Chronic Kidney Disease-Mineral and Bone Disorder
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Guest Editors: Xiong-Zhong Ruan, You-Ying Chau, and Jin-Bor Chen
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Chronic kidney disease-mineral and bone disorder (CKD-MBD) is common in CKD patients and plays pivotal roles in the altered bone metabolism and vascular/valvular calcification. Several guidelines on the management of CKD-MBD have been published but large gaps between guidelines and clinical practice still exist. The field is swamped with information coming in the forms of basic and clinical research articles. Timely published review articles and focused special issues on this topic provide quick and valuable tools for the busy medical professionals looking after CKD patients.

In this special issue, we offer a nexus for the most updated collection of reviews and clinical studies on CKD-MBD. This issue includes three review articles and six clinical studies. For better understanding the mechanisms of CKD-MBD, Dr. Y. Iwasaki et al. provide an intensive review on molecular abnormalities underlying bone fragility in chronic kidney disease. Dr. Y.-C. Hou et al. review roles of vitamin D in uremic vascular calcification. From practical viewpoints, Dr. N.-C. Chen et al. review strategies to prevent and regress the vascular calcification in dialysis patients.

Clinical studies presented in this special issue collectively cover a broad spectrum of CKD-MBD. Dr. W.-H. Wang et al. investigate the association between parathyroid hormone, 25 (OH) vitamin D, and chronic kidney disease. Dr. Y.-P. Sun et al. analyze clinical epidemiology of mineral bone disorder markers in prevalent hemodialysis patients in Xinjiang and call for an urgent need to improve dialysis adequacy in the largest province in China. Dr. Y. Liu et al. examine the practical use of the KDIGO recommended target range of CKD-MBD markers in the real world and find that achievement of the suggested serum phosphorus level is the most important factor for lowering mortality in hemodialysis patients. In line with Dr. Y. Liu’s findings, by looking at coronary artery calcification in peritoneal dialysis patients, Dr. D. Shang et al. address hyperphosphatemia and hs-CRP important initiators of vascular calcification in CKD-MBD. Parathyroidectomy (PTX) is indicated in CKD stages 3–5D with severe hyperparathyroidism that fail to respond to medical therapy. However, the impacts of PTX on serum fibroblast growth factor 23 (FGF23) and soluble Klotho levels and the optimal post-PTX intact parathyroid hormone (iPTH) levels are seldom investigated. By analyzing a larger patient cohort, Dr. S.-C. Liao et al. find no changes in serum concentrations of FGF23 and soluble Klotho in hemodialysis patients after total PTX. Dr. Q. P. Xi et al. look into the impact of post-PTX iPTH levels on all-cause mortality in dialysis patients.

Articles presented in this special issue offer novel insights into the pathogenesis of CKD-MBD and inspire new ideas in clinical management on this complex problem.
Impact of Different Levels of iPTH on All-Cause Mortality in Dialysis Patients with Secondary Hyperparathyroidism after Parathyroidectomy

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Abstract: Secondary hyperparathyroidism (SHPT) usually required parathyroidectomy (PTX) when drug treatment is invalid. Analysis was done on the impact of different intact parathyroid hormone (iPTH) after the PTX on all-cause mortality. An open, retrospective, multicenter cohort design was conducted. The sample included 525 dialysis patients with SHPT who had undergone PTX. Results: 404 patients conformed to the standard, with 36 (8.91%) deaths during the 11 years of follow-up. One week postoperatively, different levels of serum iPTH were divided into four groups: A: ≤20 pg/mL; B: 21–150 pg/mL; C: 151–600 pg/mL; and D: >600 pg/mL. All-cause mortality in groups with different iPTH levels appeared as follows: A (8.29%), B (3.54%), C (10.91%), and D (29.03%). The all-cause mortality of B was the lowest, with D the highest. We used group A as reference (hazard ratio (HR) = 1) compared with the other groups, and HRs on groups B, C, and D appeared as 0.57, 1.43, and 3.45, respectively. Conclusion: The all-cause mortality was associated with different levels of iPTH after the PTX. We found that iPTH > 600 pg/mL appeared as a factor which increased the risk of all-cause mortality. When iPTH levels were positively and effectively reducing, the risk of all-cause mortality also decreased. The most appropriate level of postoperative iPTH seemed to be 21–150 pg/mL.

1. Introduction

SHPT is a common and serious problem in patients with chronic kidney disease (CKD). With the increasing number of patients receiving long-term maintenance dialysis, SHPT appears more in patients receiving dialysis and eventually develops to refractory secondary hyperparathyroidism (rSHPT). Elevated serum concentrations of PTH may contribute to active vitamin D treatment resistance, bone and joint pain, pruritus, fractures, skeletal malformations, and cardiovascular calcification and are independently associated with all-cause and cardiovascular-related mortality [1–4]. It is believed that optimal levels of serum iPTH are different in various stages of CKD and demonstrated that either very low or high levels of serum iPTH are associated with the related mortality [5–7]. However, researchers are always interested in the following questions: what kind of PTX should rSHPT patients undergo and what is the best range of serum level of iPTH after the PTX. Since there has been a lack of relevant evidence, the present study was to investigate the associations
between different levels of serum iPTH and the mortality of patients, after the PTX. The unique features of our study hinge on the relatively large sample size with corporation of multicenters, as well as with a long-term follow-up program.

2. Materials and Methods

2.1. Selection of Patients. Patients under study were from five hospitals, including China-Japan Friendship Hospital and the Aerospace Center Hospital in Beijing, Dalian University Affiliated Xinhua Hospital in Liaoning, the Fourth Hospital of Jilin University in Jilin, and Cangzhou People’s Hospital in Hebei. Patients were followed up for 1–11 years. Inclusive criteria on patients are as follows: (1) patients who had received all prevalent dialysis with duration > 3 months, at any age; (2) patients with the level of serum iPTH > 800 pg/mL which met the PTX criteria and with hypercalcemia, hyperphosphatemia, or calcium × phosphorus (Ca × P) > 70 mg²/dl² [3]. Exclusion criteria on patients are as follows: (1) patients who did not receive dialysis but with CKD; (2) patients who had primary hyperparathyroidism; (3) patients who had received kidney transplantation; (4) patients who underwent repeated PTX; (5) patients who were lost to follow-up or with missing data.

2.2. Clinical Data

2.2.1. Baseline Characteristics. The following data were retrieved from the patients’ charts and computer-based records. Demographic details would include age, sex, primary cause of end-stage renal disease (ESRD) (diabetes, hypertension, chronic glomerulonephritis, and polycystic kidney), preoperative and postoperative PTX laboratory biochemical indexes correction of serum calcium (Ca₂⁺), inorganic P, ALP, and iPTH. Number of deaths and complications which are calculated for each outcome were counted as the events, with duration of follow-up or death recorded. Patients who were followed up for more than 1 year were included in the survival analysis as truncated values.

2.2.2. Laboratory Biochemical Index and Methods for Detection. All the samples were collected in the morning or before performing the hemodialysis, from the PTX database in the above said five hospitals. Laboratory indicators were referred to the criteria serum Ca 2.1–2.54 mmol/L (8.4–10.1 mg/dl). Serum Ca was adjusted for serum albumin according to an equation commonly used in the general population: adjusted Ca [Ca_{alb} (mg/dl)] = Ca (mg/dl) + 0.8 × [(4–Serum albumin (g/dl)) / 3] [P 1.13–1.78 mmol/L (3.5–5.51 mg/dl), and ALP 40–150 IU/L, and serum iPTH (16–65 pg/mL) was detected by enzyme-linked immunosorbent assay, from The United States DLS intact-PTH 10–8000 kit.

2.3. Management on Postoperative Patients. PTX surgical operation was standardized in the five hospitals through a training program and a unified postoperative treatment process was implemented and managed by the China-Japan Friendship Hospital. The procedures would include total parathyroidectomy (tPTX), subtotal parathyroidectomy (sPTX), and tPTX with total autotransplantation (tPTX + AT). The Kidney Disease Outcomes Quality Initiative (K/DOQI) clinical practice guidelines were referred to as the post-PTX management processes, which also include daily monitoring of serum Ca once or twice a week after surgery. Patients were provided with foods, rich in protein, calcium, and phosphorus. Post-PTX, orally administered drugs would include calcium carbonate and calcitriol. We also adjusted the dose according to each serum calcium level until calcitriol reached 4 μg per day [3]. When the serum Ca₉₀ level was lower than 1.8 mmol/L, we started to inject calcium gluconate between 1 mg and 2 mg per kg per hour. When the serum Ca₉₀ level was greater than 1.8 mmol/L, the dose of calcium supplements was gradually reduced. Appropriately, we used a high dialysate calcium concentration between 1.75 and 2.25 mmol/L after the PTX.

2.4. Grouping under Different Levels of Serum iPTh. All the postoperative PTX-related data were entered into the follow-up database. All patients were divided into four groups according to the levels of serum iPTh, one week after surgery, regardless of the PTX. The groupings were A: iPTh ≤ 20 pg/mL (n = 205, 50.7%), B: iPTh 21–150 pg/mL (n = 113, 28.0%), C: iPTh 151–600 pg/mL (n = 55, 13.6%), and D: iPTh > 600 pg/mL (n = 31, 7.7%).

3. Statistical Analysis

Stata 12.0 program was performed for statistical analysis. Normal distribution values were expressed as mean ± standard deviation (SD), and t-test was used for comparison of two means of independent samples. The abnormal distribution values were expressed as median (interquartile range) (M (QL, QU)) and rank-sum test was used to compare the differences between groups. Qualitative data were expressed as the number of cases (rate) and Chi-square test was employed for comparison of qualitative variables. Survival analysis was performed with Cox regression while survival curves were represented with Kaplan–Meier curve. Statistically significant threshold was considered as P < 0.05.

4. Results

4.1. Basic Information. Between January 2004 and December 2014, 525 patients underwent the PTX. Under the exclusion criteria, 404 patients (215 males and 189 females) were qualified for analysis (median age as 47.32 ± 11.52 years, with median dialysis vintage as 100.97 ± 54.55 months). Among them, 396 (98%) patients were with hemodialysis while another 8 (2%) patients were with peritoneal dialysis. The primary cause of ESRD would include chronic glomerulonephritis (n = 203, 50.2%), hypertensive nephropathy (n = 25, 6.2%), polycystic kidney disease (n = 20, 5.0%), and diabetic nephropathy (DN) (n = 3, 0.7%), and the rest (n = 48, 11.9%) were CKD caused by chronic pyelonephritis, Chinese herbal medicine-related nephropathy, or patients with unexplainable causes (n = 105, 26.0%) (Table 1).
Table 1: Patients characteristics by baseline iPTH category (n = 404).

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>iPTH ≤ 20 pg/mL</th>
<th>iPTH 20–150 pg/mL</th>
<th>iPTH 151–600 pg/mL</th>
<th>iPTH &gt; 600 pg/mL</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender (male)</td>
<td>112 (54.6%)</td>
<td>58 (51.2%)</td>
<td>24 (43.6%)</td>
<td>21 (67.7%)</td>
<td>215 (53.2%)</td>
</tr>
<tr>
<td>Age (year)</td>
<td>47.36 ± 11.7</td>
<td>46.58 ± 11.46</td>
<td>48.87 ± 9.66</td>
<td>46.9 ± 13.79</td>
<td>47.32 ± 11.52</td>
</tr>
<tr>
<td>Dialysis vintage (month)</td>
<td>106.59 ± 58.59</td>
<td>96.98 ± 50.85</td>
<td>90.58 ± 50.40</td>
<td>96.81 ± 44.76</td>
<td>100.97 ± 54.55</td>
</tr>
<tr>
<td>Primary cause of ESRD</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chronic glomerulonephritis</td>
<td>95 (46.3%)</td>
<td>61 (54%)</td>
<td>27 (49.1%)</td>
<td>20 (64.5%)</td>
<td>203 (50.2%)</td>
</tr>
<tr>
<td>Diabetic nephropathy</td>
<td>1 (0.5%)</td>
<td>1 (0.9%)</td>
<td>0 (0%)</td>
<td>1 (3.2%)</td>
<td>3 (0.7%)</td>
</tr>
<tr>
<td>Hypertensive nephropathy</td>
<td>21 (10.2%)</td>
<td>2 (1.8%)</td>
<td>1 (1.8%)</td>
<td>2 (6.5%)</td>
<td>25 (6.2%)</td>
</tr>
<tr>
<td>Polycystic kidney</td>
<td>13 (6.3%)</td>
<td>4 (3.5%)</td>
<td>1 (1.8%)</td>
<td>2 (6.5%)</td>
<td>20 (5%)</td>
</tr>
<tr>
<td>Other</td>
<td>25 (12.2%)</td>
<td>14 (12.4%)</td>
<td>6 (10.9%)</td>
<td>3 (9.7%)</td>
<td>48 (11.9%)</td>
</tr>
<tr>
<td>Unknown</td>
<td>50 (24.5%)</td>
<td>31 (27.4%)</td>
<td>20 (36.4%)</td>
<td>4 (12.9%)</td>
<td>105 (26%)</td>
</tr>
<tr>
<td>Serum Ca (mmol/L)</td>
<td>2.56 ± 0.24</td>
<td>2.54 ± 0.22</td>
<td>2.67 ± 0.77</td>
<td>2.52 ± 0.16</td>
<td>2.29 ± 0.37</td>
</tr>
<tr>
<td>Serum P (mmol/L)</td>
<td>2.19 ± 0.58</td>
<td>2.21 ± 0.48</td>
<td>2.17 ± 0.48</td>
<td>2.38 ± 0.40</td>
<td>1.58 ± 0.63</td>
</tr>
<tr>
<td>Serum iPTH (pg/mL)</td>
<td>1926.76 ± 933.15</td>
<td>1938.70 ± 803.63</td>
<td>1967.75 ± 717.85</td>
<td>2119.09 ± 716.01</td>
<td>1950.44 ± 854.42</td>
</tr>
<tr>
<td>Death (n, %)</td>
<td>17 (8.29%)</td>
<td>4 (3.54%)</td>
<td>6 (10.91%)</td>
<td>9 (29.3%)</td>
<td>36 (8.91%)</td>
</tr>
</tbody>
</table>

Mean ± standard deviation is described if the variable is normally distributed.

During the 1–11-year follow-up period, the median duration was 2.3 ± 2.03 years, with an all-cause mortality rate as 8.91% (n = 36). In the follow-up period, 7 patients lived for over 10 years, 8 for 8–10 years, 52 for 5–8 years, 146 for 3–5 years, and 372 for 1–3 years. No case of death was observed in the first week after the PTX. We mapped the unadjusted Kaplan–Meier curve according to all-cause mortality of the patients. During the follow-up period, we found that, with patients with SHPT in different iPTH groups, the survival outcomes were different after the PTX. Among them, patients in group B (iPTH 21–150 pg/mL) had the best outcomes while group D (iPTH > 600 pg/mL) had the worst (P < 0.05). Results of this study showed that all-cause mortality had been significantly decreased after the PTX, which also improved the long-term survival outcomes in dialysis patients with SHPT (Figure 1).

4.2. Relationship between Different Groups of iPTH and the All-Cause Mortality. We used the following criteria as the basis of grouping iPTH was administered for one week after PTX to compare the mortality rates of patients in different groups. The all-cause mortality rates appeared as 8.29% (17/205) in group A (iPTH ≤ 20 pg/mL), as 3.54% (4/113) in group B (iPTH 21–150 pg/mL), as 10.91% (6/55) in group C (iPTH 151–600 pg/mL), and as 29.03% (9/31) in group D (iPTH > 600 pg/mL) (P < 0.05), respectively. As a result, all-cause mortality in group D appeared the highest with statistical significance (P < 0.05). When calculating the causes of death, we noticed that 33 were due to cardiovascular events, 2 with cancers, and 1 with some kind of infection.

We used logistic regression model to set group A as the reference to compare the risk ratios on all-cause mortality, between different groups. Table 2 showed the multivariable adjusted odds ratio (OR) and 95% CI associated with the different groups of iPTH. Group D (OR = 5.17, 95% CI 1.93–13.88, P < 0.05) showed statistically significant difference with other groups but not with groups B (OR = 0.42, 95% CI 0.13–1.29, P > 0.05) or C (OR = 1.35, 95% CI 0.49–3.69, P > 0.05) (Table 2). Same results were obtained by Cox proportional hazards regression model. Data from further analysis revealed that the risk factors in group D (HR = 3.45, 95% CI 1.49–7.99, P < 0.05) were significantly different but neither existed in groups B (HR = 0.57, 95% CI 0.19–1.72, P > 0.05) or C (HR = 3.45, 95% CI 1.49–7.99, P > 0.05) (Table 3). The adjusted results demonstrated a significant increase in HR, associated with group D iPTH > 600 pg/mL but not with other groups. We observed an increase in the HR on deaths, with high iPTH.

For all-cause mortality, data from univariate analysis confirmed that factors as age (HR = 1.06, 95% CI 1.03–1.09, P < 0.05) and dialysis vintage1 (HR = 1.0006, 95% CI 1.000–1.001, P < 0.05) were of significant importance. We also found that factors as gender, primary cause of ESRD, and therapies...
Table 2: Results of baseline multivariate logistic regression model for all-cause mortality.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>OR</th>
<th>Standard error</th>
<th>P value</th>
<th>95% confidence interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>iPTH Group B 21–150 pg/mL</td>
<td>0.42</td>
<td>0.24</td>
<td>0.13</td>
<td>0.13–1.29</td>
</tr>
<tr>
<td>iPTH Group C 151–600 pg/mL</td>
<td>1.35</td>
<td>0.69</td>
<td>0.56</td>
<td>0.49–3.69</td>
</tr>
<tr>
<td>iPTH Group D &gt;600 pg/mL</td>
<td>5.17</td>
<td>2.6</td>
<td>0.001</td>
<td>1.93–13.88</td>
</tr>
<tr>
<td>Age</td>
<td>1.07</td>
<td>0.02</td>
<td>0.000</td>
<td>1.04–1.11</td>
</tr>
</tbody>
</table>

Adjusted for age, gender, dialysis vintage, primary cause of end-stage renal disease, serum Ca\textsubscript{ Alb}, P, and ALP.

Table 3: Results of baseline multivariate Cox regression and competing risk regression analysis for all-cause mortality.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>HR</th>
<th>P value</th>
<th>95% confidence interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>iPTH Group B 21–150 pg/mL</td>
<td>0.57</td>
<td>0.32</td>
<td>0.19–1.72</td>
</tr>
<tr>
<td>iPTH Group C 151–600 pg/mL</td>
<td>1.43</td>
<td>0.46</td>
<td>0.55–3.68</td>
</tr>
<tr>
<td>iPTH Group D &gt;600 pg/mL</td>
<td>3.45</td>
<td>0.004</td>
<td>1.49–7.99</td>
</tr>
<tr>
<td>Age</td>
<td>1.06</td>
<td>0.04</td>
<td>1.03–1.09</td>
</tr>
<tr>
<td>Dialysis vintage\textsuperscript{2}</td>
<td>1.0006</td>
<td>0.04</td>
<td>1.00–1.001</td>
</tr>
</tbody>
</table>

Adjusted for age, gender, dialysis vintage, primary cause of end-stage renal disease, serum Ca\textsubscript{ Alb}, P, and ALP.

Figure 2: Multivariate adjusted hazard ratio comparison between different groups. Note: pthg: 1 = iPTH ≤ 20 pg/mL; 2 = iPTH 21–150 pg/mL; 3 = iPTH 151–600 pg/mL; 4 = iPTH > 600 pg/mL. Adjusted for age, gender, dialysis vintage, primary cause of end-stage renal disease, serum Ca\textsubscript{ Alb}, P, and ALP.

under different sorts of dialysis did not show significant correlations with death events.

We used the predictive margins to analyze the relative hazard of mortality. Figure 2 showed the comparison on the risks of all-cause mortality in different iPTH groups. We sorted the HR values from low to high as groups B, A, C, and D. Group D had the highest HR value (P < 0.05), with statistical significance, while the others did not. We concluded that the postoperative serum iPTH levels and the risks of death presented U-shape trends. However, only group D was with statistically significant difference.

4.3. Postoperative Survival in Patients under Follow-Up Program. Patients were followed for different time spans after the PTX with different period as 1 week, 3 months, 1 year, 3 years, 5 years, 10 years, or more. Four groups of patients showed significant decreases on serum iPTH, after the PTX. Under the early follow-up program, four groups of patients showed different degrees of hypocalcemia postoperatively, during hospitalization. Hypocalcemia could be recovered or partially remitted by intravenous or oral calcium supplements. After the PTX, all patients showed different degrees of improvement or complete remission on bone pain, pruritus, and ectopic calcification during the follow-up period. When using the Cox proportional regression model to analyze the multivariable adjusted results, the increase of HR on death was noticed in patients of group B (iPTH 21–150 pg/mL). Patients in group D (iPTH > 600 pg/mL) showed the worst outcomes and in group C (iPTH 151–600 pg/mL) showed the poor outcomes. Prognosis of patients in group A (iPTH < 21 pg/mL) stood the second place among all the groups (Figure 3). Findings from our study suggested that, in CKD patients who kept receiving PTX hemodialysis (CKD5D) or with iPTH > 600 pg/mL, all-cause mortality on multivariables would significantly increase which were associated with the quality of life (P < 0.05) of the patients.

5. Discussion

In this study, more patients with chronic glomerulonephritis (50.2%) were seen than the ones with DN (0.7%). Chronic glomerulonephritis seemed one of the primary causes for ESRD while DN was less commonly seen in China [8]. Jiang et al. [9] retrospectively analyzed the clinical features of 496 patients with SHPT and found that chronic glomerulonephritis was the major primary cause of SHPT and DN was only
1.2%. The low prevalence of SHPT in patients with DN might be caused by the direct suppressive effect of high glucose concentration on PTH secretion from the parathyroid cells [10].

It is recognized that SHPT is a common complication of chronic renal failure. In the early stage of CKD, in order to adapt for the disorders of bone-mineral metabolism, parathyroid would excessively secrete the PTH, causing the disorders of bone-mineral metabolism to make the burden on cardiovascular systems aggravated and increased. SHPT is controlled under the treatments of phosphate binders, vitamin D analogs, or calcimimetics. With long-term uremia, patients would develop resistance to treatments and finally require PTX to participate. This can prevent the development of skeletal malformations and metastatic calcification, such as cardiovascular calcification. Known to each stage of CKD, the best serum PTH levels were considered different. Findings from our studies suggested that both extra high or low PTH could increase the mortality in CKD patients [5, 11, 12].

Because of the high variability in the number and location of parathyroid glands, they are difficult to be completely removed. Eventually, SHPT appears to be of high incidence in ESRD patients. Even after the PTX, the incidence remains relatively high [13, 14]. Chen et al. conducted a meta-analysis of randomized and prospective or retrospective studies. The results indicated that sPTX and tPTX + AT were equally successful in preventing recurrent SHPT and improving serum Ca, P, and PTH [13–16]. Sharma et al. [12] identified 150 patients under dialysis who underwent near-total parathyroidectomy (NTPTX) and were compared with 1,044 non-operated control patients. The results indicated that NTPTX was associated with a significant reduction in the long-term risk of deaths, in patients receiving dialysis. However, in these studies, the sample size not only was small but lacked the evaluation through prospective, long-term follow-up programs on the survival quality. However, researchers always study the following questions: what kind of PTX should rSHPT patients undergo and what is the best range of serum level of iPTH after the PTX. In China, high incidence of SHPT has been seen [10]. Our research findings showed that, in 404 patients who were with PTX, the amount of preoperative iPTH reached 1950.44 ± 42 pg/mL which called for more positive PTX treatment to be carried out.

In 2008, the Dialysis Outcomes and Practice Pattern Study (DOPPS) identified the optimal PTH level as 101–300 pg/mL, with mortality risk being the lowest under this range [11], which was consistent with the 2003 K/DOQI recommended guidelines [3]. Another study presented the results that iPTH > 600 pg/mL could increase the risk of mortality [11], which was consistent with the results of our study. However, this study was mainly aimed at the patients with different stages of CKD5D, but our research interest was focused on the patients who were receiving dialysis after the PTX therapy. Our results showed that in patients with stage of CKD5 who underwent PTX and with iPTH > 600 pg/mL the risk on all-cause mortality would increase (P < 0.05). In 2005, the DOPPS study suggested that when compared with PTH at 150–300 pg/mL level, the risk on all-cause mortality appeared to be higher than on the level of PTH 301–450 pg/mL. PTH > 600 pg/mL was also associated with higher risk on both cardiovascular and the all-cause mortality, even during hospitalization under cardiovascular disease-related diseases. PTH < 50 pg/mL seemed to be associated with mortality. Extra low or high PTH levels were associated with adverse outcomes [5].

Fernandez-Martin et al. examined 6797 adult patients with hemodialysis. They found that either high or low PTH was associated with high risk of mortality. Serum iPTH 398 pg/mL was associated with the minimum relative risk of mortality. Based on the published risk values on mortality, the lowest value being adopted was 168–674 pg/mL, for serum iPTH [6].

Only few studies specifically targeted on the value of PTH levels had been carried out. Considerable studies from home or abroad confirmed that SHPT patients could improve the survival outcomes after the PTX. Results from studies also showed that when patients underwent PTX, the levels of serum phosphorus, serum calcium, Ca × P, and iPTH were significantly reduced, ending in an effective improvement of quality of life on patients [12, 17, 18].

Komaba et al. [7] analyzed the data on 114,064 patients under maintenance of hemodialysis and evaluated the associations between the severity of SHPT and history of PTX, on both 1-year all-cause and cardiovascular mortality rates. They found that PTH < 60 pg/mL and PTH > 500 pg/mL could significantly increase both the all-cause and cardiovascular mortality rates. The results also showed that when compared to patients with iPTH > 300 pg/mL, those with the level of iPTH < 60 pg/mL might have a significant survival advantage. In our study, we noticed that low iPTH level (iPTH < 20 pg/mL) had a tendency of increasing the risk of mortality while the level of iPTH in 21–150 pg/mL could reduce the risk for both all-cause and cardiovascular mortality rates in patients under PTX dialysis.

Since the sample size was not large enough in our long-term follow-up study, no significant statistical differences were noticed. Rhee et al. [19] claimed the persistent low iPTH
(iPTH ≤ 60 pg/mL), an independent risk factor for both aortic arch calcification and mortality in hemodialysis patients. Jean et al. [20] suggested that the very low appearance of PTH level (around 10%) was associated with a significantly higher risk of mortality among non-PTX hemodialysis patients. PTH < 50 pg/mL was considered to be associated with adynamic bone disease and both of them were related to an increased risk on developing cardiovascular calcification and mortality in hemodialysis patients. Attention needs to be paid not only to high PTH but also to low PTH since both of them might influence the quality of life of patients.

6. Conclusion

Results from our study showed that, during the long-term follow-up period, all-cause mortality was associated with different iPTH levels in dialysis patients with SHPT, followed by the PTX. After the PTX, iPTH > 600 pg/mL seemed to be associated with the risk of all-cause mortality. When iPTH levels were positively and effectively decreasing, the risk of all-cause mortality also reduced. The most appropriate level of iPTH was found as 21–150 pg/mL after the PTX. We noticed that both the iPTH levels and the risk of mortality were under U-shapes. Unfortunately, because of the small sample size and inadequate number of patients that completed the whole course of the study, our findings were not able to reach the statistical significance as expected. Sampling error seemed another shortcoming of this study. However, we are planning to continue the study through increasing the sample size to confirm the postoperative best iPTH levels under the PTX and associations with the quality of life.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

Authors’ Contributions

Ling Zhang and Cheng Gang Jin were responsible for conceptualization. Qiu Ping Xi, Xi Sheng Xie, and Rui Zhang were responsible for data curation. Cheng Gang Jin and Yan Bo Li were responsible for formal analysis. Ling Zhang was responsible for funding acquisition, project administration, and supervision. Ling Zhang, Qiu Ping Xi, Rui Zhang, Yue Fei Xiao, Lin Wang, Xiao Xuan Zhang, and Shu Tong Du were responsible for investigation. Ling Zhang, Yue Fei Xiao, Lin Wang, Xiao Xuan Zhang, and Shu Tong Du were responsible for resources. Qiu Ping Xi and Rui Zhang were responsible for software. Ling Zhang and Cheng Gang Jin were responsible for validation and visualization. Qiu Ping Xi, Ling Zhang, and Xi Sheng Xie were responsible for methodology and writing of original draft. Qiu Ping Xi and Xi Sheng Xie contributed equally to this work. Qiu Ping Xi, Xi Sheng Xie, and Ling Zhang were responsible for writing, review, and editing of the manuscript.

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References


Review Article

Molecular Abnormalities Underlying Bone Fragility in Chronic Kidney Disease

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Prevention of bone fractures is one goal of therapy for patients with chronic kidney disease-mineral and bone disorder (CKD-MBD), as indicated by the Kidney Disease: Improving Global Outcomes guidelines. CKD patients, including those on hemodialysis, are at higher risk for fractures and fracture-related death compared to people with normal kidney function. However, few clinicians focus on this issue as it is very difficult to estimate bone fragility. Additionally, uremia-related bone fragility has a more complicated pathological process compared to osteoporosis. There are many uremia-associated factors that contribute to bone fragility, including severe secondary hyperparathyroidism, skeletal resistance to parathyroid hormone, and bone mineralization disorders. Uremia also aggravates bone volume loss, disarranges microarchitecture, and increases the deterioration of material properties of bone through abnormal bone cells or excess oxidative stress. In this review, we outline the prevalence of fractures, the interaction of CKD-MBD with osteoporosis in CKD patients, and discuss possible factors that exacerbate the mechanical properties of bone.

1. Introduction

Elderly people are susceptible to diseases such as hypertension, diabetes mellitus, and chronic obstructive pulmonary disease. Osteoporosis and chronic kidney disease (CKD) are also common, and the prevalence of these diseases is increasing globally, in part due to the increasing aging population. Osteoporosis under uremic conditions and management of the disease have not been widely studied. The prevalence and risk of fractures are higher in CKD patients compared to healthy people. Patients on dialysis, in particular, have an approximately fourfold greater risk for hip fractures than sex- and age-matched individuals in the general population [1, 2]. Their fracture risk correlates positively with age, duration of dialysis, high or low parathyroid hormone (PTH) level, female gender, low body mass index, and presence of peripheral vascular calcification. Several studies report that nondialysis patients aged over 50 with estimated glomerular filtration rate (eGFR) below 60 mL/min/1.73 m² also have a twofold greater risk for hip fractures than individuals without CKD [3–7]. A hip fracture critically limits activities of daily living and increases fracture-related mortality [8–10], and this trend is more evident in dialysis patients [11, 12]. Japanese dialysis patients, however, have relatively better prognosis with regard to survival after a hip fracture [13]. A tool called FRAX® that can predict fracture risk appears to be useful for predicting death among Japanese hemodialysis patients [14]. Even though it remains unclear why FRAX was useful to predict mortality in Japanese dialysis patients, elucidation of the pathogenesis of decreased bone strength and treatment of fractures in patients with CKD are important to improve survival and the quality of life in this patient population. In this review, we describe the current status of fragility fractures and their treatments in CKD patients.

2. Risk Factors of Fragility Fractures in CKD Patients

Clinicians and researchers agree that risk factors for fractures in CKD are complicated because patients have many abnormalities that may increase fracture incidence. Advanced muscle weakness [15], frailty [16], and deteriorated cognitive
function [17] are potential contributors to increased risk for falling among CKD patients. Falls are especially common in older CKD patients [18]. Lack of exposure to sunlight, which contributes to muscle strength, may be a risk factor, because the risk for hip fractures tends to be higher in high-latitude regions of the United States [19]. Despite the high prevalence of hip fractures, clinical studies have failed to elucidate why falling affects the risk for hip fractures but not fractures of other parts of the body such as vertebrae and wrist. In addition, CKD patients also have deteriorated mineral metabolism.

### 3. Definitions of CKD-MBD, Renal Osteodystrophy, and Osteoporosis

The three key bone lesions accompanying CKD are CKD-mineral bone disorder (CKD-MBD), renal osteodystrophy, and osteoporosis, but their definitions are often ambiguous. CKD-MBD is a syndrome defined by the Kidney Disease: Improving Global Outcomes (KDIGO) guidelines as a systemic mineral metabolic disorder associated with CKD, which could result in disorders of bone metabolism and/or the cardiovascular system [20]. CKD-MBD consists of three components: abnormalities of calcium, phosphorus, PTH, and vitamin D metabolism; abnormalities in bone turnover, mineralization, volume, and strength; and soft tissue calcification including vascular calcification. This disease may manifest one component or any combination of the three. According to this definition, “renal osteodystrophy” indicates bone morphologic changes in patients with CKD and is one measure of the skeletal disorder component of CKD-MBD.

Bone lesions accompanying renal dysfunction are symptoms of CKD-MBD, but worsening of mechanical bone strength is not typically mentioned. Impairment of mechanical properties of bone comes under the term “osteoporosis,” as defined by the National Institute of Health. This pathophysiology is characterized by compromised bone strength predisposing a person to increased risk of fractures [21]. In this definition, bone strength is a composite of bone mass and bone quality. Bone mass is a strong determinant of bone strength and is useful as a diagnostic tool for osteoporosis in people with extremely low bone mass. As there are no other tools to predict and/or monitor bone strength in clinical practice, bone mass measurement is considered the most informative and useful tool available to diagnosis osteoporosis. Bone mass, however, is not the only determining factor. Other factors affecting bone mechanical strength include “bone quality.” Bone quality is used to describe the ability of bone to perform mechanical load-bearing functions. This definition includes all characteristics that influence the load-bearing capacity, including bone microarchitecture and material properties [22, 23], Table 1.

A question often arises as to which plays a more important role in bone mechanical strength, bone mass or bone quality. However, the contribution of each of the two parameters remains unclear, because several cohort studies suggest that one-half of all fragility fractures are observed in postmenopausal women with a T-score above −2.5 SD, the threshold for diagnosing osteoporosis defined by the World Health Organization [24–26]. Additionally, postmenopausal women with fragility fractures had poor bone microarchitecture and altered material properties, which influence bone mechanical properties [27–29]. Therefore, bone mass measurement is strictly not the standard method for diagnosing osteoporosis.

With the progression of renal function impairment, fracture risk is remarkably high in CKD. While we suspect that osteoporosis may underlie the increased risk of fracture in CKD, the mechanism may differ from that of primary osteoporosis characterized by marked reduction in bone mass. It is also unclear whether osteoporosis (bone fragility) associated with CKD is derived from CKD-MBD or factors other than CKD-MBD.

### 4. Possible Factors Related to Weakening of Bone Strength

Both clinical and preclinical studies suggest that loss of bone strength in CKD patients has two possible components, loss of bone mass and deterioration of bone quality. The KDIGO guidelines published in 2009 do not recommend routine

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**Table 1: Components of bone quality.**

<table>
<thead>
<tr>
<th>Analytical method</th>
<th>Structural properties</th>
<th>Material properties</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bone histomorphometry</td>
<td>Trabecular number</td>
<td>Bone histomorphometry</td>
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<tr>
<td></td>
<td>Trabecular thickness</td>
<td>FTIR, Raman</td>
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<td>Trabecular connectivity</td>
<td>Cortical thickness</td>
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<td>Cortical thickness</td>
<td>HPLC</td>
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<tr>
<td>MicroCT, pQCT, HR-pQCT</td>
<td>Trabecular number</td>
<td>Back scattered electron imaging</td>
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<td>Trabecular thickness</td>
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<td></td>
<td>Trabecular connectivity</td>
<td>X-ray diffraction</td>
</tr>
<tr>
<td>Bone histomorphometry, CMS</td>
<td>Microdamage length, density</td>
<td></td>
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</tbody>
</table>

Micro-CT, microcomputed tomography; pQCT, peripheral quantitative computed tomography; HR-pQCT, high-resolution peripheral quantitative computed tomography; CMS, contact microradiograph; FTIR, Fourier transform infrared spectroscopy; HPLC, high-performance liquid chromatography; EDX, energy-dispersive X-ray spectroscopy.
bone mineral density (BMD) testing because BMD does not predict fracture risk in patients with kidney disease as it does in the general population [30]. However, a recent meta-analysis reveals that BMD is significantly lower in predialysis patients with fracture compared to those without [31]. A prospective study has shown that BMD measured by dual X-ray absorptiometry (DXA) predicts incident fracture in stages 3–5 CKD patients, and the prediction ability is comparable to that using high-resolution peripheral quantitative computed tomography [32]. Furthermore, two studies have reported the assessment of BMD using DXA to predict fractures in CKD patients including those on hemodialysis [33, 34]. Therefore, BMD measured by DXA may be useful to assess loss of bone mass or fracture risk. On the other hand, cortical bone loss that increases in advanced stage of CKD is not well depicted by DXA. Therefore, DXA still has limited clinical utility in advanced stage of CKD. More attention should be paid to other factors affecting bone strength. Factors contributing to bone strength comprising bone loss and bone quality are discussed below and summarized in Table 2.

5. Humoral Factors Related to Mineral Metabolism

Progressive changes in serum biochemical parameters such as phosphorus, PTH, 1,25(OH)2 vitamin D3, and fibroblast growth factor 23 (FGF23) levels indicate CKD-related disturbances of mineral and endocrine factors [35, 36]. Increased PTH levels powerfully impact bone mechanical properties, because PTH modifies the activities of bone cells, which regulate bone turnover leading to altered bone mass. PTH stimulates the osteoclastic resorption and remodeling speed, thereby increasing bone turnover. Reduction in cortical BMD and thickness together with increase in cortical porosity assessed by DXA or high-resolution peripheral quantitated tomography (HR-pQCT) have been reported to result in increased bone fragility [37–40]. In stable dialysis patients, Kazama et al. [41] showed that circulating PTH level correlates inversely with cortical porosity but not with cancellous bone volume assessed by bone histomorphometry. Parathyroidectomy in patients on maintenance hemodialysis reduced fracture risk [42]. Additionally, elevated serum alkaline phosphatase due to excessive PTH secretion is associated with higher risk of hip fracture [43]. However, contradicting results on the relationship between PTH level and structure-related bone strength have also been reported [44, 45]. Moreover, medical and surgical treatments for severe hyperactive parathyroid function have progressed, and moderate hyperparathyroidism is unlikely to be a major risk factor for skeletal fragility.

Disturbed bone remodeling (marked decreases in both bone resorption and bone formation) caused by suppressed PTH secretion or skeletal resistance to the action of PTH under uremic condition exits in low-turnover bone lesions in CKD [46–48]. This condition is called “adynamic bone,” and is an increasingly common occurrence [49, 50]. Adynamic bone constitutes 50% of all CKD-MBD in patients on peritoneal dialysis and 19% in patients on hemodialysis [30]. Several clinical and animal studies have suggested an increased fracture risk in adynamic bone disease [51–54].

To summarize, the relationship between fracture risk and PTH level, which alters bone remodeling and bone microstructure, remains controversial. Regardless of high or low PTH level, it is currently difficult to predict fracture risk by PTH level.

FGF23 is derived from osteocytes and is an endocrine hormone that regulates phosphate metabolism. FGF23 stimulates urinary phosphate excretion, suppresses absorption in the gut, and accelerates degradation of 1,25(OH)2 vitamin D3 in response to a high phosphate diet or a state of impaired phosphate excretion as seen in CKD. FGF23 level is elevated prior to changes in phosphate, 1,25(OH)2 vitamin D3, and PTH levels accompanying decline in GFR [55–57]. While some studies reported an association between elevated FGF23 secondary to early CKD and risk of fracture in elderly men with decreased eGFR [58–60], other reports found no significant relationship [61, 62]. Isakova et al. [63] analyzed 2234 subjects and reported that FGF23 level was not associated with bone loss or fracture risk in a community-based population of well-functioning older adults. A recent report found that elevated FGF23 induced by high phosphorus diet increased the expression of secreted frizzled-related protein 4 and Diccpf-1, which are Wnt signal inhibitors, and inhibited the Wnt signal pathway [64]. Another report showed that FGF23 also had a physiological role in local bone mineralization, regulating osteopontin indirectly through transcriptional control of tissue nonspecific alkaline phosphatase in a vitamin D- and klotho-independent manner [65]. These

| Table 2: Molecular abnormalities that affect bone loss and bone quality. |
|---------------------------|---------------------------------|---------------------------------|
| Category                  | Factor                          | Loss of bone mass               | Deterioration of bone quality |
| Humoral factors           | PTH                             | Activating bone resorption and modulating bone turnover [42, 43, 46–50] | Modulating microarchitecture [41] |
|                           | FGF23                           | Inhibiting bone formation [64]   | Inhibiting mineralization [65] |
|                           | Sclerostin                       | Inhibiting bone formation [66–68, 70, 71, 75] | Inhibiting mineralization [71] |
|                           | Vitamin D                        | Inhibiting bone formation [76, 77] | Modulating material property [74] |
| Bone aspects              | Microcrack accumulation, osteocytes apoptosis | Modulating bone turnover | Modulating material property [101–103] |

Numerals are reference numbers.
reports suggest that high FGF23 level may affect bone fragility by decreasing mineralization through inhibition of the Wnt pathway.

Sclerostin is a Wnt pathway inhibitor secreted by osteocytes. The canonical Wnt/β-catenin signaling pathway directly affects osteoblast differentiation, proliferation, survival, and bone formation. Sclerostin antagonizes Wnt signaling and inactivates the pathway. The relationship between sclerostin and fracture risk is not consistent among studies [66–69]. Serum sclerostin is high in early CKD and is maintained at a high level in the advanced stages [70–73]. In an animal study, higher serum phosphate concentration derived from a high phosphorus diet was found to elevate sclerostin expression despite increased osteocyte apoptosis [74]. Combination therapy of anti-sclerostin antibody with PTH-suppressive agent was effective in improving bone mass and mechanical properties [75]. It is possible that a high sclerostin level is an aggravating factor of bone fragility.

Vitamin D [25(OH)D3 and 1,25(OH)2D3] deficiency and altered vitamin D metabolism occur in CKD patients. Because vitamin D is required for normal bone formation and mineralization, 25(OH)D3 deficiency (<15 nmol/L) is associated with less bone formation and mineralization in trabecular bone [76]. A lower vitamin D status is associated with increased fracture incidence and risk [77–80]. Recently, Murali et al. [65] showed that 1,25(OH)2D3 inhibits local mineralization by augmenting the expression of the inhibitor osteopontin. To elucidate the involvement of 1,25(OH)2D3 in bone mineralization in CKD, further in vivo and in vitro experiments are required.

An increased incidence of bone fragility was observed in CKD irrespective of variations in PTH, 1,25(OH)2D3, FGF23, and sclerostin levels that reflect disturbances of mineral and endocrine metabolism. Factors other than CKD-MBD which may aggravate weakening of bone mechanical properties in CKD patients should be considered.

6. Uremic Conditions Deteriorate Bone Material Properties

Bone is composed of two organic materials, type I collagen and hydroxyapatite. The number of collagen crosslinks formed by both enzyme-induced and non-enzyme-induced processes as well as tissue mineral content (density) confer bone elasticity and strength. Various abnormalities in bone material properties are found in CKD patients.

The chemical composition of bone can be analyzed by vibrational spectroscopic methods such as Fourier transform infrared or Raman spectroscopy [81, 82]. These methods provide data on mineral parameters including the mineral-to-matrix ratio (indicating the degree of mineral apposition), the degree of carbonate substituting for phosphate in the apatite lattice, and crystallinity (representing the mineral crystal size and perfection). Additionally, collagen maturity can be obtained by calculating the ratio of mature crosslinks to immature crosslinks. Alterations of these parameters in the bones have been reported in animal models of CKD [51, 83–85] and bone biopsy samples from hemodialysis patients [86, 87].

Nonphysiological collagen crosslinks formed by the actions of advanced glycation end-products are modified crosslinks and are found in increased numbers in bone biopsy samples from dialysis patients [88] and animal models of CKD [51, 83–85]. Immunostaining analysis of bones in a rat model of CKD also demonstrated increase in crosslinks modified by advanced glycation end-products and reduced lysyl oxidase protein, an enzyme required for generating physiological collagen crosslinks [89]. The degree of biological bone apatite orientation, which is related to bone elasticity, was assessed by X-ray diffraction [90] and was found to be exacerbated in a rat model of early kidney injury [84]. Interestingly, in experimental uremic animals, these changes were complicated by the progression of renal dysfunction [51], and some changes were independent of bone turnover [84]. The changes were reduced by administration of AST-120, an oral charcoal adsorbent of uremic toxins [83]. AST-120 did not change mineral metabolism. Therefore, the uremic condition may modify the material properties directly.

Uremic conditions are known to create an excess oxidative stress environment [91]. Uremic condition or a specific uremic toxin inhibits osteoblasts [92–96] and osteoclasts [97]. Although whether the material properties are altered in CKD patients with fragile bone has not been confirmed, uremia-related osteoporosis causing bone fragility should exist in CKD.

7. Microcracks and Osteocyte Apoptosis

Because one of the purposes of bone remodeling is to repair microdamage that occurs in bone from daily mechanical stress, suppression or absence of remodeling will result in accumulation of microdamage. Excessive accumulation of microdamage can cause fragility fractures. Although there are no reports that indicate impaired microdamage repair in low-turnover bone associated with CKD, findings that suppressed bone turnover increases fragility and fracture risk suggest accumulation of microdamage [98–100].

The rates of osteocyte apoptosis and reduced density are higher in fractured bone than in normal bone [101–103]. Empty lacunae (absence of osteocytes in lacunae) are found in renal osteodystrophy. PTH fragment, especially the C-terminal PTH fragment, increases osteocyte apoptosis [104]. The C-terminal PTH fragment is accumulated in CKD, and the amount increases depending on renal insufficiency [105]. From these findings, increased osteocyte apoptosis appears to be associated with fragility fractures in CKD patients.

To summarize, bone fragility in CKD is probably caused by loss of bone mass and deterioration of bone quality through changes in blood levels of humoral factors and the presence of uremic toxins (Figure 1).

8. Pharmacological Therapeutics for Bone Fractures in CKD Patients

In the general population, pharmacotherapy is the mainstay of management for osteoporosis. Patients with primary osteoporosis are treated with different types of drugs. Guidelines for primary osteoporosis recommend antiresorptive drugs
Accumulated uremic toxins

Excessed oxidative stress

Abnormal bone cells
(i) Formation/resorption disorders
(ii) Skeletal resistance to PTH
(iii) Impaired cell interactions
(iv) Decreased cell viability

Diminished bone turnover (low turnover bone)

Accelerated bone turnover (high turnover bone)

Altered material properties

Altered microarchitecture

Loss of bone mass

Increased bone fragility

Reduced renal function

Increased Pi conc.
Reduced Pi clearance
Decreased klotho

Increased FGF23
Reduced 1,25(OH)_{2}D_{3}
Reduced Ca conc.

Increasing PTH conc. (secondary hyperparathyroidism)

Figure 1: Possible factors involved in bone fragility. Both mineral metabolism disorders and uremic condition induce bone fragility. The detailed mechanisms and interactions are described in the text. Gray-shaded boxes indicate the phenomena induced by mineral metabolism disorders. Detailed descriptions of components of bone quality are shown in Table 1. Pi, phosphorus; Ca, calcium; FGF23, fibroblast growth factor 23; conc, concentration.

(bisphosphonates, antagonists of osteoclasts, and selective estrogen receptor modulators) and stimulators of bone formation (teriparatide) as well as active vitamin D and calcium supplementation. However, these drugs present problems for CKD patients, because some are excreted via the kidneys. The KDIGO guidelines [30] indicate that extrapolating results from studies of osteoporosis in general population to patients with CKD stages 3–5D may not be valid, with concerns over long-term safety because the pathogenesis differs between primary osteoporosis and CKD-MBD-related osteoporosis. On the other hand, due to the increases in osteoporosis and CKD with advancing age and the proven safety profile of osteoporotic agents, the KDIGO guidelines approve the use of these agents in early CKD patients with high risk of fracture, including patients with osteoporosis and CKD stages 1-2. Potential treatments with antosteoporotic agents in different CKD stages are summarized in Table 3. Additional information for some agents will be discussed in detail below.

Although bisphosphonates have become a standard treatment for osteoporosis, bisphosphonates should not be used in patients with stages 4-5 CKD because these drugs are excreted by the kidney. The accumulation of bisphosphonates in bone also needs to be considered. Ott [106] reported the accumulation of bisphosphonate in the bone of dialysis patients treated with these agents. The use of bisphosphonate in dialysis is a growing concern, as there is the possibility of causing “frozen bone” with extremely low bone turnover. Bisphosphonate exposure over a 5.5-year period was reported to aggravate bone viscoelasticity and provoke atypical femoral fractures [107]. This phenomenon may be a consequence of reduced heterogeneity of material properties through suppressed bone turnover [108]. Bisphosphonates may increase the fracture risk through exacerbation of bone mechanical properties and increase atypical fractures [109–111]. Since the degree of bisphosphonate accumulation and the efficacy of bisphosphonates both depend on their affinity to hydroxyapatite, existing data suggest treatment durations of up to 5 years with alendronate, 3 years with zoledronate, and 1 year with risedronate, although the optimal length of a “drug holiday” has not been established [112]. If it is necessary to use bisphosphonates for the management of severe osteoporosis in patients with CKD, bisphosphonates that have low affinity to hydroxyapatite crystals, such as risedronate and ibandronate, should be chosen.

Denosumab, a human monoclonal antibody for the receptor activator of nuclear factor-kappa B, does not accumulate in the body because its point of action is limited. Its efficacy in CKD is expected to be the same as that in
Primary osteoporosis. A previous study reported that the efficacy of denosumab, which increases BMD and suppresses fractures, did not differ depending on kidney function [113]. Another study reported that denosumab significantly increased BMD of the lumber spine and femoral neck in hemodialysis patients, although the sample size was small [114]. Denosumab may induce hypocalcemia through strong suppression of bone resorption, which tends to be amplified in CKD patients [115]. Denosumab should be prescribed with active vitamin D to regulate the calcium balance.

Raloxifene, a selective estrogen receptor modulator improved BMD in postmenopausal women with CKD, and greater increases in BMD were associated with lower creatinine clearance [116]. In another study, patients on raloxifene showed slower progression of kidney disorders and significantly fewer kidney-related adverse events compared to the placebo group [117]. However, reduced serum calcium concentration and increased PTH secretion were reported.

Teriparatide is a recombinant protein of PTH (1–34) and an anabolic agent for the treatment of postmenopausal osteoporosis. Although teriparatide should be used with caution in osteoporotic patients with CKD due to higher blood PTH level in secondary hyperparathyroidism associated with CKD, intermittent PTH administration can be used to induce an anabolic effect on bone in CKD. Some studies have reported that teriparatide treatment increases BMD [118–120] and ameliorates bone turnover [120]. Subjects of these studies showed decreased endogenous PTH concentration compared to appropriate controls. The effect of teriparatide treatment on CKD patients with normal or slightly higher PTH remains unknown.

Anti-sclerostin monoclonal humanized antibodies such as romosozumab and blosozumab, a new class of drugs with novel mechanisms of action, are being developed for osteoporosis treatment. In clinical trials, romosozumab and blosozumab have been shown to increase bone mass concomitant with increase in bone formation marker and decreases in bone resorption markers [121, 122]. Increases not only in trabecular BMD but also in cortical thickness and stiffness assessed by HR-pQCT were observed in subjects taking romosozumab compared to placebo controls [123]. Although elevated levels of sclerostin have been reported in CKD stages 3 to 5D patients [73, 124, 125], there are no clinical data on anti-sclerostin antibody treatment in CKD patients. In phase 2 in clinical trial of romosozumab, subjects who had estimated creatinine clearance as low as 30 mL/min were included [121]. Since romosozumab was associated with favorable effects on bone turnover in that study population, its efficacy in improving bone fragility in CKD patients may be anticipated.

In addition to the apparent relationship between sclerostin and bone strength, blood level of sclerostin has been shown to be associated with aorta valve calcification [126] and cardiovascular mortality in CKD patients [127, 128]. Further studies are needed to investigate the efficacy of sclerostin antibody treatment not only for fracture prevention but also for reducing cardiovascular mortality in CKD patients.

Control of hyperphosphatemia is important for CKD patients to prevent cardiovascular events and reduce the risk of death. From a secondary analysis of the EVOLVE trial, cinacalcet reduced the rate of clinical fractures by 16–29% [129]. The BONAFIDE trial demonstrated that long-term treatment with cinacalcet substantially reduced PTH and diminished elevated bone turnover as well as several biomarkers [130]. Yamamoto et al. [131] reported that dialysis patients who received angiotensin-converting enzyme inhibitors or angiotensin II type 1 receptor blockers had an approximately 30% lower risk of hospitalization for any fracture. It is possible that, in addition to traditional antosteoporotic drugs, the use of inhibitors of specific pathophysiological conditions associated with renal failure is an appropriate strategy for the treatment of osteoporosis in CKD.

9. Conclusion

Determining the pathogenesis of osteoporosis and treatment efficacy is difficult in CKD patients because of the complicated mineral and bone abnormalities in these patients. As described above, many factors such as BMD, humoral factors, and alterations of material properties potentially affect bone strength. However, the factors that contribute to bone strength in the setting of CKD and their mechanisms of action remain unknown. For example, no changes in structural parameters and bone mechanical parameters were observed 6 months after kidney transplantation, even though BMD was ameliorated [132]. Other report revealed that cortical porosity is not superior to BMD determined by DXA hofr identification of HD patients with fragility fracture [133].

<table>
<thead>
<tr>
<th>Table 3: Pharmacotherapies for osteoporosis according to stage of chronic kidney disease (CKD).</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Agents</strong></td>
</tr>
<tr>
<td>---</td>
</tr>
<tr>
<td>Alendronate</td>
</tr>
<tr>
<td>Risedronate</td>
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<tr>
<td>Etidronate</td>
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<tr>
<td>Iblandronate</td>
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<tr>
<td>Minodronate</td>
</tr>
<tr>
<td>Denosumab</td>
</tr>
<tr>
<td>Raloxifene</td>
</tr>
<tr>
<td>Teriparatide</td>
</tr>
</tbody>
</table>

+: use with caution; −: avoid use.
Moreover, clinical assessment of human femoral mechanical properties by reference point indentation (RPI), which is a novel technique that allows direct measurement of bone material or biomechanical properties, indicated that BMD did not discriminate fracture cases form controls [134]. These recent studies suggest that bone strength in CKD patients may be affected by many factors in a complicated manner, and the major factor and its degree of contribution remain unidentified. Therefore, more studies are required to assess bone mechanical properties using a multitude of factors including BMD and humoral factors. If PRI can be used easily in clinical studies, we may be able to discuss the diagnosis or grading of bone fragility in CKD patients. CKD patients are at increased risk for fractures regardless of whether they are on dialysis. The KDIGO working group is scheduling a selective revision of the guidelines [135]. However, until then, patients at risk of fragility fractures still need to be managed. Researchers, clinicians, pharmacologists, nurses, drug companies, and other authorities should pay particular attention to osteoporosis in CKD patients to determine suitable management.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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Association between Parathyroid Hormone, 25 (OH) Vitamin D, and Chronic Kidney Disease: A Population-Based Study

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Identification of the accurate risk factor for CKD remains mandatory to combat the high prevalence of diseases. Growing evidence suggests the association of serum vitamin D with diverse health conditions. However, the relationship between vitamin D, intact parathyroid hormone (PTH), and calcium-phosphate metabolism and development of CKD remains controversial. We conduct this cross-sectional observational study to investigate the association between serum 25 (OH) vitamin D, intact PTH, and calcium and phosphate levels with eGFR and albuminuria, as a surrogate marker of CKD, in a community population. A total of 4080 participants were recruited. The mean age was 58.4 ± 13.3 years and 1480 (36.3%) were men. The mean eGFR was 94.1 ± 26.3 mL/min/1.73 m². The prevalence of CKD was 19.8%. Serum 25 (OH) vitamin D and log intact PTH levels were inversely correlated with eGFR but positively correlated with log albuminuria. Logistic regression analysis identified the log intact PTH as an independent factor associated with eGFR ≤ 60 mL/min/1.73 m² and proteinuria. This association was consistent when serum intact PTH was analyzed as continuous as well as categorical variables (as hyperparathyroidism). The relationship remains significant using resampling subset analysis with comparable baseline characteristics and adjustment for 25 (OH) vitamin D, calcium, and phosphate levels. This finding warranted further research to clarify the causal relationship of PTH/25 (OH) vitamin D with the risk of CKD in the general population.

1. Introduction

Chronic kidney disease (CKD) is a worldwide public health issue because of its wide association with multiple comorbidities, demanding high cardiovascular events, mortality, and expensive medical cost. The worldwide prevalence of CKD ranged from 8 to 16%, dependent on the disease definition, study design, and racial groups [1]. The estimated national prevalence of CKD is 11.9% [2] in Taiwan, which has the highest incidence and prevalence of end-stage renal disease (ESRD) worldwide [3]. Ascertainment of accurate risk factors associated with CKD is mandatory to allow timely diagnosis and intervention to decrease the burden of the disease. The associations of CKD with several traditional risk factors (such as hypertension, diabetes, obesity, smoking, dyslipidemia, and metabolic syndrome) were well established in the literature [4]. Epidemiological literatures emphasized the association between serum vitamin D level and nonskeletal disease as cardiovascular events [5–7], metabolic syndrome [8], cancer [9, 10], autoimmune disease [11], and critical illness [12]. The decrease of glomerular filtration rate restricts delivery of substrate to the 1-alpha-hydroxylase and decreases the production of 1,25 (OH) vitamin D by the kidney in CKD patients. The interplay between vitamin D, fibroblast growth factor-23 (FGF-23), intact parathyroid hormone (PTH), and calcium-phosphorus-bone metabolism is disrupted with the progression of renal function [13–15]. However, the relationship between vitamin D, intact PTH, and calcium-phosphate metabolism and development of CKD remains controversial.
A Korean population-based study found a biphasic change of serum vitamin D levels, according to CKD severity. The estimated glomerular filtration rate (eGFR) was negatively associated with serum vitamin D levels [16] in the entire population; however, the mean vitamin D values were decreased with the lowering of eGFR levels in moderate and severe CKD stages [16, 17]. A Swiss population study did not find any association between serum vitamin D levels and CKD or albuminuria [18]. In addition, the association between vitamin D levels and renal function decline was lost after adjustment for baseline eGFR [19]. Accumulating evidence indicated the association of abnormal calcium-phosphate metabolism with cardiovascular disease in the general population. Intact PTH levels were associated with hypertension [20] and abnormal calcium-phosphate metabolism was associated with coronary artery calcification [21]. The prevalence ratio of intact PTH among participants of the US National Health and Nutrition Examination Survey showed a stepwise increment of 2.30 and 4.69 for individuals with an eGFR of 45 to 59 and 30 mL/min/1.73 m² respectively [22]. While the serum value variation and the relationship between vitamin D/intact PTH and hard outcomes were evident in moderate to late stage CKD and ESRD patients, little is known about the exact role of vitamin D, intact PTH, and calcium and phosphate levels all together in the risk of CKD in healthy subjects.

We conduct this cross-sectional observational study to investigate the association between serum 25 (OH) vitamin D, intact PTH, and calcium and phosphate levels with eGFR and albuminuria and the risk of CKD in a community population.

2. Method

2.1. Patient Setting and Data Description. Participants of community health activity from August 2013 to May 2016 in the northeastern region of Taiwan were enrolled in the study. Participants aged greater than 30 years and who were not pregnant were included after obtaining informed consent (n = 4916). Individuals who had received vitamin D supplementation or any over-the-counter vitamin supplements (n = 830) or incomplete baseline biochemistry data (n = 6) were excluded from the analysis. Demographic data were assessed from questionnaires. Anthropometric and biochemistry measurements were performed at the entry of the study. Blood samples were obtained after an overnight fast, and the following parameters were determined: complete blood cell count, liver and renal biochemistry parameters, lipid profiles, fasting sugar, and intact PTH and total 25 (OH) vitamin D levels. This study was approved by the ethics committee of the institutional review board of the Keelung Chang Gung Memorial Hospital.

2.2. Definitions. CKD were defined by the National Kidney Foundation: K/DOQI classification for CKD and were determined as having persistent proteinuria or a decreased eGFR of less than 60 mL/min/1.73 m², determined by the abbreviated Modification of Diet in Renal Disease equation [23]. Serum Cr was assessed by spectrophotometric analysis using a modified kinetic Jaffe reaction with standardization of the creatinine calibration to an isotope dilution mass spectrometry reference measurement procedure. Proteinuria was determined if urine albumin-to-creatinine ratio > 30 g/g or urine protein-to-creatinine ratio > 150 g/g. Diabetes mellitus was defined as a fasting glucose level ≥ 126 mg/dL or use of any hypoglycemic medication. Hypertension was considered present if the patient received medical therapy for such a condition or if blood pressure was > 140/90 mmHg. Hypercholesterolemia was defined as a total cholesterol level ≥ 200 mg/dL. Smoking and alcohol drinking indicated any sustained past or current behaviors. Metabolic syndrome was defined according to the Adult Treatment Panel III criteria as the presence of at least three of the following five traits: visceral (abdominal) obesity, determined on the basis of the Asian waist circumference cut-offs (men: > 90 cm, women: > 80 cm); blood pressure > 130/85 mmHg or drug treatment for essential hypertension; serum high-density lipoprotein cholesterol (HDL-C) level < 40 mg/dL (1 mmol/L) in men and < 50 mg/dL (1.3 mmol/L) in women or drug treatment for low HDL-C; serum triglycerides (TG) level > 150 mg/dL (1.7 mmol/L) or drug treatment for elevated TG; and fasting plasma glucose level > 100 mg/dL (5.6 mmol/L) or drug treatment for DM. Obesity was evaluated according to the WHO classification as having a body mass index of 30 kg/m² or more [24]. Serum concentrations of 25 (OH) vitamin D were measured using a radioimmunoassay (Vitamin D Total, Roche Diagnostics, Mannheim, Germany) according to the manufacturer’s instructions. Vitamin D status was defined as “deficient” (< 20 ng/mL), “insufficient” (20–30 ng/mL), and “sufficient” (> 30 ng/mL) [25]. Hypocalcemia was defined as corrected Ca less than 8.5 mg/dL; hyperphosphatemia, p greater than 4.5 mg/dL, or phosphate binder use and hyperparathyroidism, intact parathyroid hormone more than twice the upper limit of normal, corresponding to 70 pg/mL [26]. Because of the cross-sectional nature of the study and to avoid misdiagnosis of CKD, we used two surrogate indices of CKD (eGFR < 60 mL/min and proteinuria) to establish the association between the biomarker of interest and the outcome.

2.3. Statistical Methods. Descriptive statistics were expressed as mean ± standard deviation, median, range, or percentage frequency, as appropriate. All variables were tested for normal distribution by Kolmogorov-Smirnov test. Data were log-transformed to approximate normal distribution. Student’s t-test or Mann–Whitney U test was applied to compare the mean of continuous variables. Categorical data were tested using the Chi-square test. Pearson or Spearman correlation coefficients were appropriately used to test the correlation between serum 25 (OH) vitamin D, intact PTH, and calcium and phosphate levels with eGFR or proteinuria. Logistic regression analysis was applied to identify the association between these variables with the outcome of interest, after adjusting for potential confounders, such as age or gender. Conditional logistic regression analysis was performed to
with eGFR. Logistic regression analysis identified the log intact PTH as an independent factor associated with CKD [crude: odd ratios (OR), 1.985; 95% confidential interval (CI), 1.663–2.371, \( p < 0.001 \); model 1 (adjusted for age and gender): OR, 1.618; 95% CI, 1.345–1.948, \( p < 0.001 \); model 2 (adjusted for age, gender, 25 (OH) vitamin D, calcium, and phosphate): OR, 1.796; 95% CI, 1.479–2.181, \( p < 0.001 \)]. The association of intact PTH with CKD was consistent when assessed in a categorical fashion, as hyperphosphatemia [crude: OR, 2.530; 95% CI, 2.057–3.112, \( p < 0.001 \); model 1 (adjusted for age and gender): OR, 2.074; 95% CI, 1.662–2.589, \( p < 0.001 \); model 2 (adjusted for age, gender, 25 (OH) vitamin D status, hypocalcemia, and hyperphosphatemia): OR, 2.128, 95% CI, 1.699–2.667, \( p < 0.001 \) (Table 3)]. Again, log intact PTH was an independent factor associated with proteinuria [crude: OR, 1.744, 95% CI, 1.442–2.109, \( p < 0.001 \); model 1 (adjusted for age and gender): OR, 1.477, 95% CI, 1.217–1.793, \( p < 0.001 \); model 2 (adjusted for age, gender, 25 (OH) vitamin D status, hypocalcemia, and hyperphosphatemia): OR, 1.579, 95% CI, 1.291–1.932, \( p < 0.001 \)]. The association of intact PTH with proteinuria was consistent when assessed in a categorical fashion, as hyperphosphatemia [crude: OR, 2.212, 95% CI, 1.770–2.766, \( p < 0.001 \); model 1 (adjusted for age and gender): OR, 1.865, 95% CI, 1.480–2.351, \( p < 0.001 \); model 2 (adjusted for age, gender, 25 (OH) vitamin D status, hypocalcemia, and hyperphosphatemia): OR, 1.895, 95% CI, 1.497–2.398, \( p < 0.001 \) (Table 4)]. The associations of serum 25 (OH) vitamin D, calcium, and phosphate levels with CKD or proteinuria were not significant after adjusting for confounder or analysis in a categorical manner (Tables 3 and 4).

To control as much as possible the confounding effect of baseline characteristics of patients on the outcome of study, we have resampled a subset of patients from stratified sampling by CKD stage with individualized match to age, gender, presence of hypertension, or metabolic syndrome. Stratified sampling by CKD stage identified 464 CKD patients with their matched counterparts (\( n = 928 \) normals). Only 1392 patients were included in the subset analysis after individualized matched pair of \( \pm 1 \) y/o of age. The mean age of resampling subset was 61.0 \( \pm \) 10.9 years and 39% of them were men. Hypertension was present in 73% and metabolic syndrome was present in 43% of the resampling population. Both the log intact PTH and hyperparathyroidism remained a significant risk factor for eGFR \( \leq 60 \) mL/min and albuminuria, after adjustment for age, gender, serum 25 (OH) vitamin D, and calcium or phosphate levels (Table 5).

### 4. Discussion

This community-based study found that the serum PTH levels were inversely correlated with eGFR but positively correlated with albuminuria. Either the increase of serum PTH levels or the presence of hyperparathyroidism was independently associated with the risk of CKD, after adjustments for confounders or by using the resampling subset of comparable baseline characteristics. The serum PTH levels increased in the early stage of CKD, before the change of 25 (OH) vitamin D-calcium-phosphate axis, and may serve as a potential risk factor for CKD.
Figure 2: Correlation of serum 25 (OH) vitamin D, log intact PTH, and calcium and phosphate with eGFR and log albuminuria-to-creatinine ratio.
Table 1: Demographic characteristics of all patients and stratified by CKD (n = 4080).

<table>
<thead>
<tr>
<th></th>
<th>All (n = 4080)</th>
<th>Non-CKD (n = 3273)</th>
<th>CKD (n = 807)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>58.4 ± 13.3</td>
<td>56.5 ± 12.6</td>
<td>66.1 ± 12.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Male, number (%)</td>
<td>1480 (36.3%)</td>
<td>1134 (34.6%)</td>
<td>346 (42.9%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Diabetes, number (%)</td>
<td>722 (17.7%)</td>
<td>410 (12.5%)</td>
<td>312 (38.7%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hypertension, number (%)</td>
<td>2197 (53.8%)</td>
<td>1607 (49.3%)</td>
<td>590 (73.2%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Metabolic syndrome, number (%)</td>
<td>1277 (31.3%)</td>
<td>863 (26.4%)</td>
<td>414 (51.3%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Obesity, number (%)</td>
<td>383 (9.4%)</td>
<td>264 (8.1%)</td>
<td>119 (14.7%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Physical activity, min/day</td>
<td>12.9 (0, 660)</td>
<td>12.9 (0, 660)</td>
<td>17.1 (0, 600)</td>
<td>0.137</td>
</tr>
<tr>
<td>Smoking, number (%)</td>
<td>1032 (25.3%)</td>
<td>806 (24.6%)</td>
<td>226 (28.0%)</td>
<td>0.048</td>
</tr>
<tr>
<td>Alcohol drinking, number (%)</td>
<td>642 (15.7%)</td>
<td>534 (16.3%)</td>
<td>108 (13.4%)</td>
<td>0.040</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>24.9 ± 3.8</td>
<td>24.6 ± 3.7</td>
<td>26.0 ± 3.8</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Systolic BP, mmHg</td>
<td>131.9 ± 19.4</td>
<td>129.6 ± 18.5</td>
<td>141.3 ± 20.1</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Diastolic BP, mmHg</td>
<td>78.5 ± 11.9</td>
<td>77.7 ± 11.5</td>
<td>81.9 ± 12.8</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Laboratory</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>eGFR, mL/min per 1.73 m² (MDRD)</td>
<td>94.1 ± 26.3</td>
<td>98.3 ± 22.6</td>
<td>77.1 ± 32.6</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>BUN, mg/dL</td>
<td>13.7 ± 5.5</td>
<td>12.7 ± 3.7</td>
<td>17.5 ± 8.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Serum creatinine, mg/dL</td>
<td>0.8 ± 0.3</td>
<td>0.7 ± 0.2</td>
<td>1.0 ± 0.5</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Serum albumin, g/dL</td>
<td>4.7 ± 0.3</td>
<td>4.7 ± 0.3</td>
<td>4.6 ± 0.3</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Cholesterol, mg/dL</td>
<td>209.7 ± 39.2</td>
<td>210.1 ± 37.4</td>
<td>208.2 ± 45.8</td>
<td>0.209</td>
</tr>
<tr>
<td>Triglycerides, mg/dL</td>
<td>124.4 ± 112.4</td>
<td>121.4 ± 103.3</td>
<td>156.5 ± 148.1</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>hs-CRP, mg/L</td>
<td>1.0 (0.2, 211.4)</td>
<td>0.9 (0.2, 104.1)</td>
<td>1.6 (0.2, 211.4)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Urine albumin-to-creatinine ratio, g/g</td>
<td>6.5 (0.4, 11067.4)</td>
<td>5.4 (0.6, 29.8)</td>
<td>49.7 (0.4, 11067.4)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>25 (OH) vitamin D, ug/mL</td>
<td>29.4 ± 9.4</td>
<td>29.0 ± 9.0</td>
<td>31.1 ± 10.5</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>iPTH, pmol/L</td>
<td>43.0 (3.0, 898.9)</td>
<td>42.2 (3.0, 312.2)</td>
<td>46.3 (8.3, 898.9)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Serum calcium, mg/dL</td>
<td>9.4 ± 0.3</td>
<td>9.4 ± 0.3</td>
<td>9.4 ± 0.4</td>
<td>0.055</td>
</tr>
<tr>
<td>Corrected calcium, mg/dL</td>
<td>8.8 ± 0.3</td>
<td>8.8 ± 0.3</td>
<td>8.9 ± 0.4</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Serum phosphate, mg/dL</td>
<td>3.8 ± 0.5</td>
<td>3.8 ± 0.5</td>
<td>3.8 ± 0.6</td>
<td>0.003</td>
</tr>
<tr>
<td>25 (OH) vitamin D status</td>
<td>1383 (33.9%)</td>
<td>1042 (31.8%)</td>
<td>341 (42.3%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Vitamin D sufficient, number (%)</td>
<td>1214 (52.1%)</td>
<td>1755 (53.6%)</td>
<td>369 (45.7%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Vitamin D deficient, number (%)</td>
<td>573 (14.0%)</td>
<td>476 (14.5%)</td>
<td>97 (12.0%)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

The values are expressed as means (SD) or median (min, max).
Corrected calcium = serum calcium + 0.8 * (4 – serum albumin).

Table 2: Serum 25 (OH) vitamin D status stratified by eGFR and albuminuria-to-creatinine ratio.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Vitamin D sufficient, number (%)</th>
<th>Vitamin D deficient, number (%)</th>
<th>Vitamin D insufficient, number (%)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean eGFR, mL/min</td>
<td></td>
<td></td>
<td></td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>eGFR &gt; 90 mL/min</td>
<td>607 (27.2%)</td>
<td>1227 (55.0%)</td>
<td>398 (17.8%)</td>
<td></td>
</tr>
<tr>
<td>eGFR 90–60 mL/min</td>
<td>619 (40.5%)</td>
<td>772 (50.5%)</td>
<td>139 (9.1%)</td>
<td></td>
</tr>
<tr>
<td>eGFR 60–45 mL/min</td>
<td>118 (54.1%)</td>
<td>79 (36.2%)</td>
<td>21 (9.6%)</td>
<td></td>
</tr>
<tr>
<td>eGFR &lt; 45 mL/min</td>
<td>39 (39.0%)</td>
<td>46 (46%)</td>
<td>15 (15%)</td>
<td></td>
</tr>
<tr>
<td>Mean ACR ratio, g/g</td>
<td></td>
<td></td>
<td></td>
<td>0.2586</td>
</tr>
<tr>
<td>ACR &lt; 30</td>
<td>1147 (32.8%)</td>
<td>1851 (53.0%)</td>
<td>497 (14.2%)</td>
<td></td>
</tr>
<tr>
<td>ACR 30–300</td>
<td>203 (43.5%)</td>
<td>220 (47.1%)</td>
<td>44 (9.4%)</td>
<td></td>
</tr>
<tr>
<td>ACR &gt; 300</td>
<td>33 (28.0%)</td>
<td>53 (44.9%)</td>
<td>32 (27.1%)</td>
<td></td>
</tr>
</tbody>
</table>

eGFR: estimated glomerular filtration rate; ACR: urine albumin-to-creatinine ratio.
### Table 3: OR of variables associated with eGFR less than 60 mL/min.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Crude OR (95% CI)</th>
<th>p</th>
<th>Model 1 OR (95% CI)</th>
<th>p</th>
<th>Model 2 OR (95% CI)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Continuous variables</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>25 (OH) vitamin D, ug/mL</td>
<td>1.023 (1.015–1.031)</td>
<td>&lt;0.001</td>
<td>1.001 (0.992–1.010)</td>
<td>0.795</td>
<td>1.005 (0.996–1.015)</td>
<td>0.276</td>
</tr>
<tr>
<td>Log intact PTH, pmol/L</td>
<td>1.985 (1.663–2.371)</td>
<td>&lt;0.001</td>
<td>1.618 (1.345–1.948)</td>
<td>&lt;0.001</td>
<td>1.796 (1.479–2.181)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Calcium, mg/dL</td>
<td>1.276 (1.020–1.597)</td>
<td>0.033</td>
<td>1.475 (1.168–1.862)</td>
<td>0.001</td>
<td>1.617 (1.270–2.060)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Phosphate, mg/dL</td>
<td>0.802 (0.694–0.926)</td>
<td>0.003</td>
<td>1.086 (0.919–1.283)</td>
<td>0.332</td>
<td>1.138 (0.959–1.350)</td>
<td>0.137</td>
</tr>
<tr>
<td><strong>Categorical variables</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vitamin D sufficient (yes vs no)</td>
<td>1</td>
<td></td>
<td>1</td>
<td></td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Vitamin D deficient (yes vs no)</td>
<td>0.642 (0.544–0.758)</td>
<td>&lt;0.001</td>
<td>0.872 (0.662–1.149)</td>
<td>0.330</td>
<td>0.959 (0.725–1.269)</td>
<td>0.771</td>
</tr>
<tr>
<td>Vitamin D insufficient (yes vs no)</td>
<td>0.623 (0.485–0.800)</td>
<td>&lt;0.001</td>
<td>0.726 (0.558–0.944)</td>
<td>0.017</td>
<td>0.777 (0.595–1.014)</td>
<td>0.063</td>
</tr>
<tr>
<td>Hyperparathyroidism (yes vs no)</td>
<td>2.530 (2.057–3.112)</td>
<td>&lt;0.001</td>
<td>2.074 (1.662–2.589)</td>
<td>&lt;0.001</td>
<td>2.188 (1.699–2.667)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hypocalcemia (yes vs no)</td>
<td>2.286 (2.330–3.272)</td>
<td>&lt;0.001</td>
<td>2.074 (1.662–2.589)</td>
<td>&lt;0.001</td>
<td>2.188 (1.699–2.667)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

OR: odd ratios; CI: confidence interval.
Model 1: adjusted for age and gender.
Model 2: * adjusted for age, gender, and all other continuous variables; ‡ adjusted for age, gender, and all other categorical variables.

### Table 4: OR of variables associated with proteinuria.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Crude OR (95% CI)</th>
<th>p</th>
<th>Model 1 OR (95% CI)</th>
<th>p</th>
<th>Model 2 OR (95% CI)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Continuous variables</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>25 (OH) vitamin D, ug/mL</td>
<td>1.016 (1.008–1.025)</td>
<td>&lt;0.001</td>
<td>1.000 (0.991–1.010)</td>
<td>0.941</td>
<td>1.004 (0.994–1.014)</td>
<td>0.457</td>
</tr>
<tr>
<td>Log intact PTH, pmol/L</td>
<td>1.744 (1.442–2.109)</td>
<td>&lt;0.001</td>
<td>1.477 (1.217–1.793)</td>
<td>&lt;0.001</td>
<td>1.579 (1.291–1.932)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Calcium, mg/dL</td>
<td>1.159 (0.907–1.481)</td>
<td>0.237</td>
<td>1.252 (0.980–1.599)</td>
<td>0.072</td>
<td>1.330 (1.037–1.705)</td>
<td>0.025</td>
</tr>
<tr>
<td>Phosphate, mg/dL</td>
<td>0.865 (0.740–1.012)</td>
<td>0.069</td>
<td>1.070 (0.897–1.276)</td>
<td>0.452</td>
<td>1.118 (0.935–1.338)</td>
<td>0.222</td>
</tr>
<tr>
<td><strong>Categorical variables</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vitamin D sufficient (yes vs no)</td>
<td>1</td>
<td></td>
<td>1</td>
<td></td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Vitamin D deficient (yes vs no)</td>
<td>1.386 (1.060–1.813)</td>
<td>0.017</td>
<td>0.887 (0.666–1.183)</td>
<td>0.416</td>
<td>0.963 (0.720–1.289)</td>
<td>0.801</td>
</tr>
<tr>
<td>Vitamin D insufficient (yes vs no)</td>
<td>0.971 (0.747–1.263)</td>
<td>0.826</td>
<td>0.756 (0.576–0.992)</td>
<td>0.044</td>
<td>0.802 (0.609–1.056)</td>
<td>0.115</td>
</tr>
<tr>
<td>Hyperparathyroidism (yes vs no)</td>
<td>2.212 (1.770–2.766)</td>
<td>&lt;0.001</td>
<td>1.865 (1.480–2.351)</td>
<td>&lt;0.001</td>
<td>1.895 (1.497–2.398)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hypocalcemia (yes vs no)</td>
<td>1.332 (0.542–3.272)</td>
<td>0.532</td>
<td>1.109 (0.436–2.821)</td>
<td>0.827</td>
<td>0.821 (0.320–2.107)</td>
<td>0.682</td>
</tr>
<tr>
<td>Hyperphosphatemia (yes vs no)</td>
<td>0.992 (0.759–1.298)</td>
<td>0.955</td>
<td>1.201 (0.909–1.587)</td>
<td>0.198</td>
<td>1.249 (0.943–1.653)</td>
<td>0.120</td>
</tr>
</tbody>
</table>

OR: odd ratios; CI: confidence interval.
Model 1: adjusted for age and gender.
Model 2: * adjusted for age, gender, and all other continuous variables; ‡ adjusted for age, gender, and all other categorical variables.

Optimal serum levels of vitamin D in healthy subjects and CKD patients are not well understood. Growing evidence indicated the relationship between abnormal vitamin D levels and metabolic syndrome, diabetes, cancer, and chronic diseases; however, the association of vitamin D and the risk of CKD remains controversial [27]. Our findings that serum 25 (OH) vitamin D levels increased with the decline of eGFR and the insignificant association with CKD are consistent with some studies [16, 18, 19], but not with others [28, 29]. Guessous et al. found that the prevalence of serum vitamin D levels and deficiency status were similar in CKD and non-CKD subjects in a cross-sectional Swiss population-based study [18]. They did not find any significant association of serum vitamin D levels with incident CKD, incident albuminuria, or rapid renal function decline, after a mean follow-up of 5.5 years [19]. On the other hand,
findings from the 10-year Prevention of Renal and Vascular End-Stage Disease prospective cohort demonstrated that low plasma vitamin D was associated with the risk of developing albuminuria rather than reduced eGFR in high sodium intake individuals [28]. However, investigation on normal renal function subjects found that high serum vitamin D levels were independently associated with high serum creatinine and recalled the potential role of muscle size as a contributing factor to this elevation [29]. Several factors can influence serum levels of vitamin D, including analytic method, seasonal variation, latitude, air pollution, sun exposure, and physical activity [30]. Furthermore, meta-analyses to evaluate the concentration of vitamin D and health outcome did not show convincing data for the association of this biomarker with renal outcome in healthy or CKD patients [27].

The finding that serum PTH levels and hyperparathyroidism were independent risk factors for CKD and its surrogates (eGFR and proteinuria) was novel. Molecularly, parathyroid hormone is considered as procalcific and profibrotic through promoting mRNA and protein expression of the receptor of advanced glycation end products (RAGE) and interleukin 6, enhancing abnormal calcium-phosphate homeostasis and renin-angiotensinogen-aldosterone hyperactivity; even some have advocated its possible roles as cardiovascular or uremic toxins [31–33]. On the other hand, treatment of teriparatide (human PTH) in low-density lipoprotein receptor −/− mice exhibited induction of osseous osteopontin expression and serum osteopontin levels, indicating inhibition of vascular calcification and aortic osteogenic differentiation. The study suggested possible beneficial actions of PTH at early stages of macrovascular disease in responses to diabetes and dyslipidemia [34]. Clinically, a Swedish population-based study showed that normocalcemic, vitamin D sufficient hyperparathyroidism was common and indolent in a long-term follow-up of 17 years. This condition triggered low morbidities and had no association with creatinine at follow-up [35]. However, data of the Germany Calcific Uraemic Arteriolopathy Registry indicated development of calciphylaxis in dialysis patients with mean PTH levels of 147 (IQR: 72–276) pg/mL [36]. In spite of the promising results of our study, caution in the interpretation of PTH as a powerful biomarker should be taken into account, including intermethod and interpersonal variability, target metabolite to measure, vitamin D status, baseline renal function, age, gender, menopausal status, body mass index, race, and end-organ hyporesponsiveness [37–40]. Further studies are needed to demonstrate the exact relationship between high intact PTH and development of CKD in the general population.

A peculiar finding of the present study was the elevated odd ratio of having eGFR < 60 mL/min associated with serum calcium levels, even after considering confounders. However, this association was no longer existent when the serum calcium level was assessed in a categorical manner. Physiologically, serum calcium levels decrease with the decline of eGFR. This phenomenon appears in the late stage of CKD and contributes in part to the hyperparathyroidism cascade leading to the development of CKD-bone mineral disease and adverse patient outcomes. Conversely, high serum calcium levels promote neointimal calcification and increase cardiovascular events and mortality. The paradoxical role of serum calcium levels was rarely explored in healthy or early stage CKD patients and is unclear. Association of high corrected serum calcium with both the number of the metabolic syndrome components and the number of nonconventional cardiometabolic risk factors (uric acid, homocystein, and gamma-glutamytransferase) was observed in Caucasian people, independently of the metabolic syndrome and body mass index [41]. A French multicentric study found that increased serum calcium concentration was independently and positively associated with high pulse pressure and hypertension [42], an important risk factor for CKD. Putting all these lines of evidence together, the exact impact of serum calcium levels on the renal functions of healthy people deserves further study.

### Table 5: Conditional logistic regression analysis in resampling subset of patients.

<table>
<thead>
<tr>
<th>Variable</th>
<th>eGFR ≤ 60 mL/min/1.73 m²</th>
<th>Proteinuria</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Adjusted OR (95% CI)</td>
<td>p</td>
</tr>
<tr>
<td><strong>Continuous variables</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>25 (OH) vitamin D, ug/mL</td>
<td>1.012 (0.999–1.025)</td>
<td>0.079</td>
</tr>
<tr>
<td>Log intact PTH, pmol/L</td>
<td>1.613 (1.240–2.099)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Calcium, mg/dL</td>
<td>1.421 (1.003–2.04)</td>
<td>0.048</td>
</tr>
<tr>
<td>Phosphate, mg/dL</td>
<td>1.155 (0.913–1.463)</td>
<td>0.23</td>
</tr>
<tr>
<td><strong>Categorical variables</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vitamin D sufficient (yes versus no)</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Vitamin D deficient (yes versus no)</td>
<td>1.323 (0.896–1.953)</td>
<td>0.16</td>
</tr>
<tr>
<td>Vitamin D insufficient (yes versus no)</td>
<td>1.494 (1.442–2.758)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hypocalcemia (yes versus no)</td>
<td>1.155 (1.003–2.04)</td>
<td>0.048</td>
</tr>
<tr>
<td>Hyperparathyroidism (yes versus no)</td>
<td>1.155 (0.913–1.463)</td>
<td>0.23</td>
</tr>
</tbody>
</table>

OR: odd ratios; CI: confidence interval; eGFR: estimated glomerular filtration rate.

* Adjusted for age, gender, and all other continuous variables. ** Adjusted for age, gender, and all other categorical variables.
The strength of our study was in the use of a large number of participants with high range for renal function, simultaneous measurement of biochemistry parameters that minimize bias in the variability of analytic testing, and exclusion of exogenous vitamin D use. The use of subset sampling analysis of comparable characteristics has avoided confounding effects from baseline data on the study outcome. However, some limitations of study should be addressed. First, findings from the cross-sectional design study should not be interpreted in casual terms. Second, all participants of the study came from the northeastern region of Taiwan (Keelung and its neighboring areas) and had unique geographic (latitude, 25° N), cultural, and dietary characteristics that limited generalizability of the present findings to other populations. Finally, important drivers of renal calcium-phosphorus handling, such as FGF-23 and Klotho, were not measured in the present study. Primary hyperparathyroidism could not be excluded completely, because parathyroid images were not available. Universal consensus on the target fragments, analytic method, and reference value of measurement for these two biomarkers is still in debate. Further investigation should be warranted to elucidate the interplay of FGF-23, Klotho, vitamin D, and parathyroid axis with the risk of CKD.

In conclusion, this community-based study confirmed the association of serum levels of PTH and hyperparathyroidism with the risk of CKD. The serum levels of PTH increased in parallel with albuminuria but inversely with eGFR. Using a sample of the general Taiwanese population, the current study could provide a description of the prevalence of elevated serum intact PTH levels across a broad spectrum of kidney functions. This significant association with low eGFR was independent of dietary intake of calcium and serum levels of calcium, phosphorus, and 25 (OH) D. Given its association with adverse outcomes in CKD, reducing serum intact PTH levels may be an important goal for improving CKD-MBD and may be potentially helpful for CKD prevention. The findings of the present study offer an insight for a further well-designed prospective study to clarify the causal relationship of PTH and 25 (OH) vitamin D with renal function and risk of CKD in the general population.

Competing Interests

The authors declare no competing interests regarding the publication of this paper.

Acknowledgments

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Hyperphosphatemia and hs-CRP Initiate the Coronary Artery Calcification in Peritoneal Dialysis Patients

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Background. Coronary artery calcification (CAC) contributes to high risk of cardiocerebrovascular diseases in dialysis patients. However, the risk factors for CAC initiation in peritoneal dialysis (PD) patients are not known clearly. Methods. Adult patients with baseline CaCS = 0 and who were followed up for at least 3 years or until the conversion from absent to any measurable CAC detected were included in this observational cohort study. Binary logistic regression was performed to identify the risk factors for CAC initiation in PD patients. Results. 70 patients recruited to our study were split into a noninitiation group (n = 37) and an initiation group (n = 33) according to the conversion of any measurable CAC during their follow-up or not. In univariate analysis, systolic blood pressure, serum phosphorus, fibrinogen, hs-CRP, serum creatinine, and triglycerides were positively associated with the initiation of CAC, while the high density lipoprotein and nPCR didtheoppositefunction. Multivariate analysis revealed that hyperphosphatemia and hs-CRP were the independent risk factors for CAC initiation after adjustments. Conclusions. Hyperphosphatemia and hs-CRP were the independent risk factors for CAC initiation in PD patients. These results suggested potential clinical strategies to prevent the initiation of CAC in PD patients.

1. Introduction

Cardiovascular events (CVEs) are the leading cause of death in patients on peritoneal dialysis (PD). Traditional risk factors like hypertension, diabetes, hyperlipideamia, and male gender cannot explain the abnormally high incidence of CVD in ESRD patients [1, 2]. Compared to the general population, the chronic kidney disease (CKD) and dialysis patients were characterized by mineral metabolism disorder, oxidative stress, and a poor nutritional state, resulting in more prevalent and markedly more severe vascular calcification.

Vascular calcification can occur in the arterial intimal layer in association with atherosclerosis, or in the arterial medial layer independent of atherosclerotic disease. Recently, though none of the methods can reliably distinguish between atherosclerotic and medial calcification, vascular calcification, especially the coronary artery calcification score (CaCS) assessed by the computerized tomography [3], has been reported to be an independent predictor of all-cause mortality and CVEs in CKD and dialysis patients [4–6]. A major area of interest concerns the reasons behind the development and accelerated progression of CaCS in patients with ESRD. In vitro and in vivo, HDL inhibits the osteogenic differentiation pathway [7], while phosphate will induce arterial calcification in a dose-dependent manner, which is also associated with the upregulation of proteins involved in bone formation and the phenotypic differentiation [8, 9]. In dialysis patients, age, hypercalcemia, hyperphosphatemia, PD duration, hyperlipidemia, and inflammation are considered to be related to the CAC progression [10–16].

More interested, the conversion from no CAC to any CAC reflects an important step of the disease process. In general population with zero CAC at baseline, revealing age, LDL cholesterol, systolic blood pressure, and current smoking were independent predictors of CAC onset [16, 17]. However, there were few clinical studies about the risk factors to
initiate the CAC in PD patients. Accordingly, we performed a prospective study of 70 patients with zero CAC at baseline in order to identify the initiator of CAC in PD patients.

2. Methods and Materials

2.1. Study Population. Adult PD patients treated at division of nephrology, Huashan Hospital Fudan University in China from June 2004 to march 2013 were recruited in this observational cohort study. They received regular PD treatment and underwent a series of coronary artery calcification score (CaCS) measurements by MSCT at baseline and semiannual or annual repeat scans during the follow-up. The patients with baseline CaCS = 0 and were followed up for at least 3 years or until the conversion from absent to any measurable CAC were eligible for the present analysis. The demographic characteristics, laboratory test data, and adequacy of PD were collected. Binary logistic regression was performed to identify the initiators of CAC in PD patients.

Patients were excluded: the CAC at baseline were not zero; the follow-up time of patients with the CAC remaining at zero were less than 3 years due to various reasons. All the participants provided their written informed consent, and the protocol of the study was approved by the ethics committee of Huashan Hospital at Fudan University.

2.2. Data Collection and Statistical Analysis. The recruited PD patients were divided into two groups, noninitiation and initiation group, according to their follow-up of whether CaCS remained at zero or not. The CaCS was recorded as just as we previous described [6, 14]. Demographic characteristics and comorbidities (diabetes mellitus, hypertension, and CVD) were recorded at baseline. Laboratory measurements, such as calcium-phosphate metabolism, lipids and inflammation markers, were collected every 3 months. PD adequacy was evaluated every 6 months using Baxter PD Adequest 2.0 software (Baxter Healthcare Corporation, Deerfield, IL, USA). The average values of these indexes during the total follow-up time in noninitiation group were calculated, while in initiation group the average was acquired within one year before any measurable CAC.

Statistical analysis of the data was performed using SPSS software, version 17.0. The variables expression and the statistical test were described in our previous study [14]. Binary logistic regression was used to identify the independent risk factors for CAC initiation in PD patient.

3. Results

3.1. Patients’ Characteristics. Of 550 PD patients in our PD center, 112 (21.33%) did not show any CaCS at the baseline. But as of March 2016, among these patients, only 70 patients (54.4 ± 13.4 years old, 36 men) whose baseline CaCS = 0, with a follow-up longer than 36 months or any detected CAC, were considered for the analysis. The dialysate volume was 6.8 ± 1.4 L every day and the long and short dwelling times were 12 hours and 4 to 6 hours, respectively. All patients used the Dianeeal PD4 (1.25 mmol/L calcium, 2 L) and Dianeeal PD2 (1.75 mmol/L calcium, 2 L) from Baxter Healthcare Corporation. No icodextrin was used because it is not available in China.

Among the 70 patients with a mean follow-up of 54.0 ± 16.7 months, 4 patients (5.71%) showed the CaCS in the first follow-up year, 7 (10%) in the second year, 11 (15.71%) in the third year, 4 (5.71%) in the fourth year, and 7 (10%) in later until March 2016, and they were grouped into the initiation group, and the other 37 (52.86%) remaining at zero CaCS during the follow-up time were grouped into the noninitiation group (Figure 1). And in the initiation group, the initiation CaCS, which was defined as the first detected quantitative CaCS without zero (CACS-initiated), was 29.6 (16.6, 100.7). Among these patients, 10 (30.3%) had the CaCS-initiated more than 100 and 19 (57.6%) less than 50 (Figure 2).
Table 1: Clinical characteristics of the peritoneal dialysis patients.

<table>
<thead>
<tr>
<th>Coronary artery calcification</th>
<th>Noninitiation (n = 37)</th>
<th>Initiation (n = 33)</th>
<th>Total (n = 70)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>53.9 ± 12.0</td>
<td>54.9 ± 15.1</td>
<td>54.4 ± 13.4</td>
<td>0.749</td>
</tr>
<tr>
<td>Gender (male)</td>
<td>45.9%</td>
<td>57.6%</td>
<td>51.4%</td>
<td>0.335</td>
</tr>
<tr>
<td>BMI (kg/m^2)</td>
<td>22.5 ± 3.6</td>
<td>24.0 ± 3.5</td>
<td>23.2 ± 3.6</td>
<td>0.087</td>
</tr>
<tr>
<td>Smoking</td>
<td>23.5%</td>
<td>41.9%</td>
<td>32.30%</td>
<td>0.116</td>
</tr>
<tr>
<td>Cause of ESRD</td>
<td></td>
<td></td>
<td></td>
<td>0.241</td>
</tr>
<tr>
<td>Glomerulonephritis</td>
<td>89.2%</td>
<td>78.8%</td>
<td>84.30%</td>
<td></td>
</tr>
<tr>
<td>Diabetes mellitus</td>
<td>8.1%</td>
<td>15.2%</td>
<td>11.40%</td>
<td></td>
</tr>
<tr>
<td>Others</td>
<td>2.7%</td>
<td>6.1%</td>
<td>4.30%</td>
<td></td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>125 ± 20</td>
<td>136 ± 21</td>
<td>130 ± 21</td>
<td>0.030</td>
</tr>
<tr>
<td>DBP (mmHg)</td>
<td>80 ± 11</td>
<td>82 ± 12</td>
<td>81 ± 12</td>
<td>0.322</td>
</tr>
<tr>
<td>CAPD</td>
<td>91.9%</td>
<td>93.9%</td>
<td>92.90%</td>
<td>0.742</td>
</tr>
<tr>
<td>CVD</td>
<td>8.1%</td>
<td>24.2%</td>
<td>15.70%</td>
<td>0.066</td>
</tr>
<tr>
<td>All-cause deaths</td>
<td>2.7%</td>
<td>15.2%</td>
<td>8.60%</td>
<td>0.065</td>
</tr>
<tr>
<td>Follow-up time (months)</td>
<td>52.2 ± 17.2</td>
<td>55.9 ± 16.2</td>
<td>54.0 ± 16.7</td>
<td>0.351</td>
</tr>
<tr>
<td>Baseline duration of PD (months)</td>
<td>0.2 (−0.4, 2.6)</td>
<td>0.13 (−0.3, 6.3)</td>
<td>0.2 (−0.3, 4.3)</td>
<td>0.638</td>
</tr>
<tr>
<td>CACS-initiated</td>
<td>0 (0, 0)</td>
<td>29.6 (16.6, 100.7)</td>
<td>0.9 (0, 25.9)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Calcitrol</td>
<td>83.8%</td>
<td>54.5%</td>
<td>70.0%</td>
<td>0.008</td>
</tr>
<tr>
<td>Statin</td>
<td>62.2%</td>
<td>42.4%</td>
<td>52.9%</td>
<td>0.101</td>
</tr>
<tr>
<td>Calcium carbonate</td>
<td>75.7%</td>
<td>69.7%</td>
<td>72.90%</td>
<td>0.577</td>
</tr>
</tbody>
</table>

BMI: body mass index; SBP: systolic blood pressure; DBP: diastolic blood pressure; CAPD: continuous ambulatory peritoneal dialysis; CVD: cardiovascular disease; CACS-initiated: the CACS we detected firstly when coronary artery calcification occurred.

The follow-up time of initiation group (19 men, 57.6%) was 55.9 ± 16.2 months; similarly the noninitiation group (17 men, 45.9%) was 52.2 ± 17.2 months. There was weak difference in the history of smoking (9/37 versus 14/33, p = 0.116) and BMI (p = 0.087) between the two groups. The systolic blood pressure (SBP) was higher in the initiation group (p = 0.030), while the diastolic blood pressure (DBP) was not (p = 0.322). There were no significant differences in the baseline duration of PD therapy between the groups (p = 0.638). The glucose reabsorption (p = 0.144) and the level of 25-OH-VitD (p = 0.59) between the groups were also similar. Sixty-six patients (94.3%) received continuous ambulatory peritoneal dialysis, while others received daytime ambulatory peritoneal dialysis. During the follow-up period, 6 patients died (1 in noninitiation group versus 5 in initiation group, p = 0.065) and 11 experienced CVEs (3 in noninitiation group versus 8 in initiation group, p = 0.066) (Tables 1 and 2).

In total, 45 (64.3%) patients had an average residual renal Ccr of >20 L/week and 46 (65.7%) had an average total Ccr of >60 L/week. Twenty-seven (38.6%) patients had a serum phosphate level of >5.5 mg/dL, and 52 (74.3%) had a level of >4.5 mg/dL. Among the included patients, 51 patients (72.9%) received calcium carbonate treatment (29/37 versus 23/33; p = 0.577) and 49 received calcitrol treatment (31/37 versus 18/33; p = 0.008). Else, twenty-three (62.2%) patients take the lower-lipid agent to modify the lipids in noninitiation group and fourteen (42.4%) in initiation group (23/37 versus 14/33, p = 0.101) (Tables 1 and 2).

3.2. Hyperphosphatemia and hs-CRP as Independent Risk Factors for CAC Initiation. The two groups were weakly different in BMI (p = 0.087), uric acid (UA) (p = 0.087), Homa-IR (p = 0.105), and residual CCR (p = 0.056), while they were similar in hemoglobin, AKP, ferritin, cholesterol, LDL, iPTH, and adjusted calcium. The univariate analysis showed that SBP (p = 0.030), hs-CRP (p = 0.005), fibrinogen (p = 0.019), serum creatinine (p = 0.011), triglycerides (p = 0.027), and serum phosphorus (p < 0.001) were the potential risk factors for CAC initiation in PD patients, while the HDL (p < 0.001) and nPCR (p = 0.031) may prevent the PD patients from suffering the CAC (Tables 1 and 2).

Multivariate analysis using logistic regression forward conditional method revealed that serum phosphate level and hs-CRP were independent risk factors for CAC initiation after adjusting for gender, BMI, SBP, fibrinogen, serum creatinine, UA, nPCR, triglycerides, HDL, Homa-IR, and adjusted residual CCR (Table 3).

4. Discussion

Previous study revealed that the baseline CAC was associated with the progression of CAC in general and dialysis patients [14, 16]. We sought to identify risk factors that determined incident CAC > 0 in PD patients with the hope of potential treatment preventing the vascular calcification from the very early status. In our study, we found serum phosphate level and hs-CRP were independent risk factors for CAC initiation.
Table 2: Laboratory characteristics of the peritoneal dialysis patients.

<table>
<thead>
<tr>
<th></th>
<th>Noninitiation (n = 37)</th>
<th>Initiation (n = 33)</th>
<th>Total (n = 70)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemoglobin (g/L)</td>
<td>106.0 ± 10.1</td>
<td>103.6 ± 13.4</td>
<td>104.9 ± 11.8</td>
<td>0.398</td>
</tr>
<tr>
<td>iPTH (ng/dL)</td>
<td>347 ± 153</td>
<td>441 ± 324</td>
<td>391 ± 251</td>
<td>0.121</td>
</tr>
<tr>
<td>Adjusted calcium (mg/dL)</td>
<td>10.3 ± 1.2</td>
<td>10.2 ± 0.9</td>
<td>10.2 ± 1.0</td>
<td>0.633</td>
</tr>
<tr>
<td>Phosphorus (mg/dL)</td>
<td>4.62 ± 0.79</td>
<td>5.90 ± 1.33</td>
<td>5.22 ± 1.25</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Transferrin (g/L)</td>
<td>1.84 ± 0.29</td>
<td>1.81 ± 0.24</td>
<td>1.83 ± 0.27</td>
<td>0.615</td>
</tr>
<tr>
<td>Ferritin (ug/L)</td>
<td>259 ± 88.8</td>
<td>278 ± 143</td>
<td>268 ± 117</td>
<td>0.311</td>
</tr>
<tr>
<td>Fibrinogen (g/L)</td>
<td>3.93 ± 0.85</td>
<td>4.43 ± 0.90</td>
<td>4.17 ± 0.90</td>
<td>0.019</td>
</tr>
<tr>
<td>Pro-BNP (pg/mL)</td>
<td>1987 (758, 5591)</td>
<td>2298 (784, 11154)</td>
<td>2191 (767, 8755)</td>
<td>0.362</td>
</tr>
<tr>
<td>Serum creatinine (umol/L)</td>
<td>851 ± 285</td>
<td>1065 ± 399</td>
<td>952 ± 357</td>
<td>0.011</td>
</tr>
<tr>
<td>BUN (mmol/L)</td>
<td>21.1 ± 5.1</td>
<td>23.1 ± 14.3</td>
<td>22.0 ± 10.5</td>
<td>0.428</td>
</tr>
<tr>
<td>UA (mmol/L)</td>
<td>0.43 ± 0.07</td>
<td>0.46 ± 0.09</td>
<td>0.45 ± 0.08</td>
<td>0.087</td>
</tr>
<tr>
<td>AKP (U/L)</td>
<td>84.8 ± 24.9</td>
<td>90.7 ± 59.9</td>
<td>87.6 ± 44.7</td>
<td>0.586</td>
</tr>
<tr>
<td>Albumin (g/L)</td>
<td>36.2 ± 4.1</td>
<td>36.2 ± 3.2</td>
<td>36.2 ± 3.7</td>
<td>0.944</td>
</tr>
<tr>
<td>nPCR (g/kg*d)</td>
<td>0.96 ± 0.19</td>
<td>0.87 ± 0.12</td>
<td>0.92 ± 0.16</td>
<td>0.031</td>
</tr>
<tr>
<td>hs-CRP (mg/dL)</td>
<td>1.75 ± 2.14</td>
<td>4.37 ± 5.0</td>
<td>2.98 ± 3.96</td>
<td>0.005</td>
</tr>
<tr>
<td>HbAIC</td>
<td>5.52 ± 0.65</td>
<td>5.83 ± 0.75</td>
<td>5.67 ± 0.71</td>
<td>0.070</td>
</tr>
<tr>
<td>Homa-IR</td>
<td>2.85 ± 1.96</td>
<td>4.12 ± 4.22</td>
<td>3.45 ± 3.26</td>
<td>0.105</td>
</tr>
<tr>
<td>Cholesterol (mg/dL)</td>
<td>179 ± 31</td>
<td>179 ± 40</td>
<td>179 ± 35</td>
<td>0.987</td>
</tr>
<tr>
<td>Triglycerides (mg/dL)</td>
<td>167 ± 108</td>
<td>261 ± 227</td>
<td>211 ± 180</td>
<td>0.027</td>
</tr>
<tr>
<td>LDL (mg/dL)</td>
<td>90.4 ± 22.8</td>
<td>86.7 ± 23.1</td>
<td>88.7 ± 22.8</td>
<td>0.499</td>
</tr>
<tr>
<td>HDL (mg/dL)</td>
<td>44.4 ± 9.7</td>
<td>34.8 ± 9.8</td>
<td>39.9 ± 10.8</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Lipidprotein A (mg/dL)</td>
<td>250 ± 204</td>
<td>243 ± 173</td>
<td>247 ± 189</td>
<td>0.879</td>
</tr>
<tr>
<td>25-OH-VitD (nmol/L)</td>
<td>36.8 ± 9.6</td>
<td>38.4 ± 10.1</td>
<td>37.7 ± 9.8</td>
<td>0.59</td>
</tr>
<tr>
<td>Glucose reabsorption (g/W/m²)</td>
<td>363 ± 116</td>
<td>404 ± 113</td>
<td>382 ± 116</td>
<td>0.144</td>
</tr>
<tr>
<td>Adjusted total ccr (L/W)</td>
<td>71.7 ± 20.0</td>
<td>63.1 ± 16.6</td>
<td>67.7 ± 18.8</td>
<td>0.057</td>
</tr>
<tr>
<td>Adjusted PD ccr (L/W)</td>
<td>37.9 ± 8.0</td>
<td>39.8 ± 6.9</td>
<td>38.8 ± 7.5</td>
<td>0.313</td>
</tr>
<tr>
<td>Adjusted residual CCR (L/W)</td>
<td>33.8 ± 25.1</td>
<td>23.4 ± 18.8</td>
<td>28.9 ± 22.8</td>
<td>0.056</td>
</tr>
</tbody>
</table>

iPTH: intact parathyroid hormone; pro-BNP: pro-B-type natriuretic peptide; BUN: blood urea nitrogen; UA: uric acid; AKP: alkaline phosphatase; nPCR: normalized protein catabolic rate; hs-CRP: high-sensitivity C-reactive protein; Ccr: creatinine clearance rate; LDL: low-density lipoprotein; HDL: high-density lipoprotein; Lp(a): lipoprotein (a); glucose reabsorption: the glucose reabsorption per week adjusted by body surface area.

Table 3: Multivariate analyses of the selected possible risk factors for CaCS initiation in PD patients.

<table>
<thead>
<tr>
<th>Variables analyzed by logistic regression forward conditional: gender, BMI, SBP, phosphorus, fibrinogen, serum creatinine, UA, nPCR, hsCRP, triglycerides, HDL, Homa-IR, and adjusted residual CCR.</th>
</tr>
</thead>
<tbody>
<tr>
<td>OR</td>
</tr>
<tr>
<td>p value</td>
</tr>
<tr>
<td>Step 1 Phosphorus (mg/dL)</td>
</tr>
<tr>
<td>Step 2 hs-CRP (mg/dL)</td>
</tr>
<tr>
<td>Step 3 Phosphorus (mg/dL)</td>
</tr>
</tbody>
</table>

in PD patients. These results suggested potential strategies to prevent the initiation of CAC in PD patients.

In 1261 general patients with zero CAC at baseline, age, LDL, SBP, and current smoking were independent predictors of CAC onset after 5.1 years of follow-up [17]. Results from the Multi-Ethnic Study of Atherosclerosis (MESA) also showed the age, SBP, smoking, LDL, HDL, and creatinine were related to the incident CAC risk [18,19]. In our study, the SBP in noninitiation group were controlled better than that in initiation group, which reveal the role of blood pressure in initiating the CAC in PD patients. Calcified arteries had upregulation of angiotensin 1 receptor and treatment with an angiotensin receptor blocker prevented the calcification [20]. However, in a CKD model, treatment with enalapril had no effect on vascular calcification in a CKD model [21]. The hypertension was not a common risk factor for CAC, mainly because patients with CAC have hypertension as a manifestation in vascular calcification patients, which may not be so to patients without CAC.

As we know, HDL reverse the atherosclerosis and reduce the risk of CVD due to reversing the cholesterol transport to liver, inhibition of oxidation, and inflammation [22]. Recently, Farhad Parhami demonstrates the ability of HDL to inhibit the calcification of vascular cells [7], and HDL benefits the arterial wall by affecting the endothelia BMP-signaling, essential for endothelial cell survival and prevention of vascular calcification, respectively [23]. However, we failed the correlation in multivariate analysis may be due to the impaired HDL antioxidant and anti-inflammatory properties in ESRD patients to some degree [24].
In general population, compared with no detectable CAC, people with CAC had significantly higher BMI, SBP history of smoking, and fibrinogen [25]; data from MESA also showed fibrinogen was associated with CAC presence and burden [26]. As we know, fibrinogen is not only associated with the enhanced blood viscosity as a main coagulation protein, but also involved in the atherosclerosis [27]. Thus, the control of the fibrinogen may be a potential therapy in preventing from CAC in PD patients.

Vascular calcification is associated with additional non-traditional factors that may be unique to CKD, such as disordered mineral metabolism. In previous studies, the phosphate was proved to be associated with CAC progression and the mortality in both general and CKD patients [14, 28–31]. In our study, the hyperphosphatemia was related to the CAC initiation in PD patients. The mechanisms by which phosphate induces the CAC in CKD may include promoting the osteochondrogenic phenotype change of vascular smooth muscle cells (VSMC) [32, 33], phosphate-induced apoptosis of VSMC [34–36], the inhibition of osteoclast differentiation [37], and phosphorus-mediated elevation of FGF-23 [38, 39]. In El-Abbadi’s uremic models, the high phosphate fed mice were prior to mineralization, consistent with the role as an initiating event in SMC phenotype change and calcification [9]. Hyperphosphatemia contributes to several mechanisms that initiate or advance the progression of vascular calcification and is emerging as a key regulator of calcification in patients with kidney disease [40] and supplies a potential strategy to prevent the CAC to any measurable degree as cardiovascular risk increases continuously even at very low degrees of CAC burden.

In addition, inflammation may promote vascular calcification by releasing “tumor necrosis factor α,” which triggers the Wnt signaling pathway, resulting in osteogenic differentiation of VSMCs [41, 42]. Our study shows the inflammation factor, hs-CRP, is higher in the initiation group, as determined by the multivariate analysis. This finding may also be supported by Aikawa’s study. Osteogenesis is associated with local inflammation and macrophage infiltration in atherosclerosis and atherosclerotic mineralization is linked with inflammation at the earliest stages [43].

The limitation to our study was that it was an observational, single-center, and relatively small study. Though we concluded that hyperphosphatemia and hs-CRP initiate the CAC in PD patients, we did not examine whether the control of the related factor could prevent the CAC initiation.

5. Conclusion

In summary, these findings suggest hyperphosphatemia and hs-CRP are the independent risk factors of CAC initiation in PD patients, supplying potential strategies to prevent the CAC to any measurable degree at the very early status.

Competing Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

Authors’ Contributions

Da Shang and Qionghong Xie contributed equally to the study.

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Research Article

Clinical Epidemiology of Mineral Bone Disorder Markers in Prevalent Hemodialysis Patients in the Xinjiang Uyghur Autonomous Region in China

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We investigated the clinical epidemiology of mineral bone disorder markers in prevalent hemodialysis (HD) patients in Xinjiang, the largest province in China. Data were obtained from 59 hospitals. A total of 3725 patients tracked from January 1 to December 31, 2014, were enrolled. Serum calcium (Ca) levels, phosphorus (P) levels, and intact parathyroid hormone (iPTH) levels were analyzed. Serum Ca levels were lower compared to the International Dialysis Outcomes and Practice Patterns Study (DOPPS4) and the Chinese DOPPS. The hypercalcemia rate was similar to DOPPS4 and lower than in the Chinese DOPPS. Serum P levels were higher than in DOPPS4 and lower than those in the Chinese DOPPS. Hyperphosphatemia rates were higher than DOPPS4 and lower than Chinese DOPPS. Serum iPTH levels were higher than in DOPPS4 and the Chinese DOPPS. We demonstrated higher serum P and iPTH levels in Xinjiang HD patients than in the DOPPS4 and Chinese DOPPS. In contrast, serum Ca levels were lower than the other two studies. High hypocalcemia and hyperphosphatemia rates may suggest that HD services in Xinjiang are inadequate. A multidiscipline chronic kidney disease (CKD) care program needs to be established to improve chronic kidney disease-mineral and bone disorder (CKD-MBD) target achievement in Xinjiang.

1. Introduction

Xinjiang is the largest province (1.64 million square kilometers) in China, located in the northwest with a total population of about 23 million (2015). Xinjiang is further away from the ocean than any other place on earth and is inhabited by Uyghur and Han people, and 11 other ethnic groups. The characteristics of the geographical environment and lifestyle are significantly different to that of tropical and subtropical residents.

The prevalence of chronic kidney disease in rural areas was 5.4% (2007) and 9.6% (2010) in urban areas [1, 2], and an estimated 1.2 million patients were suffering from end-stage renal disease (ESRD). In 2010, the Chinese Society of Nephrology established the nationwide renal data registration platform. This platform, known as the Chinese National Renal Data System (CNRDS), collects demographic, clinical, and laboratory data on dialysis patients [3]. The Xinjiang Quality Control Center for dialysis patients is one of the registered centers in the CNRDS. Owing to the specific habitual styles of different ethnic groups and the geographical size of Xinjiang, developing a strategy for chronic kidney disease management is challenging for nephrologists. In the present study, we report on the status of chronic kidney disease-mineral bone disorder (CKD-MBD) in Xinjiang based on the registered data from the CNRDS.

2. Materials and Methods

2.1. Database. Registered data from the Xinjiang Quality Control of Dialysis Patients database covers nearly all inpatient and outpatient dialysis medical records in the Xinjiang area. Data for the present study were obtained from 14 tertiary-level hospitals and 45 secondary-level hospitals in Xinjiang. Patients were tracked from January 1 to December 31, 2014. Enrollment criteria included (1) use of dialysis, (2) being over the age of 18, (3) use of hemodialysis (HD) for at least three months, and (4) available baseline serum calcium (Ca), phosphorus (P), and intact parathyroid hormone
(iPTH) data obtained between January 1 and December 31, 2014. Exclusion criteria were (1) comorbidities with malignancy, active infectious diseases, severe liver diseases, or liver cirrhosis and (2) acute kidney failure or chronic kidney failure with temporary dialysis. All participants received more than one fasting blood sampling for laboratory examinations in the study period. A mean value of each laboratory parameter in individual was used in statistical analysis. We confirmed hypocalcemia as a total serum calcium level of <8.4 mg/dL, hypercalcemia as a total serum calcium level of >9.5 mg, hyperphosphatemia as a serum phosphorus level of >5.5 mg/L, and high iPTH with a serum iPTH level of >300 pg/mL. The management of CKD-MBD was based on the Kidney Disease: Improving Global Outcomes (KDIGO) guidelines implemented in 2009 [4]. The data collected for Xinjiang were compared to data from the CNRDS, DOPPS4, and the Chinese DOPPS [5, 6].

All blood samples were evaluated using commercial kits and an autoanalyzer. Albumin levels were measured using the bromocresol green (BCG) method (Beckman Coulter, Inc., USA). Serum phosphorus and serum calcium were measured by spectrophotometry assay (Beckman Coulter, Inc., USA; Roche Inc., Swiss). iPTH level was measured by electrochemiluminescence immunoassay (Roche Inc., Swiss). The urea reduction ratio (URR) was calculated using the following equation: 

$$URR = \frac{\text{predialysis blood urea nitrogen (BUN) – postdialysis BUN/predialysis BUN}}{\times 100\%}.$$ 

The urea clearance index ($Kt/V$) was calculated using the following equation: 

$$Kt/V = -\ln(R-0.008\times t)+\frac{4-(3.5\times R)}{\times UF/W},$$ 

where $R$ is the ratio of postdialysis and predialysis serum urea nitrogen content, $t$ is the duration of dialysis (h), ultrafiltration (UF) is the ultrafiltrate amount (L), and $W$ is the postdialysis body weight (kg).

This study was conducted in accordance with the Declaration of Helsinki (1964). The requirement for obtaining informed consent from study patients was waived in accordance with the retrospective data review regulations of the Committee on Human Research at Hospital.

### 2.2. Statistical Analysis

SPSS 17.0 statistical software was used for data analysis. Continuous data are presented as mean ± standard deviation (SD), and categorical variables are expressed as frequency counts and percentages. Box plot was used for visual presentation of continuous variables and the median 75th and 25th percentiles. The Mann–Whitney $U$ test was used for comparisons between groups. The chi-square test and one-way ANOVA were used to analyze the associations between categorical and continuous variables, respectively. Statistical significance was set at $P < 0.05$.

### 3. Results

A total of 59 hospital-facilitated HD units participated in this study. This covered 80% of the whole population of Xinjiang in 2014. Data on 3725 patients (Hans 2352 (63.2%) and Uyghurs 1373 (36.8%)) were collected. The prevalence of end-stage renal diseases (ESRD) was 162 people per million people (pmp) in the total population, 235 pmp for the Han group, and 137 pmp for the Uyghur group. Table 1 shows the general demographic characteristics of the study cohort. The mean patient age was 52.09 years, and 64.9% of participants were male. Considering the study cohort as a whole, the main causes of ESRD were primary glomerulonephritis (42.4%), diabetic nephropathy (23.1%), and hypertension-related kidney diseases (15.4%). In comparison, causes of ESRD in the Han and Uyghur groups were glomerulonephritis (37.0% versus 51.8%) and diabetic nephropathy (25.7% versus 18.6%). The proportion of dialysis vintages of <1 year, 1–3 years, 3–5 years, 5–10 years, and >10 years were 24.5%, 41.4%, 20.3%, 12.0%, and 1.8%, respectively.

Figure 1 illustrates the distribution of serum Ca, P, and iPTH levels. The percentages of hypocalcemia and hypercalcemia were 34.3% and 21.5%, respectively. The percentage of hyperphosphatemia was 50% and iPTH > 300 pg/mL was also 50%. Elevated iPTH levels showed a significant association with increased hyperphosphatemia (Figure 2). The prevalence of P-binder use was 12.1% and 80% of the P-binders taken were calcium acetate and carbon calcium.

Table 2 shows the general characteristics of the Han and Uyghur groups. Uyghur people were younger than Han people on average (47.02 years versus 55.05 years) and the dialysis vintage was significantly shorter in Uyghur people.
Table 2: General characteristic of Han and Uygur.

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Han (N = 2352)</th>
<th>Uygur (N = 1373)</th>
<th>χ²/t</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>1497 (63.6)</td>
<td>921 (67.1)</td>
<td>4.609</td>
<td>0.032</td>
</tr>
<tr>
<td>Age (y)</td>
<td>55.05 ± 15.33</td>
<td>47.02 ± 15.01</td>
<td>3.811</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Dialysis vintage (years)</td>
<td>3.44 ± 2.63</td>
<td>2.99 ± 2.13</td>
<td>27.662</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>HD frequency (times/week)</td>
<td>2.56</td>
<td>2.59</td>
<td>2.203</td>
<td>0.138</td>
</tr>
<tr>
<td>Predialysis SBP (mmHg)</td>
<td>146 ± 21</td>
<td>151 ± 22</td>
<td>27.231</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Predialysis DBP (mmHg)</td>
<td>85 ± 13</td>
<td>90 ± 14</td>
<td>57.208</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>21.70 ± 4.98</td>
<td>21.85 ± 5.38</td>
<td>3.65</td>
<td>0.546</td>
</tr>
<tr>
<td>Urea reduction ratio</td>
<td>61.64 ± 15.77</td>
<td>55.13 ± 19.95</td>
<td>54.817</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Kt/V-urea</td>
<td>1.18 ± 0.48</td>
<td>1.05 ± 0.56</td>
<td>23.963</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hemoglobin (g/L)</td>
<td>104.03 ± 21.55</td>
<td>98.65 ± 23.41</td>
<td>3.65</td>
<td>0.072</td>
</tr>
<tr>
<td>Creatinine (μmol/L)</td>
<td>841.42 ± 312.46</td>
<td>837.26 ± 361.93</td>
<td>0.072</td>
<td>0.789</td>
</tr>
<tr>
<td>Albumin (g/L)</td>
<td>38.44 ± 5.97</td>
<td>37.37 ± 6.02</td>
<td>12.569</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Calcium (mg/dL)</td>
<td>8.92 ± 1.20</td>
<td>8.40 ± 1.16</td>
<td>2.155</td>
<td>0.032</td>
</tr>
<tr>
<td>Phosphate (mg/dL)</td>
<td>5.80 ± 0.73</td>
<td>5.67 ± 2.08</td>
<td>0.668</td>
<td>0.504</td>
</tr>
<tr>
<td>iPTH (pg/mL)</td>
<td>457.72 ± 521.83</td>
<td>500.45 ± 512.41</td>
<td>1.560</td>
<td>0.120</td>
</tr>
</tbody>
</table>

Figure 1: Distribution of serum calcium, phosphorus, and intact parathyroid hormone (iPTH) levels.

Figure 2: Comparisons of serum phosphorus, according to the different levels of intact parathyroid hormone. Boxes are median and interquartile ranges. Vertical lines represent the 25th to 75th percentile. *P < 0.001 using the Mann–Whitney U test.

compared to Han (2.99 years versus 3.44 years). The Ca serum levels, P levels, and iPTH levels were not significantly different. The all-cause mortality rate of the study population was 7.9%, 7.1% in the Han group, and 8.3% in the Uyghur group (data not shown).

4. Discussion

This cross-sectional survey is the first report on the status of CKD-MBD markers in HD patients in Xinjiang. There were 3725 patients on HD and the prevalence rate was 162 pmp in Xinjiang, but Han group (234 pmp) was higher than Uyghur group (137 pmp). It may be related that the most of Han group live in urban areas and the most of Uyghur group live in rural areas. Whether it is related to different lifestyle and genetics between Han and Uyghur will be studied. The lower prevalence of HD indicate that the patients of HD will be rapidly increased following economic and medical care improve in Xinjiang during the next 5 years.

In this study, the serum Ca levels in our patients were 8.76 mg/dL, the hypercalcemia rate was 21.5%, and hypocalcemia rate was 34.3%. When compared to DOPPS4 [5, 6], Ca levels in our patients were lower, and hypocalcemia rates were higher. The mean serum P level of our patients was 5.77 mg/dL, higher than DOPPS4 (5.2 mg/dL) [6]. The hyperphosphatemia rate was 50.0% and that was higher than all countries in the DOPPS4 report [5]. The status of CKD-MBD markers in Xinjiang was similar to the Chinese DOPPS
the present study by single-point observational design. This period and a cause-effect relationship cannot be obtained in phosphorus intake, residual renal function, and accurate as detailed data on nutritional intake, particularly dietary characteristics and Quality of life in the China Dialysis Outcomes Practice Patterns study, "Prevalence and risk factors of chronic kidney disease in the adults receiving health examination from Xinjiang," Chinese Journal of Nephrology vol. 27, no. 6, pp. 400–405, 2011.


The Strategy to Prevent and Regress the Vascular Calcification in Dialysis Patients

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The high prevalence of arterial calcification in end-stage renal disease (ESRD) is far beyond the explanation by common cardiovascular risk factors such as aging, diabetes, hypertension, and dyslipidemia. The finding relies on the fact that vascular and valvular calcifications are predictors of cardiovascular diseases and mortality in persons with chronic renal failure. In addition to traditional cardiovascular risk factors such as diabetes mellitus and blood pressure control, other ESRD-related risks such as phosphorus retention, excess calcium, and prolong dialysis time also contribute to the development of vascular calcification. The strategies are to reverse "calcium paradox" and lower vascular calcification by decreasing procalcific factors including minimization of inflammation (through adequate dialysis and by avoiding malnutrition, intravenous labile iron, and positive calcium and phosphate balance), correction of high and low bone turnover, and restoration of anticalcification factor balance such as correction of vitamin D and K deficiency; parathyroid intervention is reserved for severe hyperparathyroidism. The role of bone antiresorption therapy such as bisphosphonates and denosumab in vascular calcification in high-bone-turnover disease remains unclear. The limited data on sodium thiosulfate are promising. However, if calcification is to be targeted, ensure that bone health is not compromised by the treatments.

1. Introduction

Cardiovascular disease (CVD) is the leading cause of mortality and morbidity among patients with end-stage renal disease (ESRD) who are on chronic dialysis [1]. According to the US Renal Data System, CVD accounts for approximately 40% of mortality among patients on dialysis and is the main cause of hospitalization [2, 3]. Both traditional and nontraditional risk factors have been implicated in the development of CVD in chronic dialysis patients. Traditional risk factors are those used to predict coronary heart disease outcomes in the general population and include hypertension, smoking, hyperlipidemia, hyperglycemia, and obesity. Nontraditional risk factors (i.e., anemia, abnormal calcium/phosphorus metabolism, hyperhomocysteinemia, and malnutrition) are uremia-related factors that increase in prevalence as kidney function declines and contribute to the excess risk of CVD observed in patients with chronic kidney disease (CKD) [4].

Coronary artery calcification is much more prevalent in ESRD patients than in those without kidney diseases and contributes to extremely high morbidity and mortality. Recent evidence suggests that the interaction of traditional (i.e., age, smoking, diabetes mellitus [DM], hypertension, and dyslipidemia) and uremia-related so-called cardiovascular risk factors (e.g., hyperphosphatemia, high calcium × phosphorus product, oxidative stress, systemic inflammation, protein energy wasting, P-cresol, fetuin A, the osteoprotegerin (OPG)/receptor activator of NF-κB (RANK)/RANK ligand system, and osteopontin) contributes to excessive and accelerated vascular calcification in CKD patients [5].
Table 1: Types of vascular calcification.

<table>
<thead>
<tr>
<th>Type</th>
<th>Characteristics/risk factors</th>
<th>Complication</th>
</tr>
</thead>
<tbody>
<tr>
<td>Atherosclerotic intimal</td>
<td>Calcification of atherosclerotic plaques; eccentric lumen deformation by patchy calcification of the intima in the vicinity of lipid or cholesterol deposits as present in plaque calcification; patch or striped calcification on X-ray examination. Risk factors include hypercholesterolemia, metabolic syndrome, diabetes, and hypertension.</td>
<td>Ischemia/infarction</td>
</tr>
<tr>
<td>Arterial medial calcification</td>
<td>Calcification of the media in the absence of such lipid or cholesterol deposits, known as Mönckeberg-type atherosclerosis; tram-like or pipe calcification by X-ray examination. Risk factors include abnormal calcium-phosphate metabolism and inflammation.</td>
<td>Systolic hypertension, left ventricular hypertrophy</td>
</tr>
<tr>
<td>Heart valve calcification</td>
<td>Calcification of aortic valve or mitral valve leaflets as a consequence of abnormal calcium-phosphate metabolism, inflammation, and traditional cardiovascular risk factors such as hypercholesterolemia, metabolic syndrome, diabetes, and hypertension.</td>
<td>Heart failure</td>
</tr>
<tr>
<td>Calcific uremic arteriolopathy</td>
<td>Dermal arteriolar medial calcification and dermal fat necrosis, usually in the abdomen, thighs, breasts, and buttocks. X-ray examination of the extremities including the hands and feet reveals calcified artery in the absence of thrombosis. Risk factors include diabetes, obesity, vitamin K antagonist, and steroid.</td>
<td>Painful nodule and subcutaneous skin/fat necrosis wound</td>
</tr>
</tbody>
</table>

Intervention for these risk factors may delay the progression of vascular calcification. Cinacalcet with low-dose active vitamin D attenuated the progression of vascular and aortic valve calcification in 360 hemodialysis patients [6]. Kidney transplantation offers a means to restore kidney function and mineral metabolism at the same time. Removal of CKD-related risk factors through kidney transplantation may attenuate the rate of progression compared to those who remained on dialysis. Observation of kidney transplant recipients for 2.5–4.0 years revealed a progression of coronary artery calcification at a rate of 11% per year [7, 8].

These data suggested that vascular calcification, once it occurs, is unlikely to be reversed. Thus, therapeutic interventions that stop and reverse calcification may be of great value to patients with ESRD with vascular disease. Currently, no definite therapy has emerged. Hence, we reviewed the current strategy and treatments for vascular calcification in CKD patients.

2. Could the Extraosseous Calcification Be Reversed?

Can it reverse calcification? The answer is yes. Uremia-related vascular calcifications contain poor crystalline insoluble whitlockite and soluble amorphous calcium phosphate [9]. Although there was a portion of soluble amorphous calcium-phosphate in uremic related vascular calcification, the majority of the calcification is extremely insoluble whitlockite under physiological conditions that is difficult to mobilize the calcium deposits. However, vascular and soft tissue calcifications induced by calcitriol administration to rats partially revert after withdrawal of calcitriol treatment [10]. An active cellular process seems to be involved in regression of vascular calcification. High intake of vitamin K1 supplementation can result in regression of vascular calcification and restore the artery elasticity by virtue of its ability to activate matrix Gla protein, a local intravascular calcification inhibitor [11]. Furthermore, in clinical practice, extraosseous calcification such as tumor calcinosis and uremia-related vascular calcification such as calciphylaxis were considered controllable, at least partially, by parathyroidectomy and multiple intervention treatments including negative calcium and phosphate balance and avoiding calcification inducers and restoration of anticalcification factor balance [12–16].

3. Clinical Presentation of Calcification in CKD Patients

There are four different types of vascular calcification (Table I) [17] and one type of soft tissue calcification: intimal artery calcification or medial artery calcification (Figure 1(a)), cardiac valve calcification (Figure 1(b)), calciphylaxis (Figures 1(c)–1(e)), and tumor calcinosis (Figure 1(f)). These four types are the consequence of distinct yet overlapping pathological mechanisms, and they are by no means mutually exclusive. The reliability of distinguishing medial from intimal calcification, in theory, is easy, based on light microscopic examination. On the radiography and ultrasonography images, intimal calcification is disclosed as irregular, discrete, plaque-like calcification and medial calcification reveals tram-tract, nonstenotic diffuse calcified wall thickness. However, distinction has proved to be more difficult in clinical practice [18].
Figure 1: (a) Radial artery of a patient with end-stage renal disease showing intimal (∗) as well as medial calcification (arrow) under hematoxylin and eosin stain. (b) Calcifications can be seen on the mitral valve (arrow) in computed tomography studies. (c) Macroscopic evidence of calciphylaxis, (d) skin biopsy showing fat necrosis, composed of necrotic adipocytes, minimal inflammatory cell infiltration, and extensive calcification under hematoxylin and eosin stain, and (e) right hand radiographic evidence of severe heavy medial calcification of the radial arteries and their branches as shown by the so-called tram track phenomenon were found in a hemodialysis patient with calciphylaxis. (f) Left hip radiographs show a large radiopaque lesion on the soft tissue around the hip joint comprising multiple round calcified masses.
The contribution of intimal atherosclerotic calcification to plaque rupture complicated with obstruction is undefined. Medial artery calcification contributes to vascular stiffness, which increases pulse-wave velocity to decrease diastolic blood pressure and increase systolic blood pressure. Patients undergoing chronic hemodialysis who had intimal calcification had a higher relative risk of mortality than patients with medial calcification based on both X-ray examinations of the abdominal aorta and thigh arteries and ultrasonography of the common carotid arteries [19]. Cardiac valve calcification is significantly associated with calcification in the vascular bed, even after adjusting for traditional cardiovascular risk factors, possibly reflecting a predilection for ectopic calcification in certain subpopulations [20, 21]. Previous studies demonstrated significant correlation between valvular calcification and the coronary artery calcification score as detected by computed tomography in both ESRD and non-ESRD patients [22, 23]. The calcified regions of the cardiac valves share common features with arterial atherosclerotic plaques, with infiltration of inflammatory cells, calcium deposits, and bone matrix proteins, suggesting that valvular and vascular calcifications are likely associated syndromes [24–26]. Medial calcification develops concurrently with valve calcification and classical atherosclerosis, which is itself accelerated in a calcification develops concurrently with valve calcification calcifications are likely associated syndromes [24–26]. Medial calcification based on both X-ray examinations of the abdominal aorta and thigh arteries and ultrasonography of the common carotid arteries [19]. Cardiac valve calcification is significantly associated with calcification in the vascular bed, even after adjusting for traditional cardiovascular risk factors, possibly reflecting a predilection for ectopic calcification in certain subpopulations [20, 21]. Previous studies demonstrated significant correlation between valvular calcification and the coronary artery calcification score as detected by computed tomography in both ESRD and non-ESRD patients [22, 23]. The calcified regions of the cardiac valves share common features with arterial atherosclerotic plaques, with infiltration of inflammatory cells, calcium deposits, and bone matrix proteins, suggesting that valvular and vascular calcifications are likely associated syndromes [24–26]. Medial calcification develops concurrently with valve calcification and classical atherosclerosis, which is itself accelerated in a CKD-related inflammation state [27]. The synergistic effect could increase cardiovascular mortality.

4. How to Measure and Monitor Vascular Calcification

The evaluation of vascular calcification therapies is problematic because of lack of good methods to quantify it. Computed tomography of the aorta or coronary arteries is commonly used and is the only modality that can yield truly quantitative results. However, the cost is high and the process is not easy because the patients need to control their heart rate in order to evaluate coronary artery calcification. Clinical practice needs a simple technique to provide useful information. Radiography of the lateral abdomen (abdomen aorta) [28] or chest (aortic arch) [29] and the hand [30] can be used to detect the presence or absence of vascular calcification, and an echocardiogram can be used as a reasonable alternative to computed tomography-based imaging to detect the presence or absence of valvular calcification.

5. Strategy and Treatments for Vascular Calcification

Therapeutic interventions that stabilize or potentially reverse calcification may be of great value to patients with ESRD. Bone tissue has been detected in areas of vascular calcification, including osteoblast- and osteoclast-like cells, and in various bone-related extracellular matrix proteins, suggesting that the mechanisms of formation of mineralized vessels and bone are similar [26, 31]. However, if calcification is to be targeted systemically, caution must be exercised to ensure that bone and teeth health are not compromised. Growing evidence linking bone with different functional and structural characteristics of the arterial tree has contributed to the development of the concept of bone-vascular axis. Chemical mediators of bone metabolism such as matrix Gla protein, osteocalcin, bone morphogenetic protein, osteopontin, osteonectin, osteoprotegerin, receptor activator of nuclear factor kappa B ligand (RANKL), fetuin A, and inflammatory cytokines are also involved [32].

Although there was disappearance of a portion of uremic vascular calcification, the majority of vascular calcification that is composed of highly insoluble apatite is difficult to mobilize the calcium deposits in a short-time period. The strategies should combine the preventive and treatment approaches. The strategy goal is to reverse or stop the “calcium paradox,” that is, the lack of mineral in the bones makes them weak and excessive amount of calcium in blood vessels makes them more rigid [33]. Specific interventions in CKD patients without or with vascular calcification are aimed at restoring a new balance between pro- and anticalcification factors, at the same time considering the bone-vascular axis. In addition to traditional cardiovascular risk factors such as diabetes mellitus and blood pressure control, the strategies are as follows.

5.1. Minimize Inflammation. Recent evidence demonstrates that chronic inflammation, a nontraditional risk factor, is also commonly observed in ESRD patients. The causes of inflammation in ESRD are multifactorial, and while it may reflect underlying CVD, an acute-phase reaction may also be a direct cause of vascular injury by several pathogenetic mechanisms [34]. Many of the inflammatory markers and mediators such as interleukin 1 (IL-1), IL-6, C-reactive protein, and tumor necrosis factor alpha (TNFα) are found to promote vascular calcification in CKD patients [32, 34, 35]. The switch from cuprophane dialyzers to more biocompatible materials and ultrapure dialysate has made tremendous contributions to the lowering of inflammation in dialysis patients [36, 37]. A new dialysis membrane called the high cut-off dialyzer allows the elimination of molecules with a size of up to 45 kDa in chronic dialysis patients, which could reduce the procalcific effects of serum on vascular smooth muscle cell in vitro [38]. It was demonstrated that dietary phosphorus increased serum TNFα and malnutrition accelerated the progression of vascular calcification in uremic rats [39]. Furthermore, the introduction of high-dose iron preparations raises the future specter of inadvertent iatrogenic labile iron to accelerate early atherogenesis by increasing superoxide production and upregulating adhesion molecules [40, 41]. Thus, it could be speculated that adequate dialysis, appropriate dialyzer, use of ultrapure dialysate, avoiding malnutrition, and avoiding labile iron could improve inflammation in dialysis patients.

5.2. Maintain Appropriate Bone Turnover: Avoid Low and High Bone Turnover. CVD association with low- and high-bone-turnover disease is a biphasic relationship [42]. Under a high-bone-turnover status, the activation of osteoclast which stimulates bone resorption exceeds the bone formation by osteoblast activity through bone remodeling to
release excessive calcium and phosphate from the bone into the extracellular fluid. Under a low-bone-turnover status, defective bone mineralization releases excessive calcium and phosphate into the extracellular fluid and causes vascular calcification [43]. Hence, how to keep appropriate bone turnover is very important. As mentioned earlier, bone biopsy is the gold standard for the diagnosis of bone turnover, but it is an invasive method and cannot be routinely performed. Radiographs and bone densitometry are not helpful for the diagnosis of adynamic bone disease. Low bone turnover may be suspected based on the results of biochemical parameters such as low parathyroid hormone (PTH) levels, for example, PTH levels less than twice the upper normal limit of a particular PTH assay or classic PTH levels of 150 pg/mL according to the previous Kidney Disease Outcomes Quality Initiative (KDOQI) American guidelines, a range that has a reasonably good predictive value [44]. The predictive value may be increased by adding low bone alkaline phosphatase (bAP) levels. The best cut-off for bAP to discriminate low from nonlow bone formation rate was 33.1 U/L and to discriminate high from nonhigh bone formation rate was 42.1 U/L [45]. In fact, many studies have found the bone formation rate to be better correlated with plasma bAP levels than with either plasma total alkaline phosphatase or PTH concentrations [46–49]. Despite its weaknesses as a biomarker, PTH represents perhaps the best current option for noninvasive assessment of bone turnover. Medical or surgical treatments to control hyperparathyroidism should be emphasized, and low-bone-turnover diseases should be avoided to maintain the intermediate PTH levels (i.e., 2–9 times the upper limit of normal for a particular PTH assay) of the patients, which is currently considered a desirable range according to the Kidney Disease Improving Global Outcomes (KDIGO) guidelines [46, 50].

5.2.1. Low-Bone-Turnover Disease Treatment. Aluminum-based phosphate binders have continued to be used not only in Australia but elsewhere in the world, albeit less commonly in Europe and very little in North America [51]. Thus, as a first step, deferoxamine is considered important in the treatment of aluminum-induced forms of low-bone-turnover disease because it is easy to reverse with deferoxamine (5 mg/kg/week), which facilitates removal by dialysis. The vascular calcification of adynamic bone disease could be attributed to the low capacity of bones to accommodate a phosphate or calcium load in low-bone-turnover disease [43]. Vitamin D receptor activators restored osteoblast activity, increased the osteoid volume, and reduced intravascular calcium accumulation in adynamic animal models [52]. In clinical practice, the use of low calcium dialysate [53] and native vitamin D supplementation to achieve calcidiol levels of 20–30 ng/mL should be considered [54, 55]. Furthermore, antiresorption therapy, large doses of active form vitamin D, and high calcium dialysate should be avoided in these patients [55]. Recombinant PTH [56] and antisclerostin monoclonal antibodies as bone-stimulating agents [57] may be beneficial in these patients in the future. However, there are shortages of well-controlled clinical trials and limit to their use in the current period.

Treatment of Low-Bone-Turnover Disease [44, 46, 55, 58]

Treatment of aluminum bone disease
Avoid antiresorptive agents such as bisphosphonates and denosumab
Avoid PTH oversuppression due to calcimimetics or excessive use of the active form of vitamin D
Avoid high calcium dialysate
Avoid excessive calcium-based phosphate binder

Consider using the following: noncalcium, non-aluminum-based phosphate binders; native vitamin D to achieve calcidiol levels of 20–30 ng/mL; low calcium dialysate (1.25 mmol/L); recombinant PTH and antisclerostin monoclonal antibodies

5.2.2. High Turnover Bone Disease Treatment. Patients with more than 9 times the upper limit of normal for a particular PTH assay are considered to have high-bone-turnover disease, according to the KDIGO guidelines [46]. Bone biopsy should always be performed before hyperparathyroidism treatment if there is a marked difference between the bone metabolism marker values and the serum P, Ca, and iPTH levels that could be estimated from it or if osteomalacia is suspected because of a history of heavy exposure to aluminum and bone pain or fracture of unknown cause [44, 46, 58]. The addition of an active vitamin D analogue is appropriate for uncontrolled PTH. The selection of the best vitamin D analogue remains the subject of controversy owing to their selective ability to result in less positive calcium and phosphate balance, which are key mediators of vascular calcification [59]. Further basic and clinical investigations are needed to determine the optimal doses and type of active D analogue or dual therapies with vitamin D therapy (nutritional vitamin D plus active form or vitamin D analogue). International guidelines, including the KDOQI American commentary and the 2009 KDIGO guideline, suggest that the primary choice of a hyperparathyroidism treatment may be based on serum calcium and phosphate. Cinacalcet could markedly improve the achievement of target levels in patients with secondary hyperparathyroidism [60]. Furthermore, the efficacy of cinacalcet in preventing the progression of CV calcifications (ADVANCE) trial suggested that cinacalcet plus low doses of vitamin D may attenuate the progression of vascular calcification [6]. Parathyroid treatment including percutaneous ethanol parathyroid injection and parathyroidectomy, which is reserved for severe secondary hyperparathyroidism, could increase serum osteoprotegerin and fetuin A levels to restore anticalcification factor balance and negative extravascular calcium and phosphate balance in order to reduce or stabilize vascular calcification [61, 62].

Treatment of High-Bone-Turnover Disease [44, 46, 58]

Always consider bone biopsy before hyperparathyroidism treatment if there is a marked difference...
between the total body content of P results from (1) dietary intake, (2) gastrointestinal absorption, and (3) kidney and stool excretion. When renal function deteriorates, positive phosphate balance is inevitable. Based on epidemiological data linking hyperphosphatemia to reduced survival, the KDOQI guidelines suggested that P levels should be maintained between 3.5 and 5.5 mg/dL in dialysis patients [44]. However, KDIGO guidelines have a new suggestion to maintain P levels toward the normal range in ESRD [46]. Patients whose plasma phosphate rapidly refills from intracellular stores as dialysis removes it may respond well to increasing dialysis time with standard thrice-weekly dialysis or changing to nocturnal dialysis, either daily or thrice weekly [69]. Altering dialysis membranes and blood and dialysate flow rates have very limited effects, although hemodiafiltration appears to have a modest benefit in removing phosphate by convection [69–72]. Intestinal phosphate absorption can also be reduced by limiting or discontinuing activated vitamin D therapy. Activated vitamin D analogues, particularly paricalcitol, appear to be less phosphatemic. Cinacalcet improves the attainment of KDOQI bone metabolism serum phosphate levels in dialysis patients with various stages of secondary hyperparathyroidism [73]. The use of certain high-protein low-phosphate foods such as whey protein and egg whites can also help control phosphate levels without the threat to nutritional status presented by restriction of the usual protein-linked sources of dietary phosphate [74, 75]. Phosphate is usually present in food such as beverages and compounds that contain inorganic phosphorus, which is frequently used in the food industry to extend shelf life, enhance flavor, and improve the color of food products [72–74]. The other positive vascular phosphate balance concerns including hyperparathyroidism leading to phosphate efflux from bone [73]. In addition to adequate dialysis clearance and phosphate diet control, pharmacological phosphate binder treatments should be considered. Calcium-free-based phosphate binder could decrease mortality. Sevelamer has pleiotropic effects on lipid profiles, fibroblast growth factor 23, inflammation, uremic toxins, oxidative stress, and fetuin A and improves endothelial dysfunction. Ferric citrate could provide another iron source and avoid intraarterous iatrogenic labile iron, which accelerates early atherogenesis by vessel inflammation. Lanthanum carbonate has the most potent phosphate-binding effects in decreasing pill loading. Choosing the appropriate phosphate binder depends on the budget, pill load, beneficial additional pleiotropic effects, and side effects (Table 2) [40, 41, 76–80]. A flexible sequential approach to treatment may result in greater success to avoid phosphate positive balance.

5.3. Avoid Calcium Positive Balance. The related concept that Ca concentrations do not reflect balance should always be kept in mind during clinical practice [42, 63, 64]. Ca balance can be negative, neutral, or positive, depending on treatment with calcium salts, vitamin D, and Ca levels in the dialysate. In dialysis patients with low bone turnover whose plasma PTH levels are <150 pg/mL (16.5 pmol/L) and with preexisting severe vascular and/or other soft tissue calcifications, negative calcium balance was considered. Non-calcium-based phosphate binders with 1.25 mmol/L calcium dialysate are suggested. In dialysis patients without cardiovascular complication, neutral calcium balance was considered, and the limit Ca-based binder is set at 1.5 g elemental Ca per day [44, 46]. In addition, dialysate calcium concentration should be viewed as part of the source of positive calcium balance, as an inlet dialysate Ca concentration of 1.75 mmol/L leads to a positive Ca positive balance and possibly a negative calcium balance when dealing with a Ca dialysate of 1.25 mmol/L [65, 66]. Mild and asymptomatic hypocalcemia (e.g., in the context of calcimimetic treatment and parathyroidectomy) can be tolerated in order to avoid inappropriate calcium loading in adults [67, 68]. The increased acceptable idea is that the vascular calcification of chronic kidney disease patients should influence the choice of the negative calcium balance by non-calcium phosphate binder and 2.5 mEq/L dialysate calcium concentration as more than biochemical serum calcium levels.

5.4. Avoid Phosphate Positive Balance. The total body content of P results from (1) dietary intake, (2) gastrointestinal absorption, and (3) kidney and stool excretion. When renal function deteriorates, positive phosphate balance is inevitable. Based on epidemiological data linking hyperphosphatemia to reduced survival, the KDOQI guidelines suggested that P levels should be maintained between 3.5 and 5.5 mg/dL in dialysis patients [44]. However, KDIGO guidelines have a new suggestion to maintain P levels toward the normal range in ESRD [46]. Patients whose plasma phosphate rapidly refills from intracellular stores as dialysis removes it may respond well to increasing dialysis time with standard thrice-weekly dialysis or changing to nocturnal dialysis, either daily or thrice weekly [69]. Altering dialysis membranes and blood and dialysate flow rates have very limited effects, although hemodiafiltration appears to have a modest benefit in removing phosphate by convection [69–72]. Intestinal phosphate absorption can also be reduced by limiting or discontinuing activated vitamin D therapy. Activated vitamin D analogues, particularly paricalcitol, appear to be less phosphatemic. Cinacalcet improves the attainment of KDOQI bone metabolism serum phosphate levels in dialysis patients with various stages of secondary hyperparathyroidism [73]. The use of certain high-protein low-phosphate foods such as whey protein and egg whites can also help control phosphate levels without the threat to nutritional status presented by restriction of the usual protein-linked sources of dietary phosphate [74, 75]. Phosphate is usually present in food such as beverages and compounds that contain inorganic phosphorus, which is frequently used in the food industry to extend shelf life, enhance flavor, and improve the color of food products [72–74]. The other positive vascular phosphate balance concerns including hyperparathyroidism leading to phosphate efflux from bone [73]. In addition to adequate dialysis clearance and phosphate diet control, pharmacological phosphate binder treatments should be considered. Calcium-free-based phosphate binder could decrease mortality. Sevelamer has pleiotropic effects on lipid profiles, fibroblast growth factor 23, inflammation, uremic toxins, oxidative stress, and fetuin A and improves endothelial dysfunction. Ferric citrate could provide another iron source and avoid intravenous iatrogenic labile iron, which accelerates early atherogenesis by vessel inflammation. Lanthanum carbonate has the most potent phosphate-binding effects in decreasing pill loading. Choosing the appropriate phosphate binder depends on the budget, pill load, beneficial additional pleiotropic effects, and side effects (Table 2) [40, 41, 76–80]. A flexible sequential approach to treatment may result in greater success to avoid phosphate positive balance.

5.5. Correction of Vitamin D and Vitamin K Deficiency. KDIGO recommends the correction of vitamin D insufficiency in CKD patients (i.e., 25(OH) D level, <20 ng/mL) [44]. There is some evidence that 25(OH) D supplementation can assist with the management of secondary hyperparathyroidism and possibly prevent vascular calcification [52, 81]. Vitamin K (K1 and K2) is involved in the production of bone and matrix amino acid γ-carboxyglutamatic acid proteins, anticalcification, and bone-forming molecule. Low vitamin K concentrations increase the risks of bone fracture and vascular calcification. Experimental data suggest that vitamin K antagonist may decrease the activity of matrix-g-carboxyglutamatic acid protein, a strong inhibitor of soft tissue calcification [82]. The use of vitamin K antagonists is suggested in patients with tissue calcification [83, 84]. A subgroup analysis of participants who were ≥85% adherent to a 500 μg daily vitamin K1 treatment showed a lower CAC progression in the phylloquinone group than in the controls [85]. Vitamin K supplementation may be a simple
Table 2: How to choose phosphate binders depends on the budgets, pill loading, beneficial effects and side effects.

<table>
<thead>
<tr>
<th>Phosphate binders</th>
<th>Relative coefficient</th>
<th>Pill burden</th>
<th>Beneficial effects (pleiotropic effects or others)</th>
<th>Side effect</th>
<th>Cost</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aluminum hydroxide</td>
<td>1.5</td>
<td>Low</td>
<td>No</td>
<td>Bone accumulation</td>
<td>Low</td>
</tr>
<tr>
<td>Calcium carbonate</td>
<td>1.0</td>
<td>High</td>
<td>No</td>
<td>Vascular and soft tissue calcification</td>
<td>Low</td>
</tr>
<tr>
<td>Calcium acetate</td>
<td>1.0</td>
<td>High</td>
<td>Pleiotropic effects (lipid profiles, fibroblast growth factor 23, inflammation, uremic toxins, oxidative stress, fetuin A, and improvement of endothelial dysfunction)</td>
<td>Vascular and soft tissue calcification</td>
<td>Low</td>
</tr>
<tr>
<td>Sevelamer carbonate/Sevelamer hydrochloride</td>
<td>0.75</td>
<td>High</td>
<td>Decrease absorption of vitamins A, D, E, and K; sevelamer hydrochloride (metabolic acidosis)</td>
<td>High</td>
<td></td>
</tr>
<tr>
<td>Lanthanum carbonate (Fosrenol® chewable tablet)</td>
<td>2.0</td>
<td>Low</td>
<td>No</td>
<td>Bone accumulation</td>
<td>High</td>
</tr>
<tr>
<td>Magnesium carbonate</td>
<td>1.7</td>
<td>High</td>
<td>No</td>
<td>Hypermagnesemia</td>
<td>Low</td>
</tr>
<tr>
<td>Fe-citrate (Nephoxil®)</td>
<td>1.14</td>
<td>High</td>
<td>Supply oral iron</td>
<td>Iron overload/diarrhea?</td>
<td>High</td>
</tr>
</tbody>
</table>

means to prevent the progression of vascular calcification in hemodialysis patients, a population characterized by severe functional vitamin K deficiency [86]. It is hoped that the existing coronary artery calcification in dialysis patients in current trials be reduced.

5.6. Other Antiresorption Therapies

5.6.1. Bisphosphonates and Denosumab. Bisphosphonates are synthetic analogues of inorganic pyrophosphate that have the ability to inhibit osteoclast-mediated resorption and inhibit calcium phosphate crystal deposition in the bone and vessels. In the search for a common mediator capable of influencing bone remodeling and vascular calcification, the OPG/RANK/RANK ligand system has received recent attention. Restoring a balanced RANKL-to-OPG by modulating OPG production may represent a therapeutic strategy to preventing bone loss and vascular calcification. Denosumab is considered as OPG mimicker. Bisphosphonates and denosumab could reduce vascular calcification in chronic kidney failure animal models and some case reports [87–90]. Antiresorption therapy could reduce bone turnover and should not be used in patients with adynamic bone diseases. Similar to bisphosphonates, denosumab should not be used in patients with adynamic bone disease [91, 92].

Bisphosphonates are cleared by the kidney and should be used with caution in patients with glomerular filtration rate (GFR) less than 30 mL/min. With lower GFR, there is increased drug accumulation in bone mineral and a theoretical risk of inducing adynamic bone disease. The decision to use a bisphosphonates in a CKD patient should be individualized per patient.

Denosumab, as bisphosphonates, inhibits osteoclast-mediated bone resorption, but because it is not cleared by the kidney, there is no concern of its accumulation in patients with CKD. However, several studies in CKD patients have shown that severe and life-threatening hypocalcemia can occur; thus, frequent monitoring of serum calcium is required [93–95]. In view of the potential role of the OPG-RANK-RANKL axis in the development of vascular calcification, whether this therapeutic compound can regress vascular calcifications is worth investigating. However, well-controlled trials are needed to confirm the benefit.

5.6.2. Sodium Thiosulfate. Calciphylaxis involving arteriolar media calcification with ischemia, necrosis, and skin ulcerations could lead to 80% mortality [96]. Evidence suggests that disorders in the imbalance of the deficiencies of other calcification inhibitors are causally implicated in pathological calcification processes in the body [97]. Sodium thiosulfate, used as an antidote for cyanide poisoning, binds with vascular calcium salts to form a highly soluble calcium thiosulfate salt. The use of sodium thiosulfate in the treatment of calciphylaxis has been described primarily in case reports with partial-to-complete resolution of skin lesions. Sodium thiosulfate is generally well tolerated. Sodium thiosulfate is cleared by dialysis and, thus, doses need to be infused after each dialysis (25–50 g intravenously over 1 h). The most common adverse effects during sodium thiosulfate treatment for calciphylaxis
were nausea, vomiting, and increased anion gap acidosis [98–101]. However, the current bicarbonate dialysate may resolve sodium thiosulfate-induced metabolic acidosis.

6. Conclusion

Although some experimental targets involved in calcium deposition are emerging, no intervention has been described to reliably reverse vascular calcification. Consistent implementation of a systematic multi-interventional treatment strategy is suggested, consisting of trigger-agent cessation (calcium-based phosphate binders, excessive activated vitamin D, and vitamin K antagonist) and supplemented by minimization of inflammation, control of bone turnover, and avoidance of positive calcium and phosphate balance. Correction of vitamin D and K deficiency or antiresorption therapy in high-bone-turnover disease and intravenous sodium thiosulfate to keep extracellular calcium and phosphate negative balance as much as possible may alter the course of vascular calcification. Further studies are needed to confirm clinical effects.

Competing Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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Review Article
Role of Vitamin D in Uremic Vascular Calcification

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The risk of cardiovascular death is 10 times higher in patients with CKD (chronic kidney disease) than in those without CKD. Vascular calcification, common in patients with CKD, is a predictor of cardiovascular mortality. Vitamin D deficiency, another complication of CKD, is associated with vascular calcification in patients with CKD. GFR decline, proteinuria, or tubular dysfunction aggravates vitamin D deficiency and reduces its pleiotropic effect on the cardiovascular system. Vitamin D supplement for CKD patients provides a protective role in vascular calcification on the endothelium by (1) renin-angiotensin-aldosterone system inactivation, (2) alleviating insulin resistance, (3) reduction of cholesterol and inhibition of foam cell and cholesterol efflux in macrophages, and (4) modulating vascular regeneration. For the arterial calcification, vitamin D supplement provides an adjunctive role in regressing proteinuria, reversing renal osteodystrophy, and restoring calcification inhibitors. Recently, adventitial progenitor cell has been linked to be involved in the vascular calcification. Vitamin D may provide a role in modulating adventitial progenitor cells. In summary, vitamin D supplement may provide an ancillary role for ameliorating uremic vascular calcification.

1. Introduction

Chronic kidney disease (CKD), a complex and common disease, has multiple complications with severe impacts. The risk of cardiovascular death is 10 times higher in patients with CKD than in those without CKD. This risk is even up to 100-fold higher in young patients with CKD than in those without CKD [1]. Progressive decline in the estimated glomerular filtration rate (eGFR) is associated with an increased risk of major cardiovascular events and all-cause mortality [2]. Moreover, vascular calcification, common in patients with CKD, is a predictor of cardiovascular mortality. Vascular calcification in CKD involves two pathologies: atherosclerosis and arteriosclerosis [3]. In patients with CKD, the dysregulation of calcium and phosphate metabolism induces vascular smooth muscle calcification, and CKD complications, such as renin-angiotensin-aldosterone system (RAAS) activation or insulin resistance, induce endothelial dysfunction and atherosclerosis. These pathologies coexist during CKD progression and exacerbate vascular calcification.

Vitamin D deficiency, another complication of CKD, is associated with vascular calcification in patients with CKD [4]. GFR decline, proteinuria, or tubular dysfunction aggravates vitamin D deficiency and reduces its pleiotropic effect.
on the cardiovascular system. This review assessed the role of vitamin D in uremic vascular calcification.

2. Vitamin D Metabolism

Vitamin D is synthesized in the human skin or obtained from the diet. 7-Dehydrocholesterol in the skin is converted to pre-vitamin D3 upon exposure to ultraviolet B radiation. Vitamin D from the diet, vitamin D2 (ergocalciferol) or animal vitamin D3 (cholecalciferol), is identical to the skin-synthesized vitamin D3. The enzyme vitamin D 25-hydroxylase metabolizes ergocalciferol and cholecalciferol in the liver and converts them to the 25(OH)D forms of 25(OH)D2 and 25(OH)D3, respectively. 25(OH)D combined with vitamin D-binding protein (DBP) is delivered to the kidneys and filtered through the glomerulus [5]. The delivery of the 25(OH)D-DBP compound to the proximal tubular cells is facilitated by megalin receptor-mediated endocytosis [6]. Furthermore, 25(OH)D is converted to its active form, calcitriol, by a-α-hydroxylase and transported by intracellular DBP3; thus, 1,25(OH)2D or 25(OH)D reenters the circulation. Vitamin D receptor analogues (VDRAs), such as calcitriol, paricalcitol, and maxacalcitriol, directly act on the VDR [7].

Vitamin D has pleiotropic effects on immunity, the cardiovascular system, bone, the pancreas, the breast, and parathyroid hormone (PTH). In patients with CKD, vitamin D deficiency is common and is associated with overall and cardiovascular mortality. Vascular calcification is a crucial contributor to mortality in CKD and end-stage renal disease (ESRD). Vitamin D supplements increase the survival of patients with CKD [8, 9]. Therefore, our review focused on the effects of vitamin D on vascular calcification in patients with CKD.

3. Vitamin D Deficiency in Patients with CKD: Mechanism (Figure 1)

Recent observations have demonstrated that kidney disease seems to be associated with a high incidence of vitamin D insufficiency or deficiency [10]. Studies by González et al. demonstrated that 25-hydroxyvitamin D values are <30 ng/mL, believed to be the lower limit of normal, in the majority of patients with CKD [11]. Patients who are severely proteinuric have the lowest values. These investigators have shown that virtually all of the secondary hyperparathyroidism that occurs in the course of CKD is associated with 25-hydroxy-vitamin D values that are <30 ng/mL. It is interesting to note that, in this patient group, there is a positive relationship between 25-hydroxyvitamin D levels and 1,25-dihydroxy-vitamin D levels, in contrast to what is seen in normal individuals. Thus, when 25-hydroxyvitamin D levels are increased by therapy, one would anticipate an increase in the levels in the 1,25-dihydroxyvitamin D. It is not clear whether this is a contribution of renal 1-α-hydroxylase or the 1-α-hydroxylase at extrarenal sites; however, because of the association of low levels of 25-hydroxyvitamin D with hyperparathyroidism in the course of CKD, it is recommended that, in patients with CKD, if hyperparathyroidism is detected, then 25-hydroxyvitamin D should be measured, and if found to be <30 ng/mL, then the initial step in the therapy should be to try to correct this abnormality, as the first step in the control of hyperparathyroidism. Proteinuria, tubulointerstitial injury, GFR loss, and reduction of hepatic cytochrome p450 by pharmacologic dosage of active vitamin D are the possible mechanisms inducing vitamin D deficiency [12].

3.1. Vitamin D Deficiency because of Proteinuria. Vitamin D deficiency is common in patients with proteinuria. In nephrotic syndrome, vitamin D deficiency is common, and it predicted the remission of nephrotic syndrome in a case-control study [13]. In patients with proteinuria, such as those with diabetic nephropathy, the binding of 25(OH)D to the megalin receptor is limited; thus, fewer receptors are available for 25(OH)D-DBP reabsorption. In addition, consequent to proteinuria, proximal tubular cells are damaged; hence, fewer megalin receptors are available [14].

3.2. Vitamin D Deficiency because of GFR Decline. The prevalence of vitamin D deficiency is high in patients with an impaired GFR. An impaired GFR is predictive of vitamin D deficiency [15]. GFR decline limits the delivery of 25(OH)D to the renal tubules. The decreased renal uptake of 25(OH)D limits the formation of calcitriol. Because of GFR decline, the phosphaturic hormone fibroblast growth factor (FGF) 23 is synthesized from osteocytes. FGF23 inhibits the 1-α-hydroxylase activity in the renal proximal tubule to reduce 1,25(OH)2D production and stimulates 24-hydroxylase to produce 24,25(OH)2D [16]. In contrast to FGF23, PTH, another phosphaturic hormone, increases the 1-α-hydroxylase activity. However, the function of PTH is reduced by the retention of uremic toxins and metabolic acidosis because of GFR decline and the uncoupling of the PTH receptor-protein kinase A axis [17, 18]. The decrease in klotho protein in the blood is an early event in CKD and is progressively reduced along with the decline of renal function. Low klotho partially induces FGF23 resistance, causing an initial compensatory increase in blood FGF23 to maintain P homeostasis. The increase in FGF23 decreases vitamin D levels and is followed by elevation of PTH. Hyperphosphatemia is relatively late event in advanced CKD [19]. Thus we call the FGF/PTH as the killers, but the phosphate as the chief instigator and vit-D as a victim in the development of CKD-MBD.

3.3. Vitamin D Deficiency because of Tubulointerstitial Damage. Renal tubular epithelial cells possess 1-α-hydroxylase, which converts 25(OH)D to 1,25(OH)2D, and 24-hydroxylase, which converts 25(OH)D to 24,25(OH)2D. Serum 1,25(OH)2D reduces the 1-α-hydroxylase activity in renal cells and promotes 24-hydroxylase gene activity for enhancing 1,25(OH)2D inactivation [20]. CKD progression in addition to tubulointerstitial damage reduced the activity of 1-α-hydroxylase and 25-hydroxylase. In addition, in uremic rats, indoxyl sulfate upregulated nuclear factor-κB expression in renal tubular cells and subsequently activated 24-hydroxylase [21, 22]. Increased 24-hydroxylase and decreased
1-α-hydroxylase activities cause a prominent reduction of endogenous 25(OH)D and 1,25(OH)2D products, thus increasing their decay. The severity of vitamin D deficiency increases with the progression of tubulointerstitial damage.

3.4. Therapeutic 1,25(OH)2D Usage. In CKD patients with secondary hyperparathyroidism (SHPT), vitamin D, particularly the active form of vitamin D, inhibits the parathyroid gland. Nigwekar et al. addressed the question of calcidiol deficiency, one potential factor that was not mentioned is a reduction in hepatic conversion of calciferol into calcidiol, as shown in a chronic kidney failure rat model [23]. This reduction is secondary to downregulation of the major cytochrome P450 isoforms involved in 25-hydroxylation of vitamin D in rats. Furthermore, the mechanism underlying decreased cytochrome P450 activity seems to be related to secondary hyperparathyroidism. This mechanism also could explain the poor response obtained when treating some patients with CKD with 1-α-hydroxyvitamin D [24]. The normal serum physiological 1,25(OH)2D concentration is approximately 20–30 pg/mL (50–75 pmol/L) [25]. The therapeutic dose of 1,25(OH)2D (usual dose in micrograms) is crucial for treating SHPT in patients with CKD. However, 1,25(OH)2D is the end product of the vitamin D pathway and inhibits 1-α-hydroxylase and 25-hydroxylase through feedback inhibition. A therapeutic dose of 1,25(OH)2D may downregulate 25(OH)D levels, thus reducing 25(OH)D availability in extra-renal tissues and organs and increasing 25(OH)D deficiency [26].

4. The Mechanisms of Cardiovascular Calcification in CKD

Vascular calcification is a prominent feature of arterial disease in CKD and may have an impact on cardiovascular mortality through modulating both arteriosclerosis (arterial stiffening) and atherosclerosis. According to the anatomical site, vascular calcification can be divided into three categories: arteriosclerosis, arteriosclerosis, and cardiac valve calcification.

During the progression of CKD, atherosclerosis and arteriosclerosis occur simultaneously because of mixing effects by hyperparathyroidism, renal bone dystrophy, metabolic syndrome, hypertension, retention of uremic toxin, and transformation of adventitial progenitor cells. The following are the mechanisms.

4.1. Traditional Concept of Mechanisms of Vascular Calcification in CKD: On Endothelium [27]. Atherosclerosis involves the intima layer of arterial vessels. Lipid-laden plaque within the tunica intima is a hallmark of atherosclerosis, and atherosclerosis are mainly composed of macrophages with high low-density lipoprotein (LDL) and triglyceride levels. In addition to dyslipidemia, oxidative stress and chronic inflammation contribute to endothelial dysfunction and subsequent atherosclerosis. In patients with diabetes mellitus with preserved renal function, microalbuminuria, an indicator of endothelial dysfunction, predicts the presence and progression of coronary arterial calcification [28, 29]. Oxidative stress induces endothelial dysfunction. Oxidative stress is
triggered by risk factors contributing to endothelial shearing stress change, such as RAAS aldosterone activation and hyperfiltration in diabetes mellitus, and chronic inflammation exacerbates insulin resistance or metabolic syndrome [30, 31]. With deteriorating renal function, the accumulation of indoxyl sulfate damages the endothelial cells by enhancing monocyte adhesion, thus increasing endothelial oxidative stress induced stimulated by inflammatory cytokines, and inhibits endothelial progenitor cell-associated neoangiogenesis [32]. During early-stage CKD, the dysregulation of the calcium-phosphate balance influences endothelial injury and subsequent endothelial dysfunction. However, the disruption of endothelial-derived relaxing factors may signal an early stage in atherosclerosis. Hyperlipidemia, hypertension, metabolic syndrome, protein bound uremic toxins (indoxyl sulfate/p-cresol sulfate), and CKD are the major causes of endothelial injury, partly through increase of inflammation or oxidative stress. Major cell players are endothelial cells (or valve interstitial cells; VICs), leukocytes, and intimal smooth muscle cells (SMC). Focal calcification within atherosclerotic plaques is due to both active (osteogenic) and passive (cellular necrosis) processes. The phenotypic osteocyte in calcified vessels/valves may secrete Wnt inhibitors, which may fight back inhibition of bone formation.

### 4.2. Bone Turnover and Vascular Calcification [33]

CKD progression results in less vitality of bones in patients with CKD than in normal people. Thus, low bone turnover is an innate characteristic of CKD. High PTH serum levels stimulate indolent bone cells and lead to high-turnover bone disease, with the characteristics of relatively higher bone resorption than bone formation [34]. The high bone turnover status in SHPT can induce an increase in bone demineralization, which increases calcium and inorganic phosphate release from the bones into circulation. Most CKD patients developed high PTH levels after stage 3 of CKD. Patients may present prominent soft tissue calcification and/or vascular clarification [35]. In low or high bone turnover, serum calcium and phosphate concentrations increase and excessive calcium and phosphate precipitate in the vessels [36–38]. In overtreatment of CKD patients with Ca-salts, VDRA, aluminum, or parathyroidectomy may cause them to develop low turnover bone disorders and low serum PTH levels. In patients with low bone turnover status, the decreased bone mineralization makes it difficult for calcium and inorganic phosphate to enter into bone, resulting in increased serum calcium and inorganic phosphate. Thus, patients may present with prominent vascular calcification. Both high and low bone turnover disorders are characterized by a relatively higher degree of bone resorption than bone formation, which may contribute to the elevated serum calcium and inorganic phosphate levels, and aggravate vascular calcification/ossification. Therefore, correcting the high or low turnover status of bones is crucial for alleviating vascular calcification.

### 4.3. PTH and Vascular Calcification

The elevation of FGF23 increases the degradation of 25(OH)D by enhancing the activity of 24-hydroxylase and inhibition of 1-α-hydroxylase. The expression of klotho from distal renal tubules decreases with decreasing 25(OH)D. In addition, indoxyl sulfate prevents the calcitriol-induced inhibition of parathyroid cell proliferation. PTH acts on PTH receptors on osteoblasts and drives the proliferation of hematopoietic stem-progenitor cells (HSPCs) in the bone marrow either directly [39] or through the stimulation of granulocyte-colony stimulating factor, which consequently, through osteoblast loss and reduced CXCL12 expression by the cells inside the niche, fosters HSPC transmigration into the vascular sinuses [40, 41]. The circulating CD34 progenitor cells and CD34-positive vascular endothelial growth factor receptor-2-positive endothelial progenitor cells, which are correlated with vascular calcification, worsen the endothelial dysfunction [42]. Previously, smooth muscle cells, mesenchymal stem cells, and pericytes were considered to be the major precursors of ectopic chondrogenic cells in calcified vessels. In recent studies, endothelial cells have been demonstrated to participate in tissue calcification by providing osteochondrogenic cells via the endothelial-to-mesenchymal transition. Recent animal study showed elevated PTH induces endothelial to chondrogenic transition in aortic endothelial cells [43]. It also showed that cinacalcet ameliorates aortic calcification in uremic rats via suppression of endothelial-to-mesenchymal transition [44]. These data showed high PTH levels may contribute to the development of the intima (endothelial) calcification (atherosclerosis) other than the phosphate induced medial layer calcification.

### 4.4. Hyperphosphatemia and Vascular Calcification

In early CKD, compensatory mechanisms mediated by FGF23 maintain phosphaturia at a sufficient level. With increasing renal function impairment, phosphate retention induces compensating elevation of the phosphaturic hormones FGF23 and PTH. Phosphate retention occurs very early in the course of CKD, and it contributes to the genesis of SHPT [45]. In dialysis patients with calciphylaxis, hyperphosphatemia and calcium × phosphate product, but not PTH, were found to be risk factors in case-control studies [46, 47]. Hyperphosphatemia stimulates endothelial cells to release microparticles, which reduce the secretion of annexin II, reduce angiogenesis, increase the production of reactive oxygen species, and enhance inflammation, resulting in apoptosis of the endothelial cells [48]. Serum phosphate influences the endothelial response; inorganic phosphate induces endothelial dysfunction by producing oxidative stress and reducing nitric oxide production, by a higher magnitude than does indoxyl sulfate [49]. In CKD, abnormal mineral metabolism, predominantly hyperphosphatemia and hypercalcemia, facilitates the progression of the active process of osteogenesis in vascular smooth muscle cells (VSMC) resulting in arteriosclerosis calcification. VSMCs cultured in higher phosphorus concentrations express genes as markers of osteoblasts and induce both calcification in extracellular tissues and osteochondrogenesis [50]. Hyperphosphatemia plays an important role in the development of vascular calcification.
4.5. FGF23/Klotho and Vascular Calcification. FGF23, secreted by osteocytes, induces left ventricle hypertrophy. However, studies have reported direct effects of FGF-23 and klotho on vascular calcification. Recent clinical and observational data suggest that FGF23 is linked to cardiovascular mortality as well as subclinical indices of cardiovascular pathology such as left ventricular hypertrophy, vascular calcification, and endothelial dysfunction [51]. In patients at various CKD stages, plasma FGF23 is an independent biomarker of vascular calcification [52]. In observation studies, FGF23 seemingly exerted an anticalcification effect. FGF23-knockout mice show severe vascular calcification in addition to hyperphosphatemia. FGF23 mutation is associated with ectopic calcification [53]. However, in animal study, these results demonstrate that FGF23-klotho signaling is absent in mouse arteries and that the vascular response was unaffected by FGF23 treatment [54]. However, when the effects of FGF23 have been blocked with monoclonal anti-FGF23 antibodies in an experimental animal model of CKD, even if hyperparathyroidism was better controlled, the net result was a net increase in animal mortality [55], and these data cast some doubt on the putative direct pathogenic effect of FGF23. In end-staging kidney disease, impaired compensatory mechanisms by further downregulation of klotho promote osteochondrocytic differentiation of VSMCs through phosphate hoarding and increased transcription factors. On the other hand, klotho protein expressed in VSMCs suppresses osteochondrocytic differentiation with inhibition of phosphate uptake. Both renal and vascular klotho protect VSMCs against vascular calcification. In klotho-deficient vessels, the deficiency is associated with vascular calcification, and it mediates the resistance of FGF23 [56]. From the evidence above, the interaction between FGF23 and klotho on vascular calcification needs further investigation.

4.6. Decrease of Calcification Inhibitors and Vascular Calcification. Renal function impairment reduces endogenous calcification inhibitors, such as fetuin-A, matrix γ-carboxyglutamic acid protein (MGP), pyrophosphate, osteoprotegerin, and bone morphogenetic protein. Fetuin-A, a hepatic secreting protein, is a crucial inhibitor of extraskeletal calcification that exerts its action by inhibiting the de novo formation and precipitation of calcium phosphate [57]. Low fetuin-A concentrations are associated with high coronary arterial calcification scores in patients undergoing HD [58]. Fetuin-A is synthesized in the liver as a negative acute-phase protein. Low fetuin-A reflects malnutrition in patients undergoing HD, and low serum fetuin-A is associated with more severe vascular calcification and subsequent cardiovascular mortality [22]. MGP, synthesized by VSMCs [59], is observed at the interface between normal tissues and the mineralized lesions of calcified arteries in patients with CKD or diabetes mellitus. Pyrophosphate, degraded by alkaline phosphatase on the bone lining cells, serves as inhibitors of vascular calcification. Bisphosphate is resistant to alkaline phosphatase degradation, and it helps to ameliorate calcification. However, it increase serum iPTH after lowering serum calcium concentration [27, 60, 61]. Studies have reported that the vitamin K-dependent γ-carboxylation of glutamate residues is mandatory for MGP’s ability to chelate minerals and inhibit calcification [62]. In rats, uremia impairs vitamin K recycling by reducing the γ-carboxylase activity [63]. In patients undergoing maintenance HD, low vitamin K is common because of low protein intake and high energy wasting; vitamin K antagonists, such as warfarin, are strongly associated with arterial calcification [64–66]. Exposure to vitamin K antagonists has been recognized as a predictor of vascular calcification in patients undergoing maintenance HD [23, 67].

4.7. Adventitial Cell and Vascular Calcification. Mesenchymal stem cell-like cells are present in the vascular wall, particularly in the inner layer of the tunica adventitia. After intimal injury, intimal endothelial progenitor cells are present in the reendothelialization area. Submural muscular progenitor cells provide rapid replacement after insult to the tunica media according to the outside-in paradigm [68]. Soluble mediators released from adventitial progenitor macrophages, such as TGF-β and platelet-derived growth factor, activate the sonic hedgehog (Hh) signal and subsequently smooth muscle transformation from SMCs through Gli1. Perivascular Gli1 progenitors are key contributors to injury-induced organ fibrosis [69]. Gli1+ cells located in the arterial adventitia are progenitors of VSMCs and contribute to neointima formation and repair after acute injury to the femoral artery. Gli1+ cells are critical adventitial progenitors in vascular remodeling after acute and during chronic injury [70]. Thus, Gli1+ adventitial cells play a critical role in vascular calcification in CKD.

5. Vitamin D Supplements Have Therapeutic Effects on Vascular Calcification in CKD (Table 1)

In addition to vitamin D-dependent vascular calcification because of excessive use of VDRAs, vitamin D deficiency is related to vascular calcification in CKD. The normal 25(OH)D level in the blood is 30–80 ng/mL (75–200 nmol/L). Although a consistent conclusion has yet to be reached, most professionals have reported levels of 20–30 ng/mL (50–75 nmol/L) as vitamin D deficiency [4]. Vitamin D supplements are typically prescribed at levels lower than 30 ng/mL (75 nmol/L) in patients with CKD; vitamin D deficiency is associated with higher mortality in these patients. Although the cutoff value of serum 25(OH)D and vascular calcification remains controversial, vitamin D deficiency is associated with vascular calcification in patients with CKD. In uremic vascular calcification, a low serum 25(OH)D level is related to more severe calcification in patients with CKD [71]. Luo et al. reported that, in patients with CKD not undergoing HD, a serum 25(OH)D level lower than 20 ng/mL was associated with increased arterial stiffness [72]. In patients with ESRD, 25(OH)D negatively correlated to the severity of coronary arterial calcification, and a lower serum 25(OH)D level is associated with aortic pulse velocity [59]. In an animal study, vitamin D deficiency accelerated vascular calcification and atherosclerosis, independent of the expression of LDL receptors on vessels [73]. In LDL-knockout mice, a low vitamin
D diet stimulated the osteogenic expression of SMCs [74]. In uremic vascular calcification with vitamin D deficiency, vitamin D supplements exert a protective role.

5.1. Vitamin D Supplements for Endothelial Dysfunction. As stated in the previous section, multiple factors affect endothelial function during early-stage CKD. The expression of VDRs on endothelial cells remains controversial; therefore, we discuss the effects of vitamin D on hormone release.

5.1.1. RAAS Inactivation. Vitamin D deficiency has been associated with systemic and intrarenal RAAS activation in humans after angiotensin II infusion [16]. Vitamin D analogues or endogenous vitamin D supplements in addition to RAAS blockade agents have been reported to exert additive effects for reducing proteinuria [75]. Active vitamin D downregulates renin expression by suppressing renin gene transcription [76]; it reduces urinary angiotensinogen and intrarenal RAAS blockade [77]. These observations reveal that vitamin D supplements reduce the activation of the RAAS system in patients with CKD, with or without angiotensin converting enzyme inhibitors or angiotensin-receptor blockers.

5.1.2. Reduction of Cholesterol and Inhibition of Foam Cell and Cholesterol Efflux in Macrophages. The activation of VDRs by 1,25(OH)2D reduces liver and serum cholesterol levels because VDRs suppress the expression of the small heterodimer partner and activation of cholesterol 7-α-hydroxylase (CYP7A1). CYP7A1 is the rate-limiting enzyme in bile and reduces the serum cholesterol concentration [78, 79]. Oxidized LDL cholesterol retention in the vascular wall is harmful for the activation of immune cells, thus decreasing cholesterol efflux and releasing proinflammatory cytokines [80]. The differentiation of monocytes into M1 macrophages by interferon-γ is associated with higher endothelial stress and atherosclerotic plaque formation. Oh et al. reported that, in patients with diabetes mellitus, macrophages incubated with 1,25(OH)2D suppressed the formation of foamy cells by reducing acetylated or oxidized LDL cholesterol uptake [81]. Riek et al. reported that monocytes in patients with diabetes mellitus tend to differentiate to M2 macrophages on incubation with 1,25(OH)2D, vitamin D3, and endoplasmic reticulum stress is alleviated [82]. Vitamin D supplements reduce hypertension and atherosclerotic changes in mice [83]. Thus, vitamin D plays a role in reducing the formation of atheromas or atherosclerotic changes.

5.1.3. Vascular Regeneration. 1,25(OH)2D directly influences VSMC regeneration through VDRs. Wu-Wong et al. reported that vitamin D downregulated thrombotic molecules from VSMCs from a human aortic cell culture. 1,25(OH)2D modified the vascular tone by regulating nitric oxide release from VSMCs [84]. Nutritional vitamin D supplements provide circulating CD45-negative and CD117-, stem cell antigen-1-, and fetal liver kinase 1-positive angiogenic myeloid cells, which are considered to promote vascular regeneration. 1,25(OH)2D promotes reendothelialization in injured endothelial cells by increasing stromal cell-derived factor, which is associated with the homing of angiogenic myeloid cells [85]. However, the interaction between progenitor cells of angiogenesis and supplements of vitamin D should modulate the vascular regeneration by affecting the interaction between endothelial cells and VSMCs.

5.2. Vitamin D Supplements for Arterial Calcification: Mechanisms

5.2.1. Application in Renal Osteodystrophy. In high-turnover bone disease, PTH suppression depends on active or nutritional vitamin D supplements. Active vitamin D suppresses

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the chief cells of PTH, and several derivatives reduce vascular calcification through anti-inflammatory effects [86]. However, hypercalcemia and hyperphosphatemia are common, and they are associated with further vascular calcification. Vitamin D derivatives, such as paricalcitol, are associated with a reduced incidence of hypercalcemia and hyperphosphatemia and lower severity of vascular calcification [86, 87]. Nutritional vitamin D inhibits PTH in patients with CKD [88, 89] and is associated with a lower incidence of hypercalcemia than active vitamin D. Nutritional vitamin D is converted to 1,25(OH)2D in the parathyroid gland through autocrine or paracrine mechanisms, and it binds to 24-hydroxylase to avoid 1,25(OH)2D degradation [90]. In low turnover disease, both vitamin D analogues and nutritional vitamin D alleviate vascular calcification. Mathew et al. reported that, in mice with adhesive bone disease, vitamin D analogues restored osteoblast activity, increased the osteoid volume, and reduced intravascular calcium accumulation [91]. At physiological concentrations, osteoblasts are activated and bone formation is accelerated. Eldecalcitol [1α,25-dihydroxy-2β-(3-hydroxypropyloxy) vitamin D3] is an analogue of 1α,25-dihydroxyvitamin D3 [1,25(OH)2D3], bearing a hydroxypropyloxy residue at the 2β position. In preclinical studies, eldecalcitol suppressed bone resorption to a greater extent than alfalcacical but had a similar effect on bone formation and calcium metabolism, resulting in a greater increase in bone mineral density in ovariectomized rats [92]. Because eldecalcitol stimulates intestinal calcium absorption and improves calcium balance in addition to its skeletal effects, combination treatment with antiresorptive agents may be able to show better effects than native vitamin D and calcium supplementation in preventing fractures in patients with high bone turnover bone disease. Furthermore, during the treatments, the patients should keep negative extrasosseous calcium balance and minimize total positive calcium balance as possible.

Further studies are warranted to clarify these issues. Under controlled concentrations of calcium and phosphate, vascular calcification can be controlled by correcting renal osteodystrophy.

5.2.2. Restoring Calcification Inhibitors. Vitamin D analogue supplements help to restore vascular calcification inhibitors. Hansen et al. reported that fetuin-A significantly increased in patients undergoing HD who were receiving alfalcacidol rather than paricalcitol [93]. In addition, in arteries of patients with CKD, VDRAs restored the mRNA expression of klotho. The local restoration of klotho reversed the anticalcifying effect of FGF23 [56]. Moreover, VDRAs increased the expression of osteopontin in aortic cells from uremic mice [94]. Cianciolo et al. reported that VDRAs reduced endothelial progenitor cells’ expression of osteocalcin, which is a calcification promoter [95]. Therefore, vitamin D might restore calcification inhibitors in patients with uremia and alleviate vascular calcification.

From the perspective of endothelial dysfunction and arterial calcification during GFR decline, vitamin D supplements should provide protection against vascular calcification.

5.3. Effects of Vitamin D Supplements on Vascular Progenitor Cells: Vitamin D/Sonic Hedgehog Signaling and Gli1+ Cells in Vascular Calcification of CKD (Figure 2). Studies using primary cultured human keratinocytes have reported that 1,25(OH)2-dihydroxyvitamin D3 suppresses cyclin D1 and Gli1. The blockade of VDR by siRNA resulted in increased expression of cyclin D1 and Gli1 during embryogenesis and maintains stem cell populations in certain adult tissues. The potential mechanisms of cross-talk between Hh signaling and calcitriol-VDR signaling suggest a cooperative role during multiple stages of human development and diseases [98]. The most common target associated with the Hh pathway is Gli1, which controls the expression levels of multiple genes related to Hh signaling, cell cycle progression, cell adhesion, and apoptosis [99]. These findings suggest that vitamin D deficiency contributes to vascular calcification through increased Gli1 expression in CKD.

6. Vitamin D Supplements for Vascular Calcification: Clinical Evidence

Vitamin D deficiency is common in CKD and is associated with vascular calcification; therefore, vitamin D supplements...
are considered. In early CKD with a preserved GFR, vitamin D supplements have additive effects in reducing blood pressure, without affecting calcium or phosphate concentrations. Clinical studies have reported an antiproteinuric effect of vitamin D supplements. The supplements result in efficient sugar control by reversing insulin resistance. In advanced CKD, active vitamin D provides control over PTH. In addition, retrospective studies have reported reduced cardiovascular mortality in patients undergoing maintenance HD who were receiving active vitamin D supplements [87, 100, 101]. Therefore, adequate vitamin D supplements have been used as a cardioprotective agent.

Because of its potential role in cardiovascular protection, vitamin D has been used in several clinical trials. The VITAL study initiated in 2008 reported the effects of VDRAs in patients with CKD. Furthermore, 24 weeks of paricalcitol at a dosage of 1-2 μg/day alleviated proteinuria in patients with diabetic nephropathy. Serum alkaline phosphatase and intact PTH were reduced in patients receiving paricalcitol [102]. Thethi et al. reported that, in patients with diabetes mellitus having an eGFR between 15 and 59 mL/min/1.73 m², a daily supplement of paricalcitol did not improve brachial artery flow-mediated dilatation or nitroglycerine-mediated dilation [103]. In two recent double-blind RCTs (PRIMO and OPERA studies) in nondialysis CKD stages 3–5 patients active vitamin D (paricalcitol) failed to demonstrate the improvement in clinically cardiac outcome but did demonstrate an increased risk of hypercalcemia [104, 105]. Active vitamin D analogues, particularly the nonselective forms, are associated with hypercalcemia and hyperphosphatemia because of a direct effect on the intestinal absorption of calcium and phosphate.

In contrast to active vitamin D or vitamin D analogues, nutritional vitamin D is associated with a lower incidence of hypercalcemia [106, 107], and it acts as the substrate of extrarenal 1-α-hydroxylase. Cholecalciferol at a dosage of 25,000 IU every 2 weeks was effective, without inducing hypercalcemia or hyperphosphatemia in patients undergoing HD [107]. For nutritional vitamin D supplementation, a six-month course of oral cholecalciferol treatment was given to adult (age: 53.8 ± 17.3) long-term maintenance dialysis patients with vitamin D insufficiency. The cholecalciferol replacement did not demonstrate an increased risk of hypercalcemia [108]. In the study of cardiovascular disease in young adults with childhood onset of ESRD, the Berlin pediatricians Briese et al. [109] pointed out interesting differences with a quite similar study reported by Heidelberg [4]: the prevalence of coronary calcifications (10 versus 92%) and of cardiac valve calcifications (0 versus 32%) was quite lower in the Berlin study (more use of cholecalciferol) than in the Heidelberg one (less use of cholecalciferol), while the technique of evaluation was comparable. These findings suggest that nutritional vit-D supplement may prevent the development of vascular calcification [110]. Notably, both high and low vitamin D levels are associated with vascular calcification, and supplementation with excessive or insufficient exogenous vitamin D has been associated with vascular calcification in vivo and human studies [111]. Assimon et al. reported that ergocalciferol reduced the plasma concentration of adhesion molecules in patients undergoing HD [112]. Several clinical trials have analyzed the effects of nutritional vitamin D on patients with CKD and ESRD. Hewitt et al. [113] reported that cholecalciferol administered to patients with ESRD for more than 6 months reduced tartrate-resistant acid phosphatase-5b but not the pulse-wave velocity. Kidir et al. reported that cholecalciferol improved arterial diastolic function in patients undergoing dialysis [108]. Since the mechanism of vascular calcification in CKD is complex, early use of nutritional vit-D in CKD may be helpful although its therapeutic effect is still unproven.

7. Native Vitamin D Supplements for CKD Patients: The Timing for Supplement and the Dosage

7.1. Check Serum 25(OH)D Level Since CKD Stage 3. Under normal renal function, high or low serum 25(OH)D levels do not adequately affect 1,25(OH)2D levels. However, in advanced kidney diseases (glomerular filtration rate [GFR] < 25 mL/min), the serum 25(OH)D and 1,25(OH)2D levels exhibit strong relationships because diminished 25(OH)D can be converted to its bioactivated form 1,25(OH)2D by the residual renal 1-α-hydroxylase in CKD. Higher physiological 25(OH)D levels can upregulate serum 1,25(OH)2D levels in CKD patients [114]. Thus, the most appropriate time for initiation of therapy will be stages 3-4 of CKD with low serum 25(OH)D levels. Regular check serum levels of 25(OH)D will be the adequate therapeutic strategies.

7.2. The Advantage of Native Vitamin D in CKD. Compared with active vitamin D (calcitriol), 25-hydroxy vitamin D (calcifediol) has a longer plasma half-life and less potency, with fewer effects on hypercalcemia; thus, it has become a crucial agent for vitamin D replacement in CKD patients. A previous study of the pharmacokinetics of oral cholecalciferol and calcifediol revealed that calcifediol given daily, weekly, or as a single bolus is approximately 2-3 times more potent in increasing plasma 25 (OH) D3 concentrations than cholecalciferol [36]. Recently, a modified-release oral formulation of calcifediol was designed to gradually raise serum 25-hydroxyvitamin D to minimize the induction of CYP24 (the cytochrome P-450 enzyme that specifically catabolizes vitamin D and its metabolites) and was found to reduce iPTh more effectively in patients with secondary hyperparathyroidism [115].

Native vitamin D supplementation prevents secondary hyperparathyroidism in early CKD; KDOQI suggests its use is not beneficial in advanced CKD because of the lack of 1-α-hydroxylase in the kidneys [116]. However, another study indicated that 1,25(OH)2D levels were increased after supplementation with native vitamin D in hemodialysis patients and suggested that there was enough extrarenal 1-α-hydroxylase activity to produce serum levels of 1,25(OH)2D even in ESRD [117]. We also had shown that cholecalciferol, in combination with paricalcitol, additively lowers the iPTh levels in a significant number of HD patients with SHPT. A dose
of 5000 IU/week of cholecalciferol could maintain serum 25(OH)D3 levels above 30 ng/dL as early as 8 weeks after beginning supplementation [118]. The combination therapy of native vitamin D and active vitamin D supplements has fewer adverse effects of hypercalcemia and hyperphosphatemia and can improve bone quality efficiently. In patients with adynamic bone disorder (low bone turnover), the viability of osteoblasts and osteoclasts is low. Providing native vitamin D or intermittent PTH supplements may rescue the function of osteoblasts, improve bone turnover, and promote bone health.

7.3. The Dosage of Native Vitamin D in CKD Patients. Public-health authorities are responsible for ensuring the recommended daily vitamin D intake of 600–800 IU in the general population (under circumstances of limited or no sun exposure). With regard to patient care, the antifracture effects of vitamin D have been documented for daily vitamin D doses of 800–2,000 IU, whereas daily vitamin D doses of up to 4,000 IU (and probably even 10,000 IU) are considered to be safe with regard to acute vitamin D toxic effects leading to hypercalcemia [4].

Vitamin D supplementation to maintain 25-OHD concentrations at 20–30 ng/mL or higher (but <50 ng/mL) with or without VDRA therapy is inexpensive, appears safe, and may have additional health benefits in patients with > stage 3 CKD [119]. Daily vitamin D intake of 600–800 IU in CKD patients without high PTH levels is recommended. In CKD with high PTH levels, a minimum daily dose of 2,000 IU of vitamin D3 (equivalent to 14,000 IU/wk) likely is required to achieve serum 25(OH)D concentrations >30 ng/mL [120].

8. Conclusion

Vitamin D deficiency is common in patients with CKD because of GFR decline, renal tubular dysfunction, and proteinuria. With CKD progression, multiple factors exacerbate vascular calcification, including vitamin D deficiency. VDRAs or nutritional vitamin D supplements facilitate the alleviation of vitamin D-dependent or vitamin D-independent vascular calcification. Nonselective VDRAs may increase vascular calcification by inducing hyperphosphatemia and hypercalcemia. Nutritional vitamin D supplements may provide an ancillary role for ameliorating uremic vascular calcification.

Competing Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

References


Research Article

Association between the Achievement of Target Range CKD-MBD Markers and Mortality in Prevalent Hemodialysis Patients in Taiwan by Using the Kidney Disease: Improving Global Outcomes Clinical Guidelines

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Background. This study evaluated the association between achieving target chronic kidney disease-mineral and bone disorder (CKD-MBD) marker levels and mortality in Taiwanese hemodialysis (HD) patients. Target levels were based on the Kidney Disease: Improving Global Outcomes (KDIGO) guidelines.

Methods. We performed a retrospective medical record review of 1126 HD patients between 2009 and 2013. A logistic regression model was used to evaluate the relationship between achieving target marker levels and the risk for all-cause and cardiovascular (CV) mortality. Reference target ranges were $7.9 \leq \text{calcium (Ca)} \leq 9.9 \text{mg/dL}$, $2.4 \leq \text{phosphate (P)} \leq 4.7 \text{mg/dL}$, and $144 \leq \text{intact parathyroid hormone (iPTH)} \leq 648 \text{pg/mL}$.

Results. Achievement of target P levels was associated with a lower risk for all-cause mortality compared to achievement of either target Ca or iPTH levels. Achieving target P + iPTH levels (OR 1.32) was associated with a lower odds ratio for all-cause mortality compared to achieving target Ca + P (OR 1.66) and Ca + iPTH (OR 1.43) levels. Similar trends were observed for CV mortality risk.

Conclusions. The present study demonstrated that achieving serum P levels within the KDIGO target range is the most important factor for lowering mortality in HD patients.

1. Introduction

Chronic kidney disease-mineral and bone disorder (CKD-MBD) has emerged as an important factor in the care of CKD patients. The Kidney Disease: Improving Global Outcomes (KDIGO) guidelines define CKD-MBD as a systemic disorder of mineral and bone metabolism due to CKD and manifested by either one or a combination of the following: (1) abnormalities of calcium (Ca), phosphorus (P), parathyroid hormone (PTH), or vitamin D metabolism; (2) abnormalities in bone turnover, mineralization, volume, linear growth, or strength; and (3) vascular or other soft-tissue calcification [1]. In the past several decades, many epidemiologic studies have revealed that CKD-MBD markers are associated with higher rates for both all-cause and cardiovascular (CV) mortality [2–7]. Accordingly, clinical guidelines have set the target range for three CKD-MBD markers (namely, Ca, P, and PTH) in CKD-MBD management. The two clinical guidelines adopted by the majority of clinicians are the US National Kidney Foundation Kidney Disease Outcomes Quality Initiative (NKF KDOQI) guidelines implemented in 2003 [8] and the Kidney Disease: Improving Global Outcomes (KDIGO) guidelines implemented in 2009 [1]. Owing to variations in health support systems and living habits around the world, discrepancies between target ranges and achievement rates in CKD-MBD markers have been reported [9–12]. In the Dialysis Outcomes and Practice Patterns Study, only 4.6% to 5.5% of patients were within range for all four CKD-MBD
markers (serum P, albumin-corrected Ca, Ca-P product, and iPTH), and only 23% to 28% patients fell within the guidelines for at least three markers [9]. Fouque et al. [13] investigated the CKD-MBD marker targets in the KDIGO guidelines by using data from the French Phosphate and Calcium Observatory Study and found low achievement rates for Ca (59.4%), P (32.6%), and PTH (60.7%) targets and an extremely low rate of target achievement for all three markers (12.3%). Another report from Chinese dialysis patients also revealed suboptimal achievement rates based on the NFK KDOQI clinical guidelines for target Ca (38.6%), P (37.6%), and PTH (26.5%) levels [14].

The NFK KDOQI guidelines for CKD-MBD recommend that clinicians maintain serum Ca, P, and iPTH levels within a narrow range [8]. In contrast, the KDIGO CKD-MBD guidelines recommend maintaining serum Ca, P, and iPTH levels in a reference range determined by individual laboratory tests [1]. However, the recommendations of these two clinical guidelines are primarily based on retrospective observational studies. Therefore, the ability of these recommendations to predict mortality and morbidity across time and countries has subsequently been challenged. To date, there have been no randomized controlled trials which have determined the ideal ranges for CKD-MBD markers, and so we can only rely on the results from epidemiological observational studies.

As the largest hospital-facilitated HD center in Taiwan, we evaluated the relationship between achievement rates of KDIGO determined CKD-MBD marker levels and mortality between 2009 and 2013.

2. Materials and Methods

2.1. Study Participants. Patients who received regular outpatient HD three times per week at Kaohsiung Chang Gung Memorial Hospital in Taiwan were enrolled in the study. These patients were tracked from January 1, 2009, until December 31, 2013. In total, the medical records of 1126 HD patients were reviewed and the demographic data and CKD-MBD laboratory marker levels were compiled. Patients were excluded if (1) they did not receive regular HD thrice weekly and/or (2) they did not receive continuous HD for at least 3 months in the study hospital.

2.2. Retrospective Analysis of Patient Medical Records. We retrospectively analyzed serial hemogram results and biochemical data for each patient between January 2009 and December 2013. Baseline characteristics included the baseline levels of blood albumin, hemoglobin (Hb), cholesterol, triglyceride, fasting blood glucose, ferritin, intact PTH (iPTH), noncorrected Ca, P, potassium (K), uric acid, Kt/V urea score, and cardiothoracic (CT) ratio. Laboratory values for blood analysis were measured monthly with the exceptions of ferritin (measured every 3 months), iPTH (measured every 6 months), and Kt/V urea score (Daugirdas method) (measured every 6 months) [15]. Blood sampling was performed in the mid-week session before routine hemodialysis (Wednesday, Thursday). Kt/V scores were calculated using the following equation: Kt/V urea score = \[-\ln(R-0.008\times t)\times UF/W\], where R is the ratio of postdialysis BUN to predialysis BUN, t (in hours) is the duration of dialysis, UF (L) is the ultrafiltrate amount, and W (kg) is the postdialysis body weight. The reference values in our laboratory for CKD-MBD markers are as follows: serum Ca 7.9–9.7 mg/dL, serum P 2.4–4.7 mg/dL, and serum iPTH 14–72 pg/mL. All blood samples were analyzed using commercial kits and an autoanalyzer (Hitachi 7600-20i, Hitachi Ltd., Tokyo, Japan). Albumin was measured by the bromocresol green (BCG) method. iPTH was measured by chemiluminescence immunoassay (Siemens Healthcare Diagnostics Inc., USA). To determine the cardiothoracic (CT) ratio for each patient, chest radiography was performed following HD. Cardiac size was measured as the distance between parallel lines drawn down to the most lateral points on both sides of the heart. Thoracic width was measured as the distance between parallel lines drawn down the inner aspect of the widest points of the rib cage. The CT ratio was defined as cardiac size/thoracic width. All patients received HD with dialyzers that had an effective surface area > 2.0 m². Majority subjects received dialysate Ca 3.0 mEq/L. Dialysate Ca 3.5 mEq/L or 2.5 mEq/L was used when the patients presented with hypocalcemia or hypercalcemia in the routine monthly blood examinations.

2.3. Approval. The study protocol was approved by the Committee on Human Research at Kaohsiung Chang Gung Memorial Hospital (101-1595B) and was conducted in accordance with the Declaration of Helsinki (1964). The requirement for obtaining informed consent from study patients was waived in accordance with the retrospective data review regulations of the Committee on Human Research at Kaohsiung Chang Gung Memorial Hospital.

2.4. Patient Demographics and Baseline Characteristics. The demographics and baseline characteristics of the study population are represented as numbers and percentages for categorical data and as means ± standard deviation for continuous data. Demographics included age, sex, HD vintage, etiology of renal failure, erythropoietin (EPO) use, vitamin D analogs use, antihypertensive agent use, iron use, and parathyroidectomy status. The study endpoints were all-cause and cardiovascular (CV) mortality.

2.5. Mortality versus Patient Responses according to KDIGO Guidelines (Logistic Model). According to the KDIGO clinical practice guidelines, patients who achieved targeted levels of serum Ca (7.9–9.9 mg/dL) during the study constituted the target response group. For P levels, “normal values of laboratory” according to KDIGO recommended values were 2.4–4.7 mg/dL in our HD center. For iPTH levels, the target response range was 144–648 pg/mL. We calculated the mean values of CKD-MBD markers (Ca, P) in each study subject in the first three continuous months when the subjects were enrolled for observation. iPTH levels were measured every six months and the first three values were used for mean calculation. Then the mean values were used for odds ratio (OR) analysis. A logistic regression model was used to demonstrate the association between the risk
for mortality and the achievement of the target response range for corrected Ca, P, and iPTH levels in the study population. All of the patients in the target response group were defined as the reference group. Crude and adjusted OR with corresponding 95% confidence intervals (CI) were calculated using logistic regression analysis. Two categories of covariates were considered: patient demographics and baseline characteristics. A p value < 0.05 was considered statistically significant. All statistical analyses were performed using SAS 9.3.

3. Results

3.1. Patient Demographics and Baseline Characteristics. A total of 1126 HD patients were enrolled in the study. The mean duration of follow-up was 3.38 ± 1.08 years. The mean age of the study population was 60.0 ± 12.4 years old, and the mean HD vintage was 5.74 ± 5.34 years. Over the 5-year observation period, 240 patients experienced all-cause mortality, and 48 patients experienced CV mortality (Table 1). Additional patient characteristics, including laboratory parameters at baseline, HD adequacy indices, and CT ratios, are shown in Table 2.

3.2. Logistic Regression Analysis of Risk of Mortality versus Patient Responses according to KDIGO Guidelines. We examined the relationship between achieving target levels of three separate CKD-MBD markers (Ca, P, and iPTH) according to the KDIGO clinical guidelines and the risk for all-cause and CV mortality over the five-year study interval. The unadjusted logistic regression model demonstrated that hypocalcemia (<7.9 mg/dL) was significantly associated with a higher OR (OR 2.41, 95% CI 1.22–4.77, and p = 0.01) for all-cause mortality, and the trend still appeared after adjusted model analysis (models 2 and 3) (Table 3). There were also no significant differences between Ca levels and CV mortality by multivariable adjusted analysis. Hyperphosphatemia (P > 4.7 mg/dL) was significantly associated with an increased OR for all-cause and CV mortality according to multivariable adjusted analysis. Hyperphosphatemia (P > 4.7 mg/dL) was significantly associated with an increased OR for all-cause and CV mortality according to multivariable adjusted logistic regression models (OR 1.82, 95% CI 1.17–2.82, and p = 0.007; OR 2.62, 95% CI 1.19–5.76, and p = 0.01, resp.) (Table 4). Using an iPTH reference range of 144–648 pg/mL, all three models demonstrated that low (<144 pg/mL) iPTH levels were significantly associated with higher OR for all-cause mortality, but not for CV mortality. High (>648 pg/mL) iPTH levels were not significantly associated with higher OR for all-cause and CV mortalities by multivariable adjusted analysis (Table 5).

We also examined the relationship between simultaneously achieving target levels for all three CKD-MBD markers (Ca, P, and iPTH) according to the KDIGO clinical guidelines and the risk for mortality. The reference group included patients who achieved targets simultaneously for all three CKD-MBD markers (Table 6). Patients who achieved target Ca levels had a lower risk for all-cause mortality compared to those who only achieved target P or iPTH levels by multivariable adjusted models. Multivariable adjusted model analyses of the risk for mortality associated with simultaneously achieving two target CKD-MBD marker levels revealed that achieving target P and iPTH (P + iPTH) levels (OR 1.32) was associated with a lower OR for all-cause mortality compared to achieving Ca + P (OR 1.66) and Ca + iPTH (OR 1.43) levels. Achieving target Ca + P levels demonstrated the highest OR (compared to Ca + iPTH and P + iPTH) for all-cause mortality in all three logistic regression models. Analysis of the risk of CV mortality revealed similar trends.
<table>
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</table>

Table 2: Summary of laboratory parameters.

Hb, hemoglobin; iPTH, intact parathyroid hormone.

where achieving target $P + iPTH$ levels showed the lowest OR compared to achieving target levels for any other combination of CKD-MBD markers. Patients who did not achieve any of the three-target CKD-MBD marker levels demonstrated increased risk for all-cause mortality in all three logistic regression models, compared to patients who achieved any two-target CKD-MBD marker levels. In contrast, this trend was not apparent for risk of CV mortality according to case-mix adjusted and multivariable adjusted models (Table 6).

Patients number in each achievement of CKD-MBD markers was shown in supplementary Tables 1 and 2 in Supplementary Material available online at http://dx.doi.org/10.1155/2016/1523124.

4. Discussion

In the present study, we examined the association between mortality and achievement of KDIGO recommended target ranges of CKD-MBD markers (Ca, P, and iPTH) in a cohort of HD patients. Compared to reference serum Ca levels (7.9–9.9 mg/dL), serum Ca levels $< 7.9$ mg/dL and $> 9.9$ mg/dL were associated with an increased risk for all-cause mortality in all three logistic regression models (unadjusted, case-mix adjusted, and multivariable-adjusted). However, only patients with Ca $< 7.9$ mg/dL in the three models yielded a statistically significant increase in the risk for all-cause mortality. Our results are in slight disagreement with those previously reported by Fouque et al. who demonstrated a U-shape relationship between HR for mortality and total serum Ca concentrations [13]. We suppose the different patient number recruitment and study design may contribute this variable results. Of note, there have been no randomized controlled trials that have determined the ideal ranges for CKD-MBD markers. A further study in the future is needed.

When we applied KDIGO recommended serum P levels to logistical regression analyses of P level-associated mortality risk, serum P levels $< 2.4$ mg/dL and $> 4.7$ mg/dL were generally associated with an increased risk for both all-cause and CV mortality. By multivariable adjusted analysis, we found that serum P levels $> 4.7$ mg/dL had a significantly increased OR for all-cause and CV mortality. In previous reports, risk for mortality was associated with increased P levels in CKD patients [2, 3, 5, 6]. However, these studies differed from ours with respect to study design and patient population and thus used different cut-off P levels for mortality. The present study enrolled a relatively small number of patients and did not stratify serum P levels extensively, and thus a cut-off serum P level for highest risk for mortality cannot be determined from the present study. Nevertheless, the trend for the association between mortality and high serum P levels demonstrated in this study was similar to previous reports. The present study did not find an association between serum P $< 2.4$ mg/dL and mortality. Low P levels ($\leq 3.5$ mg/dL) at baseline were found to be significantly associated with higher rates for CV mortality in a 3-year HD cohort study in Japan [16]. Another report from a French HD cohort failed to demonstrate a significant association between low P levels ($\leq 2.79$ mg/dL) at baseline and mortality over 30 months of follow-up [13]. A report from the United Kingdom demonstrated a lower hazard ratio for all-cause mortality in incident dialysis patients with serum P levels $< 3.5$ mg/dL [12]. In the HEMO study, lower levels of serum P ($\leq 4.0$ mg/dL) did not increase the hazard ratio for all-cause mortality [17]. These inconsistencies highlight the need for further studies in order to clarify the association between low serum P levels and mortality in dialysis patients.
As previously established, high iPTH levels were associated with an increased risk for mortality in HD patients. A J-curve association between PTH levels and risk for mortality has previously been reported [4, 7, 16, 18]. We evaluated the association between KDIGO recommended serum iPTH levels (144–648 pg/mL based on our laboratory test) and risk for all-cause and CV mortality. We found that serum iPTH levels <144 pg/mL and >648 pg/mL were significantly associated with an increased risk for all-cause mortality, but not for CV mortality. This result is different from the result previously reported by Fouque et al. which did not find a J-curve relationship between iPTH levels and mortality risk [13]. According to the KDIGO recommended target range for serum iPTH levels, iPTH levels >585 pg/mL were not significantly associated with an increased risk for mortality. The authors attributed this finding to the fact that relatively few patients with serum iPTH levels exceeding 700 pg/mL were included in the study [13]. Our study failed to demonstrate a positive association between high serum iPTH levels and CV mortality in a sample of 1126 patients, and it is possible that this finding may be due to the relatively small sample size analyzed.

We also examined the relationship between achieving KDIGO determined target levels of all three CKD-MBD markers and the risk for all-cause and CV mortality. Case-mix adjusted and multivariable adjusted logistic regression analyses revealed that patients who achieved target serum Ca levels demonstrated a lower risk for all-cause mortality compared to those who achieved target serum P or iPTH levels. When examining the association between simultaneously achieving target levels for two CKD-MBD markers and the risk for mortality, we found that patients who achieved target P + iPTH levels had a lower risk for all-cause and CV mortality than those who achieved either Ca + P or Ca + iPTH targets by fully adjusted model analysis. Based on this finding, we suggest that achieving target serum Ca level or simultaneously achieving P + iPTH levels should be prioritized in the management of CKD-MBD in prevalent HD patients.

Although these results provide valuable CKD-MBD management data obtained over a long period of 5 years, the present study still has some limitations. First, the study was retrospective and the study population was limited to patients in a single HD center. Thus, center-specific effects cannot be
Table 5: Odds ratio of all-cause and CV mortality by achievement of KDIGO clinical guideline, iPTH target.

<table>
<thead>
<tr>
<th>iPTH (pg/mL), all-cause</th>
<th>OR (95% CI)</th>
<th>Model 1</th>
<th>Model 2</th>
<th>Model 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;144</td>
<td>1.70 (1.25~2.33)</td>
<td>2.00 (1.31~3.05)</td>
<td>1.95 (1.21~3.14)</td>
<td>0.006</td>
</tr>
<tr>
<td>144~648</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>&gt;648</td>
<td>1.67 (1.03~2.70)</td>
<td>2.14 (1.21~3.77)</td>
<td>1.47 (0.77~2.81)</td>
<td>0.24</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>iPTH (pg/mL), CV</th>
<th>OR (95% CI)</th>
<th>Model 1</th>
<th>Model 2</th>
<th>Model 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;144</td>
<td>1.12 (0.60~2.12)</td>
<td>1.20 (0.54~2.70)</td>
<td>1.68 (0.70~4.02)</td>
<td>0.24</td>
</tr>
<tr>
<td>144~648</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>&gt;648</td>
<td>0.69 (0.21~2.31)</td>
<td>1.00 (0.28~3.54)</td>
<td>0.54 (0.13~2.21)</td>
<td>0.39</td>
</tr>
</tbody>
</table>

Logistic regression model:
Model 1: unadjusted.
Model 2: adjusted for age, sex, hemodialysis vintage, etiology of renal failure, EPO, vitamin-D analogs, antihypertensive agent, iron use, and parathyroidectomy.
Model 3: model 2 + baseline laboratory results (albumin, hemoglobin, cholesterol, triglyceride, glucose (fasting), ferritin, Ca, P, potassium, uric acid, Kt/V urea, and cardiac-thoracic ratio).

Table 6: Odds ratio of all-cause and CV mortality by achievement of KDIGO clinical guideline.

<table>
<thead>
<tr>
<th>All-cause</th>
<th>OR (95% CI)</th>
<th>Model 1</th>
<th>Model 2</th>
<th>Model 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>All</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>Ca + P</td>
<td>1.52 (1.05~2.21)</td>
<td>1.85 (1.16~2.95)</td>
<td>1.43 (0.77~2.68)</td>
<td>0.25</td>
</tr>
<tr>
<td>Ca + iPTH</td>
<td>0.96 (0.60~1.52)</td>
<td>1.39 (0.81~2.39)</td>
<td>1.32 (0.46~3.77)</td>
<td>0.60</td>
</tr>
<tr>
<td>P + iPTH</td>
<td>1.07 (0.46~2.52)</td>
<td>1.50 (0.59~3.78)</td>
<td>1.32 (0.46~3.77)</td>
<td>0.60</td>
</tr>
<tr>
<td>Ca</td>
<td>1.85 (1.14~3.00)</td>
<td>4.02 (2.12~7.63)</td>
<td>&lt;0.001</td>
<td>3.16 (1.52~6.39)</td>
</tr>
<tr>
<td>P</td>
<td>3.95 (1.65~9.46)</td>
<td>4.54 (1.61~12.8)</td>
<td>0.004</td>
<td>3.55 (1.05~12.0)</td>
</tr>
<tr>
<td>iPTH</td>
<td>2.51 (1.08~5.83)</td>
<td>4.69 (1.56~14.1)</td>
<td>0.005</td>
<td>4.70 (1.44~15.4)</td>
</tr>
<tr>
<td>None</td>
<td>2.19 (1.06~4.52)</td>
<td>1.99 (0.77~5.13)</td>
<td>0.15</td>
<td>3.91 (1.00~15.3)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>CV</th>
<th>OR (95% CI)</th>
<th>Model 1</th>
<th>Model 2</th>
<th>Model 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>All</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>Ca + P</td>
<td>1.30 (0.60~2.85)</td>
<td>1.30 (0.49~3.41)</td>
<td>1.39 (0.51~3.77)</td>
<td>0.51</td>
</tr>
<tr>
<td>Ca + iPTH</td>
<td>1.75 (0.78~3.92)</td>
<td>2.58 (1.04~6.39)</td>
<td>2.96 (1.08~8.13)</td>
<td>0.03</td>
</tr>
<tr>
<td>P + iPTH</td>
<td>0.77 (0.10~5.96)</td>
<td>1.40 (0.17~11.9)</td>
<td>1.01 (0.11~9.66)</td>
<td>0.99</td>
</tr>
<tr>
<td>Ca</td>
<td>1.39 (0.50~3.89)</td>
<td>4.13 (1.32~13.0)</td>
<td>4.35 (1.20~15.9)</td>
<td>0.02</td>
</tr>
<tr>
<td>P</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>iPTH</td>
<td>3.71 (1.01~13.6)</td>
<td>3.47 (0.62~19.4)</td>
<td>2.40 (0.40~14.3)</td>
<td>0.33</td>
</tr>
<tr>
<td>None</td>
<td>1.58 (0.35~7.14)</td>
<td>0.82 (0.10~6.86)</td>
<td>1.43 (0.12~16.9)</td>
<td>0.77</td>
</tr>
</tbody>
</table>

Logistic regression model:
Model 1: unadjusted.
Model 2: adjusted for age, sex, hemodialysis vintage, etiology of renal failure, EPO, vitamin-D analogs, antihypertensive agent, iron use, and parathyroidectomy.
Model 3: model 2 + baseline laboratory results (albumin, hemoglobin, cholesterol, triglyceride, glucose (fasting), ferritin, Ca, P, iPTH, potassium, uric acid, Kt/V urea, and cardiac-thoracic ratio).

avoided. Second, CKD-MBD care was managed by multiple nephrologists in one center. Thus, differences in clinical practice among the different nephrologists could have resulted in heterogeneous CKD-MBD management. Unfortunately, the impact of this limitation on the clinical analysis performed in the present study cannot be addressed. Third, the sample size was relatively small, and other traditional and/or nutritional factors that may influence survival in HD patients were not evaluated in the present study. Finally, the effects of different drugs (e.g., cinacalcet or P-binders such as lanthanum carbonate, sevelamer hydrochloride, and sevelamer carbonate) on HD patient survival were not evaluated. However, these drugs are not covered by medical reimbursement schemes in Taiwan, and thus their use in Taiwanese HD patients is limited. Furthermore, we believe that these drugs would not have had a significant influence on the results presented in this study.

5. Conclusions

The present study provides valuable data on the management of CKD-MBD in HD patients followed up at one HD center...
over a five-year period. The results presented herein, with regard to the association between different CKD-MBD markers and the risk for all-cause and CV mortality in prevalent HD patients, are generally accordant with those cited by previous epidemiological studies. When applying KDIGO recommended target ranges for CKD-MBD markers, the present study demonstrated that patients who achieved the target range for serum Ca levels had lower risk for mortality compared to those who achieved the target range for serum P or iPTH levels. Furthermore, patients who achieved the target ranges for both P and iPTH levels demonstrated a significantly lower risk for mortality compared to those who achieved target ranges for Ca and P levels or Ca and iPTH levels.

**Competing Interests**

The authors declare that there is no conflict of interests regarding the publication of this paper.

**Acknowledgments**

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**References**


Research Article
Changes in Serum Concentrations of Fibroblast Growth Factor 23 and Soluble Klotho in Hemodialysis Patients after Total Parathyroidectomy

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Background. We examined the changes in circulating fibroblast growth factor 23 (FGF23) and Klotho concentrations in hemodialysis patients after parathyroidectomy (PTX).

Methods. We enrolled a cohort of hemodialysis patients who received PTX. Postoperatively, patients received calcium supplements and/or vitamin D analogue (calcitriol) to maintain serum calcium within 7.0–8.0 mg/dL. Information on clinical parameters including bone-mineral metabolic variables was collected pre-PTX and on days 5 and 90 after PTX. Concomitantly, serum full-length FGF23 and α-Klotho levels were measured. The relationship between FGF23 and clinical parameters was analyzed by single linear regression.

Results. Forty-six participants (33 women; 13 men) were enrolled in the study. Their mean age was 56.49 years. Serum FGF23 and α-Klotho concentrations were elevated on days 5 and 90 after PTX compared to baseline (p > 0.05). Serum FGF23 concentrations negatively correlated with serum calcium concentrations pre-PTX (Beta −0.31; R² 0.0949; p = 0.040), day 5 post-PTX (Beta −0.31; R² 0.0982; p = 0.036), and day 90 post-PTX (Beta −0.39; R² 0.1528; p = 0.008).

Conclusions. There was no change in circulating FGF23 and Klotho concentrations after PTX in hemodialysis patients given postoperative calcium supplements and/or vitamin D analogue. Serum FGF23 concentrations pre-PTX and at days 5 and 90 after PTX were inversely related to serum calcium concentrations.

1. Introduction

Secondary hyperparathyroidism (SHPT) is a common complication of chronic kidney disease (CKD) and occurs as a consequence of calcium, phosphate, and vitamin D homeostasis. Recently, fibroblast growth factor 23 (FGF23) has been identified as a protein which plays a crucial role in phosphate regulation. FGF23 is primarily secreted by osteocytes and is involved in controlling the metabolism of phosphate, parathyroid hormone (PTH), and 1,25 dihydroxyvitamin D (1,25(OH)2D3) [1, 2]. In the early stage of CKD, elevated FGF23 levels increase fractional phosphate excretion and subsequently reduce serum phosphate and 1α-hydroxylase levels. These changes in turn reduce 1,25(OH)2D3 formation and increase PTH secretion [3–5]. In the late stage of CKD, FGF23 cannot regulate phosphate homeostasis and it adversely affects outcome in CKD by contributing to disease progression, left ventricular hypertrophy, and increased mortality [5–8].

Klotho is an antiaging gene, which encodes a transmembrane protein that forms a complex with multiple growth factor receptors [9]. FGF23 exerts its effects by forming a complex with Klotho [10]. Klotho also acts as a humoral factor when it is cleaved and released into the circulation.
Previous studies have demonstrated that the circulating soluble form of Klotho can enhance the excretion of phosphate in the proximal nephron and promote calcium reabsorption in the distal nephron [11, 12]. The concentrations of circulating FGF23 and Klotho were found to progressively decline after parathyroidectomy (PTX). However, circulating Klotho levels only decreased transiently before increasing progressively thereafter [13].

The literature on serial changes in circulating FGF23 and Klotho concentrations after PTX in hemodialysis (HD) patients is scarce, and thus we conducted a study to monitor these values in a cohort of HD patients after PTX. We aimed to examine the changes in circulating FGF23 and Klotho concentrations until 90 days after PTX. We also evaluated the relationships between clinical parameters and circulating concentrations of FGF23 and Klotho after PTX.

2. Materials and Methods

2.1. Patients. We enrolled HD patients who received PTX at Kaohsiung Chang Gung Memorial Hospital in Taiwan. The patients were tracked from January 1, 2014, until December 31, 2016. The inclusion criteria were (1) patients receiving regular HD for at least 3 months and (2) follow-up for at least 3 months after PTX. The exclusion criteria were (1) patient age greater than 90 years old; (2) patients transferred to other medical facilities; and (3) patients whose information was incomplete and/or those who were lost to follow-up after PTX. The indications for PTX included uncontrolled pruritus, generalized bone pain, resistance to medical treatment, and high intact PTH (iPTH) levels (>1000 pg/mL). The PTX procedure included a total parathyroidectomy and autotransplantation of 140 mg of hyperplastic tissue of the parathyroid gland into subcutaneous forearm tissue. All patients received calcium supplements and/or vitamin D analogue (calcitriol) on day 1 after PTX to maintain serum calcium concentrations around 7.0–8.0 mg/dL.

2.2. Laboratory Measurements. Routine hematological and biochemical investigations were performed one day prior to PTX (baseline). Serum albumin, calcium (Ca), phosphate (P), alkaline phosphatase (ALP), and bone-alkaline phosphatase (B-ALP) concentrations were measured again at 5 days and 90 days after PTX. All blood samples were measured using commercial kits and an autoanalyzer (Hitachi 7600-210, Diagnostic Indianapolis, USA). B-ALP was measured by colorimetric assay as the hydrolysis of p-nitrophenyl phosphate according to instructions from the supplier (Roche Diagnostic Indianapolis, USA). The concentrations of circulating FGF23 and Klotho after PTX were measured using a commercially available enzyme-linked immunosorbent assay (ELISA) kit (DiaSorin, USA). Serum Klotho concentration was measured using human soluble α-Klotho assay kit (Immuno-Biological Laboratories, Japan). Serum 25-hydroxyvitamin D concentration was measured using human soluble 25-hydroxyvitamin D assay kit (DiaSorin, USA).

2.3. Statistics. The baseline characteristics were summarized as total number, percentage, and mean, SD. Changes in FGF23 and α-Klotho before PTX and after PTX (at 5 and 90 days) were evaluated by independent one-way repeated ANOVA. The FGF23 and α-Klotho measurements before PTX and after PTX (day 5 and day 90) were visually summarized using a boxplot graph. We evaluated the relationship between FGF23 and other parameters using single linear regression. A p value < 0.05 was considered statistically significant. All statistical analyses were performed by STATA (version 11.1).

3. Results

3.1. Participant Demographic Characteristics. A total of 46 participants were enrolled in the final study. The mean age of the study population was 56.49 years, and the ratio of women: men was 33:13. The majority of patients (n = 40/46) had comorbid hypertension. Laboratory parameters at the baseline showed markedly elevated levels of ALP, B-ALP, and iPTH (Table 1). Compared to baseline, there was no significant increase in the serum concentrations of FGF23 and α-Klotho at 90 days after PTX (Figure 1) (the detailed data sheet was shown in the Supplementary Table 1 in Supplementary Material available online at http://dx.doi.org/10.1155/2016/6453803).

3.2. Correlation between FGF23 Concentrations and Mineral Bone Metabolism Parameters. We performed single linear regression analysis to investigate the relationship between serial changes in FGF23 concentrations (baseline and 5 and 90 days post-PTX) and mineral bone metabolism parameters. There was a significant negative correlation between serum FGF23 concentrations and serum calcium concentrations at baseline (Beta −0.31; R^2 0.0949; p = 0.040), at 5 days post-PTX (Beta −0.31; R^2 0.0982; p = 0.036), and at 90 days post-PTX (Beta −0.39; R^2 0.1528; p = 0.008) (Table 2). We assumed a closer approximation to a standard distribution curve when FGF23 concentrations were transformed to logarithms. The results showed increased correlation between log FGF23 concentrations and serum creatinine concentrations from baseline to 90-day PTX (no statistical significance). Serum log FGF23 concentrations were negatively correlated with serum calcium concentrations from baseline to 90 days post-PTX (only significant at 90 days post-PTX, Beta −0.32; R^2 0.1002; p = 0.034). Moreover, there were no significant correlations between either FGF23 or log FGF23 concentrations and mineral bone metabolism markers [P, calcium-phosphate product (Ca×P), intact PTH, ALP, B-ALP, 25(OH)2D3] from baseline to 90-day PTX (Table 2).
Figure 1: The serum concentrations of FGF23 and α-Klotho in the study period. The boxes represent the medium values and the vertical lines the maximum and minimum values in the participants measured.

### Table 1: Baseline characteristics in study subjects (n = 46).

<table>
<thead>
<tr>
<th>Variables</th>
<th>n</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (yrs) (mean, SD)</td>
<td>56.49</td>
<td>9.95</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>33</td>
<td>71.74</td>
</tr>
<tr>
<td>Male</td>
<td>13</td>
<td>28.26</td>
</tr>
<tr>
<td>HD vintage (yrs) (mean, SD)</td>
<td>6.14</td>
<td>4.31</td>
</tr>
<tr>
<td>Vitamine D use</td>
<td>8</td>
<td>16.33</td>
</tr>
<tr>
<td>Calcium salt use</td>
<td>8</td>
<td>16.33</td>
</tr>
<tr>
<td>Comorbidity</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diabetes mellitus</td>
<td>6</td>
<td>13.04</td>
</tr>
<tr>
<td>Hypertension</td>
<td>40</td>
<td>86.96</td>
</tr>
<tr>
<td>Coronary</td>
<td>2</td>
<td>4.35</td>
</tr>
<tr>
<td>SBP (mean, SD)</td>
<td>139.10</td>
<td>26.46</td>
</tr>
<tr>
<td>DBP (mean, SD)</td>
<td>78.10</td>
<td>14.04</td>
</tr>
<tr>
<td>Lab parameters (mean, SD)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hb (g/dL)</td>
<td>11.13</td>
<td>1.73</td>
</tr>
<tr>
<td>Hct (%)</td>
<td>33.97</td>
<td>5.18</td>
</tr>
<tr>
<td>Albumin (g/dL)</td>
<td>4.82</td>
<td>4.59</td>
</tr>
<tr>
<td>BUN</td>
<td>41.39</td>
<td>20.99</td>
</tr>
<tr>
<td>Ca (mg/dL)</td>
<td>9.21</td>
<td>10.58</td>
</tr>
<tr>
<td>P (mg/dL)</td>
<td>9.82</td>
<td>1.33</td>
</tr>
<tr>
<td>CaXp (mg^2/dL^2)</td>
<td>54.40</td>
<td>20.52</td>
</tr>
<tr>
<td>ALP (U/L)</td>
<td>229.94</td>
<td>197.18</td>
</tr>
<tr>
<td>Bone-ALP (µg/L)</td>
<td>65.00</td>
<td>39.43</td>
</tr>
<tr>
<td>Al (µg/dL)</td>
<td>2.26</td>
<td>1.14</td>
</tr>
<tr>
<td>Ferritin (ng/mL)</td>
<td>363.84</td>
<td>234.53</td>
</tr>
<tr>
<td>iPTH (pg/ml)</td>
<td>1391.22</td>
<td>746.68</td>
</tr>
<tr>
<td>25(OH)D3 (ng/mL)</td>
<td>14.54</td>
<td>7.48</td>
</tr>
</tbody>
</table>

HD, hemodialysis; SBP, systolic blood pressure; DBP, diastolic blood pressure; Hb, hemoglobin; Hct, hematocrit; BUN, blood urea nitrogen; Cr, creatinine; Ca, calcium; P, phosphate; CaXp, calcium-phosphate product; ALP, alkaline phosphatase, Al, aluminum; iPTH, intact parathyroid hormone.

3.3. Relationship between FGF23 and α-Klotho Concentrations. FGF23 concentrations and α-Klotho concentrations were not significantly correlated at any time point (baseline and 5 and 90 days post-PTX) (Figure 2).

4. Discussion

In the present study, we examined the serial changes in serum FGF23 and α-Klotho concentrations in HD patients after PTX. There were no significant changes in the concentrations of FGF23 or α-Klotho at baseline, 5 days post-PTX, or 90 days post-PTX. In addition, we did not find a significant correlation between FGF23 and α-Klotho concentrations during the study period. These results differ from previous studies, which showed a progressive decrease in FGF23 concentrations from pre-PTX to 10 days after PTX [13, 14]. In another study, FGF23 concentrations at days 1 and 3 correlated with serum phosphorus and calcium-phosphorus levels [14]. One report from Takahashi et al. [13] showed a progressive decline in FGF23 concentrations together with significant reductions in serum Ca, P, and iPTH concentrations from pre-PTX to 30 days after PTX and stable FGF23 concentrations from 30 to 90 days after PTX. This is in contrast to the results of another study, which examined changes in FGF23 levels after PTX for primary hyperparathyroidism and demonstrated no change between day 1 and 6 weeks after PTX. Meanwhile, serum FGF23 concentrations did not correlate with serum P, Ca, iPTH, 25(OH)2D, or B-ALP concentrations in the pre-PTX and post-PTX state [15]. The present study measured serum FGF23 concentrations at three time points: pre-PTX and 5 days and 90 days after PTX. Our patients received calcium supplements and vitamin D analogue (calcitriol) after PTX to keep serum calcium concentrations in the range of 7.0–8.0 mg/dL. It is worth noting that certain diets and medications can also modify FGF23 concentrations including a vegetarian diet, phosphate restriction or phosphate binders, vitamin D analogues, parathyroidectomy, cinacalcet, and kidney transplantation [16]. In parathyroidectomized...
### Table 2: Relationships between FGF-23 and clinical parameters.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Baseline</th>
<th>Day 5</th>
<th>3 months</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Beta</td>
<td>$R^2$</td>
<td>$p$ value</td>
</tr>
<tr>
<td>FGF-23</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cr</td>
<td>-0.13</td>
<td>0.0161</td>
<td>0.402</td>
</tr>
<tr>
<td>Ca (mg/dL)</td>
<td>-0.31</td>
<td>0.0949</td>
<td>0.040</td>
</tr>
<tr>
<td>P (mg/dL)</td>
<td>-0.01</td>
<td>0.0001</td>
<td>0.949</td>
</tr>
<tr>
<td>iPTH (pg/ml)</td>
<td>-0.10</td>
<td>0.0098</td>
<td>0.514</td>
</tr>
<tr>
<td>25(OH)D$_3$ (ng/mL)</td>
<td>0.09</td>
<td>0.0087</td>
<td>0.538</td>
</tr>
<tr>
<td>ALP (U/L)</td>
<td>0.15</td>
<td>0.0217</td>
<td>0.414</td>
</tr>
<tr>
<td>Bone-ALP (µg/L)</td>
<td>0.23</td>
<td>0.0521</td>
<td>0.413</td>
</tr>
<tr>
<td>CaxP (mg$^2$/dL$^2$)</td>
<td>-0.09</td>
<td>0.008</td>
<td>0.556</td>
</tr>
</tbody>
</table>

**log FGF-23**

| Cr                        | -0.19    | 0.0355  | 0.210    | -0.18    | 0.0314  | 0.238    | 0.20     | 0.0415  | 0.174    |
| Ca (mg/dL)                | -0.21    | 0.0440  | 0.167    | -0.22    | 0.0487  | 0.145    | -0.32    | 0.1002  | 0.034    |
| P (mg/dL)                 | 0.09     | 0.0080  | 0.559    | -0.01    | 0.0001  | 0.949    | 0.11     | 0.0129  | 0.458    |
| iPTH (pg/ml)              | -0.04    | 0.0018  | 0.778    | -0.10    | 0.0097  | 0.515    | -0.07    | 0.0044  | 0.661    |
| 25(OH)D$_3$ (ng/ml)       | -0.08    | 0.0063  | 0.600    | -0.08    | 0.0058  | 0.615    | 0.12     | 0.0153  | 0.413    |
| ALP (U/L)                 | 0.12     | 0.0150  | 0.497    | 0.25     | 0.0626  | 0.160    | 0.18     | 0.0339  | 0.305    |
| Bone-ALP (µg/L)           | 0.15     | 0.0216  | 0.601    | 0.25     | 0.0642  | 0.362    | -0.08    | 0.0059  | 0.786    |
| CaxP (mg$^2$/dL$^2$)      | 0.03     | 0.0007  | 0.859    | -0.06    | 0.0041  | 0.677    | 0.02     | 0.0003  | 0.903    |

**Figure 2:** The correlation between serum FGF23 and α-Klotho concentrations in the study period.
fluids [22]. cKL stimulates FGF23 production and is a product of mKL (cKL) [23]. Only the cKL protein was detected in the blood, urine, and cerebrospinal fluid [11, 22]. The α-Klotho gene product is expressed in a membrane-bound form (mKL) in association with FGF23 and FGF receptors (FGFRs) and signals through the MAPK cascade [10, 20]. Two soluble species have been reported: an alternatively spliced secreted form (sKL) and an endoproteolytic cleavage product (mKL) in association with FGF23 and FGF receptors (FGFRs) and signals through the MAPK cascade [10, 20]. The α-Klotho gene product is expressed in a membrane-bound form (mKL) in association with FGF23 and FGF receptors (FGFRs) and signals through the MAPK cascade [10, 20]. Two soluble species have been reported: an alternatively spliced secreted form (sKL) and an endoproteolytic cleavage product (mKL) (cKL) [23]. Only the cKL protein was detectable in human and rodent plasma and cerebrospinal fluid [22]. cKL stimulates FGF23 production in vivo and results in elevated FGF23 levels [24]. Takahashi et al. [13] reported a 13% decrease from baseline in serum soluble Klotho concentrations on the day after PTX and a subsequent increase to peak levels of 34% above baseline at 90 days post-PTX [13]. The present study showed a nonsignificant increase in serum α-Klotho concentrations after PTX on days 5 and 90 (Supplementary Table 1). This is in keeping with the findings reported by Takahashi et al. [13].

We found a negative correlation between FGF23 concentrations and serum calcium concentrations at pre-PTX and days 5 and 90 after PTX. However, there was no significant correlation between FGF23 concentrations and serum P, iPTH, or 25(OH) D3 concentrations. This is in contrast to the report by Sato et al. [14], which demonstrated a correlation between FGF23 concentrations on days 1 and 3 after PTX and serum phosphate and calcium-phosphate product levels [14]. Of note, serum calcium concentrations decreased to less than 8 mg/dL on day 3 after PTX and calcium supplements were not administered to any patient before day 2 in the aforementioned study [14]. In contrast, our patients received calcium supplements and calcitriol one day after PTX to maintain serum calcium concentrations above 7.0 mg/dL. It is possible that this discrepancy in study protocol contributed to the different results obtained between the two studies.

The present study has some limitations. Firstly, our patients received calcium supplements and/or vitamin D analogue one day after PTX. These interventions may alter calcium-phosphate homeostasis in dialysis patients and further complicate the complex regulation of FGF23 and Klotho post-PTX. Therefore, it is not yet possible to accurately outline the mechanism whereby PTH regulates FGF23 and Klotho production after PTX in dialysis patients. Secondly, a study in wild-type mice placed on an iron-deficient diet showed an increase in Fgf23 mRNA expression in bone, as well as increased circulating levels of C-terminal FGF23 fragments in the absence of any change in intact FGF23 levels [25]. Since the present study measured serum full-length FGF23 concentrations, we cannot be certain of the contribution of circulating FGF23 fragments to the values obtained in the assay. A similar problem was encountered in the measurement of circulating Klotho levels. It would be useful to obtain a response curve of circulating Klotho levels after PTX in dialysis patients using separated component measurements for Klotho. Furthermore, the present study did not measure circulating cKL concentrations, which may play a role in stimulating FGF23 production [24]. Furthermore, this clinical study cannot propose a mechanism to explain how FGF23 regulates bone-mineral metabolism after PTX. Finally, the present study enrolled a relatively small number of participants. A well-designed randomized controlled study enrolling more participants is needed to further validate the results of the current study. Despite the limitations stated above, the present study provides a clinically relevant analysis of changes in FGF23 and Klotho levels after PTX in dialysis patients. Future studies aimed at following up these changes over a longer period of time and delineating the interactions between calcium supplements/vitamin D analogues and FGF23/Klotho are warranted.

5. Conclusions
The present study demonstrated no change in circulating FGF23 and Klotho concentrations after PTX in HD patients receiving calcium supplement and/or vitamin D analogues postoperatively. There was a negative correlation between serum FGF23 and calcium concentrations at pre-PTX and on days 5 and 90 after PTX.

Competing Interests
The authors declare that there are no competing interests regarding the publication of this paper.

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