (9.0, 9.9)	9.3 (8.8, 9.7)
Alkaline phosphatase, IU/L	80 (64, 101)	77 (65, 89)	83 (63, 110)
Albumin, g/dl	4.0 (3.8, 4.2)	4.1 (3.9, 4.4)	3.9 (3.6, 4.1)
Glucose, mg/dl	130±61	133±51	128±70
25OHVitD, ng/ml	30 (22, 40)	32 (28, 40)	25 (16, 40)
Intact PTH, pg/ml	147 (80, 332)	150 (118, 346)	97 (76, 313)
Intact FGF23, pg/ml	3401 (529, 9706)	3799 (466, 9429)	1937 (700, 9936)
C-terminal FGF23, RU/ml	6547 (1787, 12,339)	5777 (1294, 11,591)	6547 (3221, 12,975)

Data are shown as mean±SD or median (first and third quartiles), and categorical variables as counts (percentages). AVF, arteriovenous fistula; 25OHVitD, 25-hydroxyvitamin D; PTH, parathyroid hormone; FGF23, fibroblast growth factor 23.
[a]Participants in group A received a 2-day diet with a phosphate-to-protein ratio of 8 mg/g, followed by a 5-day washout period, and then received a 2-day diet with a phosphate-to-protein ratio of 10 mg/g. Those in group B received the diets in the opposite order.
[b]Low dialysate calcium means dialysate calcium concentration of ≤2.5 mEq/L.

hypotension, while eating during hemodialysis over a total of 21 dialysis sessions.

Discussion

This is the first study to investigate the effect of low-phosphate diets on FGF23 levels and the optimal amount of dietary phosphate restriction in patients undergoing hemodialysis. We conducted a randomized, crossover trial to compare the short-term beneficial effects of a very-low-phosphate diet and a low-phosphate diet on mineral parameters. The mixed-effects models indicated a lack of potential bias due to period or carryover effects. We found that the very-low-phosphate diet did not provide an additional benefit for FGF23 reduction measured with either assay, but the phosphate-lowering effects of 2-day low-phosphate diets exhibited dose-response relationships and demonstrated that the optimal dose of dietary

Table 3. Comparison of outcomes after consuming a low-phosphate diet for 2 days

Outcome	Diet of Phosphate-to-Protein Ratio 8 mg/g (*n*=31)		Diet of Phosphate-to-Protein Ratio 10 mg/g (*n*=32)		Between-Group Difference (*n*=29)	
	Before	After	Before	After	Effect Estimate (Cohen *d*)[a]	*P* Value[b]
Primary outcome						
Intact FGF23, pg/ml	5598±6614	4578±4943	5394±6247	4372±5124	−0.01	0.87
Secondary outcomes						
Phosphate, mg/dl	4.9±1.1	3.9±1.0	4.8±1.0	4.4±1.0	0.62	0.002
Intact PTH, pg/ml	208±211	186±195	225±255	204±240	−0.003	0.82
C-terminal FGF23, RU/ml	6354±5621	6184±5937	6129±5716	5960±6160	−0.13	0.63

Data are presented as mean±SD. FGF23, fibroblast growth factor 23; PTH, parathyroid hormone.
[a]Cohen *d* is calculated as the difference between two means divided by a SD for the data. The sign of Cohen *d* indicates the direction of the effect. The positive value of the Cohen *d* means that the very-low-phosphate diet (diet of phosphate-to-protein ratio 8 mg/g) provides a greater lowering effect on a primary or secondary outcome, and the negative value corresponds to a greater increasing effect. The Cohen *d* values of 0.2, 0.5, and 0.8 are suggested to correspond to small, medium, and large effects, respectively.
[b]For non-normally distributed data, Wilcoxon signed-rank tests are performed. For normally distributed data, paired *t* tests are performed.

Table 4. Comparison of estimated daily dietary intake between the two study diets

Nutrients	Diet of Phosphate-to-Protein Ratio 8 mg/g (*n*=31)		Diet of Phosphate-to-Protein Ratio 10 mg/g (*n*=33)		Between-Group Difference[c] (95% CI) (*n*=29)
	Baseline[a]	During Study[b]	Baseline[a]	During Study[b]	
Calorie, kcal	1551±312	1612±346	1522±343	1564±326	−14 (−132 to 104)
Protein, g	56±15	66±13	55±16	65±12	1 (−7 to 8)
Calcium, mg	264±110	366±84	254±122	405±104	55 (−11 to 122)
Phosphate, mg	729±289	557±117	704±211	663±135	128 (4 to 251)[d]
Phosphate-to-protein ratio, mg/g	12.8±2.2	8.4±0.5	13.1±2.5	10.1±0.5	1.5 (0.4 to 2.6)[d]

Data are provided as mean±SD, except where indicated. 95% CI, 95% confidence interval.
[a]We calculated a 3-day average value of estimated daily dietary intake before the study.
[b]We calculated a 2-day average value of estimated dietary intake during study period.
[c]Paired *t* tests were performed for between-group differences.
[d]*P* value <0.05.

phosphate intake should be as low as 8 mg/g phosphate-to-protein ratio, equivalent to an intake of 10 mg phosphate per kg of body wt. These results highlight the benefits of dietary phosphate restriction in the management of hyperphosphatemia after only 2 days of controlled study diets, providing a rationale for recommending a low-phosphate diet to patients on dialysis for management of hyperphosphatemia.

On the basis of evidence from mainly protein-restricted studies of nondialysis populations, KDOQI clinical practice guidelines recommend a phosphate-restricted diet with a phosphate-to-protein ratio of 10–12 mg/g (3). In fact, it is not practical for patients on dialysis to adopt a unique diet with such a low phosphate-to-protein ratio without adequate support from nutritional counseling. In this study, we demonstrated that the very-low-phosphate diet yielded larger phosphate-lowering effect than the low-phosphate diet. A decrease in daily phosphate-to-protein ratio of 2 mg/g for 2 days achieved a reduction in serum phosphate of 0.6 mg/dl. The rapid decrease of serum phosphate was associated with increase of serum calcium (P=0.04). Our results imply that short-term dietary phosphate restriction rapidly improves serum phosphate level and that a phosphate-restricted diet with a phosphate-to-protein ratio of 8 mg/g might be recommended for patients on dialysis with hyperphosphatemia.

Despite the considerable evidence in healthy humans and nondialysis patients with CKD that dietary phosphate restriction lowers FGF23(1,13,14,17,19), we measured FGF23 with intact and C-terminal assays and observed no additional reduction in either FGF23 levels in response to the very-low-phosphate diet compared with the low-phosphate diet. There are several possible reasons for this discrepancy. The main explanation is that the difference between the two study diets regarding phosphate-to-protein ratio was not large enough to achieve a dose-response FGF23-lowering effect. In this active-controlled trial, we ethically defined the low-phosphate diet with a phosphate-to-protein ratio of 10 mg/g as the active comparator, which is the lower limit of 10–12 mg/g as suggested by KDOQI guidelines, and that value is recommended for hemodialysis patients with hyperphosphatemia (3). To test our hypothesis that lowering dietary phosphate intake has a better FGF23-lowering effect, we crafted the very-low-phosphate diet with a phosphate-to-protein ratio of 8 mg/g as the experimental treatment (5). We observed the less than anticipated phosphate-to-protein ratio contrast between the two study diets (mean difference, 1.5 mg/g; 95% CI, 0.4 to 2.6). This is an important and unexpected limitation, which may be explained by difference in dietary adherence rate, 61% during the very-low-phosphate period versus 71% during the low-phosphate period, and a higher phosphate-to-protein ratio of extra intake by the participants during study periods than that of study diet. Because our study is the first to our knowledge that compares the effect of low-phosphate diets with different phosphate-to-protein ratios on FGF23 levels and demonstrates that diet fails to affect FGF23 in a dose-response manner in patients on dialysis, future studies investigating the effect of low-phosphate diets may be more likely to detect differences in FGF23 levels if low-phosphate diets are designed with a sufficient difference in phosphate-to-protein ratio.

Another possibility for the neutral result of our study is that the study duration was probably too brief for a dialysis population in whom FGF23 levels are usually 1000 times above normal and are chronically elevated. Although we demonstrated the additional reduction in serum phosphate levels in response to the low-phosphate diet with a phosphate-to-protein ratio of 8 mg/g for 2 days, patients on dialysis might require a longer duration of dietary phosphate restriction to lower FGF23 levels. We chose a study duration of 2 days because changes to FGF23 levels through dietary modulation can occur within 2 days of changing diet in previous studies (13,14). In contrast to these findings from healthy volunteers, data from CKD studies indicate that 1 week of dietary intervention was required to lower FGF23 levels (1,17). Future studies should assess the effect of low-phosphate diet in a dialysis population for a longer duration.

There is growing evidence that eating during hemodialysis treatment can improve nutritional status, quality of life, and survival (20). However, it is usually discouraged because of concerns of postprandial hypotension. In our study, participants with the afternoon shift consumed study lunch during hemodialysis. There was a total of

21 dialysis sessions requiring participants to eat during hemodialysis treatment, but we observed no adverse events including postprandial hypotension. Similarly, a recent study by Choi *et al.* (21) indicated that eating during hemodialysis did not increase symptomatic hypotension events. Consuming meals during hemodialysis is a favorable practice and low-phosphate hospital diets are a useful way to provide meals during hemodialysis treatment.

Our study has a few other limitations. First, we designed our study to assess the short-term beneficial effects of low-phosphate diets; thus, the duration of dietary intervention was only 2 days. Any extrapolation to the effects of low-phosphate diets over longer periods is not recommended. Second, our study included hemodialysis patients with a mean vintage of 10 years and high FGF23 levels. Therefore, the phosphate-lowering effects of low-phosphate diets that we observed are specific to a dialysis population. Third, we did use nonfasting blood measurements, and some would argue that morning or fasting blood work is the norm in clinical practice, and suggest to use fasting morning blood work for ascertaining differences in mineral parameters. Given that our goal was to evaluate the short-term effect of different diets on mineral parameters, and these values are known to have a circadian change throughout the day, examining fasting morning specimens could underestimate the effect of dietary intervention. Finally, the investigators were not blinded because individualized study meals were required. However, the low-phosphate diets were prepared using similar natural food materials and methods such that the participants could not infer their intervention assignments from the appearance or taste of the study meals. Thus, the participants were blinded after assignment to interventions. Furthermore, the selected outcomes were objective, and outcome assessments were made by independent technicians blinded to the assigned intervention; therefore, potential bias due to a lack of blinding of participants and personnel could be ignored.

In conclusion, the FGF23-lowering effect of the very-low-phosphate diet was similar to that of the low-phosphate diet during the 2-day period. The very-low-phosphate diet has an additional phosphate-lowering effect compared with the low-phosphate diet.

Data Sharing

Individual-level, deidentified participant data will be made available upon request by emailing the corresponding author of this article. The data will be available for 3 years after publication.

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Disclosures

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Supplemental Material

This article contains the following supplemental material online at http://cjasn.asnjournals.org/lookup/suppl/doi:10.2215/CJN.04250419/-/DCSupplemental.

Supplemental Appendix 1. Study protocol.

Supplemental Appendix 2. Characteristics of the study diets.

Supplemental Appendix 3. Methods for chemical analysis of dietary composition of the study meals.

Supplemental Appendix 4. Actions to promote dietary adherence.

Supplemental Table 1. Ingredients of the study diets.

Supplemental Table 2. Nutrient compositions of the study diets according to participant body weight.

Supplemental Table 3. Assessment of carryover effects and period effects by mixed-effects models.

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