SERUM 25-HYDROXYVITAMIN D LEVEL COULD PREDICT THE RISK FOR PERITONEAL DIALYSIS-ASSOCIATED PERITONITIS

Hai-Chen Pi,^{1*} Ye-Ping Ren,^{2*} Qin Wang,² Rong Xu,¹ and Jie Dong¹

Renal Division,¹ Department of Medicine, Institute of Nephrology, Peking University, Beijing, China; and Department of Nephrology,² Second Affiliated Hospital of Harbin Medical University, Heilongjiang, China

◆ **Background:** As an immune system regulator, vitamin D is commonly deficient among patients on peritoneal dialysis (PD), which may contribute to their impaired immune function and increased risk for PD-related peritonitis. In this study, we aimed to investigate whether vitamin D deficiency could predict the risk of peritonitis in a prospective cohort of patients on PD.

◆ **Methods:** We collected 346 prevalent and incident PD patients from 2 hospitals. Baseline demographic data and clinical characteristics were recorded. Serum 25-hydroxyvitamin D (25[OH]D) was measured at baseline and prior to peritonitis. The mean doses of oral active vitamin D used during the study period were also recorded. The outcome was the occurrence of peritonitis.

◆ **Results:** The mean age of patients and duration of PD were 58.95 ± 13.67 years and 28.45 ($15.04 - 53.37$) months, respectively. Baseline 25(OH)D level was 16.15 ($12.13 - 21.16$) nmol/L, which was closely associated with diabetic status, longer PD duration, malnutrition, and inflammation. Baseline serum 25(OH)D predicted the occurrence of peritonitis independently of active vitamin D supplementation with a hazard ratio (HR) of 0.94 (95% confidence interval [CI] $0.90 - 0.98$) after adjusting for recognized confounders (age, gender, dialysis duration, diabetes, albumin, residual renal function, and history of peritonitis). Compared to the low tertile, middle and high 25(OH)D level tertiles were associated with a decreased risk for peritonitis with HRs of 0.54 (95% CI $0.31 - 0.94$) and 0.39 (95% CI $0.20 - 0.75$), respectively.

◆ **Conclusions:** Vitamin D deficiency evaluated by serum 25(OH)D rather than active vitamin D supplementation is closely associated with a higher risk of peritonitis.

a variety of infectious diseases. During the past few years, the link between vitamin D deficiency and infection susceptibility has been investigated in the general population (3–5).

The prevalence of vitamin D deficiency evaluated by serum 25-hydroxyvitamin D (25[OH]D) levels was found to be substantially high, ranging from 50% to 98% in the dialysis population (6–9). It is also well established that patients on dialysis suffer from an impaired immune system and are highly susceptible to infections compared to the general population (10). For patients on peritoneal dialysis (PD), peritonitis is the most common infectious complication, contributing to treatment failure, hospitalization, and death. Whether vitamin D deficiency contributes to the increased risk for PD-related peritonitis and poor outcome is uncertain. To date, evidence concerning this theory is limited, with only 2 retrospective studies having shown a positive association between oral active vitamin D therapy and the decreased risk for peritonitis (11,12). Both studies, however, failed to report serum 25(OH)D levels for either the intervention or control groups, leaving uncertainty about the association between vitamin D deficiency and peritonitis risk in each case.

Therefore, we aimed to determine whether serum 25(OH)D levels could predict the occurrence of peritonitis in a large cohort of PD patients from 2 PD units. Of note, the external active vitamin D supplementation, comorbidity load, and nutritional status may be potential confounders for the association of serum 25(OH)D and peritonitis risk. And, individuals with vitamin D deficiency are prone to be weak and malnourished (13). Therefore, active vitamin D supplementation, comorbidity load, nutritional status, and physical performance would need to be taken into account in determining the above link.

SUBJECTS AND METHODS

STUDY DESIGN AND SUBJECTS

This prospective cohort study was carried out at the PD center of Peking University First Hospital and the Second

Correspondence to: Jie Dong, Renal Division and Institute of Nephrology, Peking University First Hospital, 100034 Beijing, P. R. China.

dongjie@medmail.com.cn

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The nonclassical effects of vitamin D on the immune system, cardiovascular disease, and cancer, unrelated to its effects on bone metabolism, have been broadly investigated in recent years. Among these, the immunomodulating actions of vitamin D have received considerable attention. Vitamin D signaling pathways regulate both innate and adaptive immunity, thereby maintaining the associated inflammatory response within physiological limits (1,2). Vitamin D deficiency contributes to microinflammation and a defective immune system, which are associated with

* Hai-Chen Pi and Ye-Ping Ren contributed equally to this work.

Affiliated Hospital of Harbin Medical University. Inclusion criteria for participants were: (1) age ≥ 18 years; and (2) prevalent and incident patients between March 1, 2011, and November 31, 2011. Patients were excluded if they had systemic infection, acute cardiovascular events, active hepatitis or tumor, operation, or trauma during 1 month prior to study. All patients were followed until death, transfer to hemodialysis, renal transplantation or May 31, 2013 (the end of study). All the subjects received conventional glucose-based, lactate-buffered PD solutions (Ultrabag, Baxter Healthcare, Guangzhou, China). The Ethics Committees of the 2 hospitals approved this study protocol, and written informed consent was obtained from each participant.

BASIC DEMOGRAPHIC AND LABORATORY DATA

Basic demographic data, including age, gender, body weight, height, duration of PD, comorbidities, and the presence of peritonitis history, were collected at baseline. The Charlson index was used to assess the comorbidity load. Physical performance was assessed using the Karnofsky index (14). All laboratory samples except for serum 25(OH)D were analyzed by standard laboratory techniques in the local hospitals including hemoglobin (Hb), serum albumin (ALB), calcium, phosphate, intact parathyroid hormone (iPTH), and high sensitive C-reactive protein (hs-CRP). Dialysis adequacy and residual renal function (RRF) were also measured. Dialysis adequacy was defined as weekly total Kt/V (TKt/V) and total creatinine clearance (Tccr). Residual renal function was defined as the mean of residual creatinine clearance and residual urea clearance. The normalized protein equivalent of total nitrogen appearance (nPNA) was calculated using the Bergstrom formula (15). Serum iPTH was measured by the chemiluminescence assay (reference range: 15 ~ 65 pg/mL). Serum hs-CRP was measured by immune rate nephelometric analysis.

SERUM 25(OH)D MEASUREMENTS AND ORAL VITAMIN D SUPPLEMENTS

Extra serum samples were obtained at baseline and then at 6-month intervals during follow-up. Samples were centrifuged at 3,000 rpm for 10 minutes, and then stored at -80°C until analysis for 25(OH)D. Baseline serum 25(OH)D was measured for each subject. For subjects that developed peritonitis, frozen samples at the nearest time point prior to peritonitis onset were examined for serum 25(OH)D. For subjects who experienced more than 1 episode of peritonitis during the follow-up period, we measured serum 25(OH)D prior to their first episode only. Serum 25(OH)D was measured by enzyme-linked immunosorbent assay (ELISA, Immunodiagnostic Systems Ltd, Bolden, UK) in Peking University First Hospital. The ELISA kit we used can not cross react with 1,25(OH) $_2$ D.

During the study period, medication regimens and oral active vitamin D (calcitriol or alfacalcidol) supplements for each patient were recorded at 1-month intervals and mean weekly doses were then calculated. Alfacalcidol doses were converted to the calcitriol equivalent by multiplying by 0.75 (16).

DIAGNOSIS OF PERITONITIS

The outcome was defined as the occurrence of peritonitis during follow-up. Peritonitis was defined as the presence of at least 2 of the following conditions: (1) abdominal pain or tenderness, (2) presence of white blood cells (100 cells/mL) in peritoneal effluent, with at least 50% polymorphs, and (3) positive dialysate culture results. In cases where abdominal pain or tenderness were unavailable, cloudy fluid combined with at least 1 of the latter 2 conditions were considered diagnostic (17).

Examination of effluent included cell counts, causative micro-organisms, and drug sensitivities. For each episode of peritonitis, we recorded the initial effluent white blood cell count (day 0) and then the serial white blood cell counts on scheduled follow-up visits on days 1, 3, 5, 7, and 14. The disease severity score (DSS) (range: 0–5 points) was calculated as the sum of points for pain and fever (18).

TREATMENT FOR PERITONITIS

Peritonitis was treated with the standard antibiotic protocol, modified from the International Society for Peritoneal Dialysis (ISPD) guidelines (19). In general, initial antimicrobial therapy for peritonitis consisted of intra-peritoneal administration of a third generation cephalosporin plus cefazolin (both with a single dose of 1 g once daily). Antibiotic treatment was modified once the culture results and antimicrobial sensitivities became available. Patients were switched to hemodialysis and their PD catheters were subsequently removed if they showed a lack of improvement after 7–14 days of appropriate antibiotic therapy, if PD dialysate grew yeast species or tuberculosis at any time, and if a clinical tunnel infection was diagnosed at the start as suggested by the ISPD guidelines (19).

STATISTICAL ANALYSIS

Statistical analysis was performed by SPSS for Windows software version 13.0 (SPSS Inc., Chicago, IL, USA). Parametric data are presented as means $\pm$ standard deviation. Non-parametric data are presented as median values with their intervals from the 25th to 75th percentile. Categorical variables are expressed as a percentage or ratio.

Subjects were divided by baseline or pre-peritonitis serum 25(OH)D levels. Differences were compared using the analysis of variance (ANOVA), Fisher's exact test, chi-squared test, and Kruskal-Wallis H test between 3 groups, as appropriate, or by using the unpaired Student's *t*-test, Mann-Whitney U test, and chi-squared test between 2 groups, as appropriate.

We aimed to determine the prognostic value of baseline 25(OH)D for the incidence of peritonitis. Multivariate Cox regression models were built to determine this association, adjusting for: (1) age, gender, dialysis duration, diabetes, albumin, RRF and history of peritonitis; (2) Charlson index and Karnofsky index; (3) whether or not taking active vitamin D supplements, serum calcium, phosphate, and iPTH. For

these models, patients were censored for all other reasons for stopping PD, including death, transfer to hemodialysis, renal transplantation, or the study end date (May 31, 2013). The multivariate Cox regression models were built without forward and backward selection processes. Odds ratios (ORs), risk ratios (RRs) and their 95% CI were also calculated. All probabilities were 2-tailed, and the level of significance was set at 0.05.

RESULTS

DEMOGRAPHIC DATA AND CLINICAL CHARACTERISTICS

There were 572 patients receiving PD therapy in Peking University First Hospital and the Second Affiliated Hospital of Harbin Medical University between March and November 2011. In total, 61 patients were excluded based on the exclusion criteria and 165 patients refused to participate in this study. A total of 346 PD patients (155 men and 191 women; 324 prevalent and 22 incident patients) were included (Figure 1), and they had a mean age of 58.95 ± 13.67 years and duration on dialysis of $28.45 (15.04 - 53.37)$ months. Among these 346 PD patients, 133 (38.44%) had diabetes mellitus (DM) and 75 (21.68%) had peritonitis history. The baseline data showed averages of 111.49 ± 15.43 g/L for Hb and 36.66 ± 4.46 g/L for ALB.

Baseline serum 25(OH)D of all patients was $16.15 (12.13 - 21.16)$ nmol/L. Patients were divided into 3 groups according to tertiles of serum 25(OH)D. Compared to patients in the high tertile, those in the low tertile tended to be diabetic, hypoalbuminemic, and had lower RRF and higher Charlson index ($p < 0.05$). Patients in the middle and low tertile groups were more likely to have peritonitis history with a longer time on dialysis, and they also had higher Hs-CRP values ($p < 0.05$). Moreover, peritonitis patient-months increased according to serum 25(OH)D tertiles. There were no differences in age, gender, Hb, Tkt/V, calcium, phosphate, iPTH, nPNA, Karnofsky index, whether or not they were taking oral active vitamin D, and vitamin D doses for vitamin D users between groups (Table 1).

Figure 1 — Study flow chart. PD = peritoneal dialysis.

OUTCOMES

At the end of the study, 259 patients were still being maintained on PD. The median follow-up time was 16.48 (13.14 – 19.45) months. A total of 96 episodes of peritonitis in 74 (21.39%) PD patients were recorded during the observation period. The peritonitis rate was 58.38 patient-months. The time-to-peritonitis occurrence was 7.89 (3.17 – 12.90) months. Among 96 peritonitis cases, 30 episodes were due to gram-positive organisms, 23 gram-negative organisms, 4 due to polymicrobial infections, and 3 to fungi; there were 29 due to culture-negative organisms and 7 no culture; 75 (78.13%) episodes lead to treatment success, 14 were (14.58%) transferred to hemodialysis, and 7 (7.29%) patients died.

BASELINE SERUM 25(OH)D AND THE RISK FOR PERITONITIS

The predictive role of baseline 25(OH)D for the occurrence of peritonitis was examined in multivariate Cox regression models. After adjustment for age, gender, dialysis duration, DM, ALB, RRF, and history of peritonitis, baseline 25(OH)D (1 nmol/L) as a continuous variable was an independent predictor of peritonitis (HR = 0.94; 95% CI 0.90 – 0.98). When patients were tertiled by baseline serum 25(OH)D, Kaplan–Meier analysis showed that time-to-peritonitis episode was significantly shorter for patients in the low tertile compared to patients in the high tertile group ($p = 0.003$, Figure 2). In addition, baseline 25(OH)D as a categorical variable was used in multivariate Cox regression models. As shown in Table 2, the middle and high tertiles of baseline 25(OH)D levels were associated with a significantly decreased risk of peritonitis compared to the low tertile group (HR = 0.54; 95% CI 0.31 – 0.94; HR = 0.39; 95% CI 0.20 – 0.75, respectively; Model 1). The difference in risk between middle and high tertile groups did not reach statistical significance. After additionally adjusting for Charlson index and Karnofsky index (Model 2), or whether or not the patient was taking active vitamin D supplements (calcitriol or alfacalcidol), calcium, phosphate, and iPTH (Model 3), the prognostic value of baseline 25(OH)D levels as both continuous and categorical variables remained unchanged. Sensitivity analysis showed that the prognostic value of baseline 25(OH)D level did not change when we excluded subjects who had a history of peritonitis before enrollment (date not show). The use of active vitamin D supplements *per se* could not predict the risk for peritonitis (HR = 0.82; 95% CI 0.48 – 1.40). Furthermore, there was no interaction effect between the use of active vitamin D supplements and baseline 25(OH)D levels on the risk for peritonitis.

CLINICAL CHARACTERISTICS OF PERITONITIS BASED ON PRE-PERITONITIS SERUM 25(OH)D LEVEL

Of 96 episodes of peritonitis, 62 were examined with serum 25(OH)D within 6 months of the episode using frozen samples. The median pre-peritonitis 25(OH)D value was

TABLE 1
Baseline Demographic Data and Clinical Characteristics of Patients Tertiled by Baseline Serum 25(OH)D Levels

Variables	Tertile of Baseline Serum 25(OH)D (nmol/L)			P value ^a
	Low (<13.51) (n=115)	Middle (13.51–19.13) (n=116)	High (>19.13) (n=115)	
Age (years)	59.99±12.13	59.86±14.96	56.97±13.68	0.17
Male, n (%)	49 (42.61)	52 (44.83)	54 (46.96)	0.80
Dialysis duration (months)	38.80 (17.74–62.95)**	31.79 (16.53–58.75)*	24.35 (12.85–40.80)	0.01
Diabetes, n (%)	59 (51.30)***	40 (34.48)#	34 (29.57)	0.002
Charlson index	5 (3–8)**	4 (2–7)	2 (3–6)	0.01
Karnofsky index	80 (70–90)	80 (70–90)	90 (70–90)	0.08
Peritonitis history, n (%)	28 (24.35)*	31 (26.72)*	16 (13.91)	0.04
Number of previous peritonitis episodes ^b	2 (1–2)	1 (1–2)	1 (1–2)	0.09
Hemoglobin (g/L)	110.15±16.64	112.15±16.01	112.24±13.37	0.52
Albumin (g/L)	35.90±4.60*	36.63±4.63	37.50±4.01	0.02
Total Kt/V	1.84 (1.63–2.08)	1.79 (1.62–2.01)	1.91 (1.65–2.30)	0.16
Total CCr (L/1.73 m ² /week)	52.17 (46.40–62.90)	53.05 (42.20–66.35)	55.87 (47.50–81.58)	0.07
RRF (mL/min)	1.05 (0–2.92)*	1.11 (0–3.25)*	1.95 (0.56–4.30)	0.01
nPNA (g/kg/day)	0.82±0.18	0.87±0.23	0.86±0.19	0.11
Hs-CRP (mg/L)	3.93 (1.11–12.38)*	3.43 (1.47–10.47)*	1.95 (0.57–5.93)	0.01
Calcium (mmol/L)	2.39±0.24	2.35±0.25	2.31±0.27	0.07
Phosphate (mmol/L)	1.58±0.45	1.56±0.43	1.65±0.42	0.26
iPTH (pg/ml)	101.80 (32.32–192.20)	95.98 (48.09–229.80)	116.25 (48.80–267.03)	0.11
Taking oral active vitamin D (n, %)	40 (34.78)	45 (38.79)	44 (38.26)	0.12
Doses of oral active vitamin D for users (μg/week) ^c	0.33 (0.13–0.86)	0.61 (0.25–1.20)	0.64 (0.32–1.06)	0.61
Peritonitis rate (patients-months)	38.23	69.67	95.38	—
Peritonitis during follow-up (n, %)	35 (30.43)**	23 (19.83)	16 (13.91)	0.01

25(OH)D = 25-hydroxyvitamin D; CCr = creatinine clearance rate; RRF = residual renal function; Hs-CRP = high sensitive C-reactive protein; iPTH = intact parathyroid hormone.

^a Boldface type indicates significance between the 3 groups.

^b The number of previous peritonitis episodes for patients who have peritonitis history.

^c Alfacalcidol doses were converted to the calcitriol equivalent by multiplying by 0.75.

* $P < 0.05$ compared to the high tertile group.

** $P < 0.005$ compared to the high tertile group.

*** $P < 0.001$ compared to the high tertile group.

$P < 0.05$ compared to the low tertile group.

19.63 (11.86 – 23.12) nmol/L and the median time between the measurement of pre-peritonitis serum 25(OH)D and the peritonitis episode was 1.43 (0.56 – 2.43) months. As shown in Table 3, there were no significant differences in DSS and dialysate white cell counts on days 0, 3, and 5, or with respect to the distribution of the causative organism between episodes dichotomized by the median pre-peritonitis 25(OH)D level. However, patients in the low pre-peritonitis 25(OH)D group tended to experience subsequent peritonitis during follow-up compared to those in the high group ($p = 0.04$).

DISCUSSION

This study indicated that vitamin D deficiency was prone to be prevalent in diabetic PD patients with long duration on dialysis, accompanied by malnutrition and inflammation. The vitamin D status evaluated via serum 25(OH)D level could predict the occurrence of peritonitis in this prospective PD

cohort, which was independent of comorbidity load, physical performance, active vitamin D supplementation, calcium, phosphate, iPTH, and other recognized confounders. Low pre-peritonitis 25(OH)D was linked to the risk of subsequent peritonitis. This is the first prospective study indicating an association between vitamin D status and peritonitis risk in a PD population. By contrast, 2 previous studies indicating that oral active vitamin D is associated with peritonitis risk in retrospective PD cohorts did not examine serum 25(OH)D values.

Vitamin D has an important role in regulating the immune function, reflected by mediating the maturation, differentiation, and biochemical function of most cells of the immune system (1). Those functions rely on the process of converting the circulating form of 25(OH)D to its active form, 1,25(OH)₂D mediated via 25-hydroxyvitamin D-1α-hydroxylase in cells of the immune system (20). Our data first verified the prognostic value of 25(OH)D deficiency for peritonitis risk in the PD population. There are several mechanisms for the immunologic

potentiating function of vitamin D. First, Rook *et al.* showed that vitamin D could increase the antibacterial activity in human monocyte cell lines (21). Second, the expression of cathelicidin, an antimicrobial peptide with broad spectrum activity, can be upregulated by vitamin D in a wide range of human cells (20). Third, microinflammation is associated with immune dysfunction for patients with chronic kidney disease (10), and vitamin D contributes to maintaining inflammatory response within physiological limits (1).

Figure 2 — Kaplan–Meier analysis of the probability of remaining peritonitis-free according to tertiles of baseline serum 25(OH)D level. Time-to-peritonitis episode was significantly shorter for patients in the low tertile compare to patients in the high tertile ($p=0.003$).

Of note, in our study, the prognostic value of 25(OH)D was independent of oral calcitriol ($1,25[\text{OH}]_2\text{D}$) or alfacalcidol (precursor of $1,25[\text{OH}]_2\text{D}$). This phenomenon may be explained by the fact that the ability of vitamin D to influence human immune responsiveness seems to be highly dependent on 25(OH)D status, the main circulating vitamin D metabolite (5). Calcitriol and alfacalcidol were thought to be exclusively active for calcium homeostasis in patients with chronic kidney disease, as the characteristic short half-life for active vitamin D forms does not exert an immunomodulating action on many extrarenal cells. To the contrary, cholecalciferol supplementation could increase serum 25(OH)D obviously, alter calcitriol-responsive monocyte proteins, and decrease inflammatory cytokines during hemodialysis (22). Therefore, it is recommended that native vitamin D should be combined with active vitamin D supplements in patients with chronic kidney disease (23).

We are aware that patients in the low 25(OH)D tertile group appeared to be “sicker” than those in the middle and high groups. So we added Charlson index and Karnofsky index into the Cox regression model, and baseline 25(OH)D was still an independent predictor of peritonitis. These findings indicated that the link between vitamin D deficiency and susceptibility to infection could not be totally explained by malnutrition and frailty. However, we also recognized that the weakness of patients could not be assessed by Charlson index and Karnofsky index comprehensively. Accordingly, the confounding effect of general frailty is not excluded. Future studies need to be performed to

TABLE 2
Multivariate Cox Regression Models for Predicting Peritonitis

Variables	Model 1		Model 2		Model 3	
	HR (95% CI)	P value [#]	HR (95% CI)	P value [#]	HR (95% CI)	P value [#]
Tertile of serum 25(OH)D						
Low	ref	0.01	ref	0.01	ref	0.01
Middle	0.54 (0.31–0.94)	0.03	0.50 (0.28–0.88)	0.02	0.53 (0.30–0.92)	0.02
High	0.39 (0.20–0.75)	0.01	0.38 (0.19–0.76)	0.01	0.38 (0.20–0.75)	0.01
Age (per year)	1.02 (1.00–1.04)	0.07	1.02 (1.00–1.05)	0.06	1.02 (1.00–1.04)	0.06
Gender (male as reference)	0.64 (0.39–1.07)	0.09	0.68 (0.40–1.15)	0.15	0.61 (0.36–1.03)	0.06
Dialysis duration (per month)	1.00 (0.99–1.01)	0.56	1.00 (0.99–1.01)	0.62	1.00 (0.99–1.01)	0.51
Diabetes	0.75 (0.44–1.26)	0.27	0.72 (0.39–1.33)	0.29	0.72 (0.42–1.25)	0.25
Albumin (1 g/L)	0.97 (0.91–1.03)	0.27	0.98 (0.92–1.04)	0.51	0.96 (0.91–1.03)	0.27
RRF (1 mL/min)	0.91 (0.81–1.01)	0.09	0.92 (0.82–1.03)	0.17	0.90 (0.80–1.01)	0.08
Peritonitis history	1.33 (0.76–2.34)	0.32	1.35 (0.77–2.38)	0.30	1.26 (0.71–2.24)	0.42
Charlson index (1 point)	—	—	1.02 (0.96–1.09)	0.54	—	—
Karnofsky index (1 point)	—	—	1.00 (0.98–1.02)	0.95	—	—
Taking oral active vitamin D	—	—	—	—	0.85 (0.51–1.42)	0.53
Calcium (1 mmol/L)	—	—	—	—	1.32 (0.38–4.57)	0.66
Phosphate (1 mmol/L)	—	—	—	—	0.76 (0.41–1.42)	0.39
iPTH (100 pg/mL)	—	—	—	—	1.07 (0.93–1.23)	0.36

RRF = residual renal function; 25(OH)D = 25-hydroxyvitamin D; iPTH = intact parathyroid hormone; HR = hazard ratio.

Model 1: adjusted for age, gender, dialysis duration, diabetes, albumin, RRF, and history of peritonitis.

Model 2: adjusted for Model 1 plus Charlson index and Karnofsky index.

Model 3: adjusted for Model 1 plus whether or not taking active vitamin D supplements, serum calcium, phosphate, and iPTH.

[#] Boldface type indicates significance.

TABLE 3
DSS, Dialysate White Cell Counts, and Causative Organisms of Peritonitis by Median Pre-Peritonitis 25(OH)D Level

Variables	Pre-peritonitis serum 25(OH)D (nmol/L)		P value
	Low (≤ 19.63) (n=31)	High (>19.63) (n=31)	
DSS	1 (0–2)	1 (1–2)	0.23
Dialysate WBCs (n/mm ³)			
Day 0	1385 (640–3027.5)	1920 (512.5–3010)	0.95
Day 3	54 (13–699)	28 (4.5–350)	0.22
Day 5	24 (3–76.75)	10 (3–595)	0.93
Causative organisms			0.10
Gram-positive (n, %)*	12 (42.86)	8 (25.81)	—
Gram-negative (n, %)*	7 (25)	12 (38.71)	—
Polymicrobial and Fungi (n, %)*	0 (0)	4 (12.9)	—
Culture negative (n, %)*	9 (32.14)	7 (22.58)	—
Having subsequent peritonitis (n, %)	11 (35.48)	4 (12.90)	0.04

DSS = disease severity scores; 25(OH)D = 25-hydroxyvitamin D; WBCs = white blood cells.

* The number and percentage of specific organism to total causative organisms examined.

determine the mechanisms for immune-effect of vitamin D and its association with peritonitis risk in the PD population. More interventions related to vitamin D supplementation are urgently needed to determine their benefits in decreasing the risk for peritonitis and any other systemic infections.

Of note, the median level of serum 25(OH)D in our subjects was remarkably lower than the 50 nmol/L recommended for normal individuals (24); we could not use 50 nmol/L as a cut-off point in our study because only 3 patients had baseline serum 25(OH)D levels higher than this. The serum 25(OH)D levels in this study are also lower than their hemodialysis counterparts as reported previously (25,26), but comparable to PD patients (8). Since the benefits of external vitamin D supplementation in modulating immune function is based on improving vitamin D status, the therapeutic regimen for vitamin D repletion should be explored in terms of improving the vitamin D status of the host—that is, an increase in serum 25(OH)D levels. Unfortunately, vitamin D replacement strategies applied in present clinical trials vary in frequency and dose for treating infection (4). A variable regime for cholecalciferol (10,333 IU weekly – 100,000 IU monthly) has also been confirmed to be effective and safe with good compliance for hemodialysis patients (27–29). We hope that more studies will be performed to explore the association between prescribed vitamin D dose and the 25(OH)D level achieved.

This study has some distinct advantages over previous studies. First, a series of serum samples were collected at baseline and then at 6-month intervals. This provided a unique chance to explore the association between baseline/pre-peritonitis serum vitamin D status and peritonitis risk. Second, oral vitamin D supplements as a critical confounder were recorded prospectively during the study period, thereby avoiding recall bias induced by retrospective data collection. Third, other recognized confounders were also taken into account simultaneously when determining the association between

25(OH)D and peritonitis risk. The relatively large sample size and prospective design are also merits of this study.

However, the present study is not without limitations. For instance, as an observational study, a cause-effect relationship could not be established. Furthermore, the potential mechanisms for the immunomodulating effect of serum vitamin D status on peritonitis risk are not clear. Measurements of innate and acquired immune dysfunction related to vitamin D deficiency would be helpful to address this issue. Also, there is a possibility of selection bias since 30% of prevalent PD patients were not included in this study. Since the peritonitis rate and the spectrum of pathogenic bacteria in the present study are comparable to our previous studies (17,30), subjects of this study can be considered representative. In addition, the study period was relatively short and the frequency of peritonitis was low. If the observation period is lengthened and more outcome events recorded, more statistical power to solidify our main findings would be realized.

In conclusion, vitamin D deficiency evaluated by serum 25(OH)D levels rather than active vitamin D supplementation at the present doses contributes to a higher risk of peritonitis. Clinicians and researchers should explore the potential immunological actions of vitamin D and its contribution to peritonitis in the PD population. More studies are needed to determine the effect of oral vitamin D supplements on decreasing peritonitis rates and attaining target vitamin D repletion.

DISCLOSURES

The authors have no financial conflicts of interest to declare.

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REFERENCES

1. Sterling KA, Eftekhari P, Girndt M, Kimmel PL, Raj DS. The immunoregulatory function of vitamin D: Implications in chronic kidney disease. *Nat Rev Nephrol* 2012; 8:403–12.
2. Hewison M. Antibacterial effects of vitamin D. *Nat Rev Endocrinol* 2011; 7:337–45.
3. Ginde AA, Mansbach JM, Camargo CA, Jr. Association between serum 25-hydroxyvitamin D level and upper respiratory tract infection in the Third National Health and Nutrition Examination Survey. *Arch Intern Med* 2009; 169:384–90.
4. Yamshchikov AV, Desai NS, Blumberg HM, Ziegler TR, Tangpricha V. Vitamin D for treatment and prevention of infectious diseases: a systematic review of randomized controlled trials. *Endocr Pract* 2009; 15:438–49.
5. Lang PO, Samaras N, Samaras D, Aspinall R. How important is vitamin D in preventing infections? *Osteoporos Int* 2013; 24:1537–53.
6. Drechsler C, Verduijn M, Pilz S, Dekker FW, Krediet RT, Ritz E, et al. Vitamin D status and clinical outcomes in incident dialysis patients: Results from the NECOSAD study. *Nephrol Dial Transplant* 2011; 26:1024–32.
7. Wang AY, Lam CW, Sanderson JE, Wang M, Chan IH, Lui SF, et al. Serum 25-hydroxyvitamin D status and cardiovascular outcomes in chronic peritoneal dialysis patients: a 3-y prospective cohort study. *Am J Clin Nutr* 2008; 87:1631–8.
8. Taskapan H, Ersoy FF, Passadakos PS, Tam P, Memmos DE, Katopodis KP, et al. Severe vitamin D deficiency in chronic renal failure patients on peritoneal dialysis. *Clin Nephrol* 2006; 66:247–55.
9. Shah N, Bernardini J, Piraino B. Prevalence and correction of 25(OH) vitamin D deficiency in peritoneal dialysis patients. *Perit Dial Int* 2005; 25:362–6.
10. Kato S, Chmielewski M, Honda H, Pecoits-Filho R, Matsuo S, Yuzawa Y, et al. Aspects of immune dysfunction in end-stage renal disease. *Clin J Am Soc Nephrol* 2008; 3:1526–33.
11. Rudnicki M, Kerschbaum J, Hausdorfer J, Mayer G, König P. Risk factors for peritoneal dialysis-associated peritonitis: the role of oral active vitamin D. *Perit Dial Int* 2010; 30:541–8.
12. Kerschbaum J, Vychytil A, Lhotka K, Prischl FC, Wiesholzer M, Machhold-Fabrizii V, et al. Treatment with oral active vitamin D is associated with decreased risk of peritonitis and improved survival in patients on peritoneal dialysis. *PLoS One* 2013; 8:e67836.
13. Granado-Lorencio F, Blanco-Navarro I, Perez-Sacristan B, Millan I, Donoso-Navarro E, Silvestre-Mardomingo RA. Determinants of fat-soluble vitamin status in patients aged 65 years and over. *Eur J Clin Nutr* 2013; 67:1325–7.
14. Mor V, Liliberte L, Morris JN, Wiemann M. The Karnofsky performance status scale. An examination of its reliability and validity in a research setting. *Cancer* 1984; 53:2002–7.
15. Bergstrom J, Heimbürger O, Lindholm B. Calculation of the protein equivalent of total nitrogen appearance from urea appearance. Which formulas should be used? *Perit Dial Int* 1998; 18:467–73.
16. Matsumoto T, Ito M, Hayashi Y, Hirota T, Tanigawara Y, Sone T, et al. A new active vitamin D3 analog, eldecalcitol, prevents the risk of osteoporotic fractures—a randomized, active comparator, double-blind study. *Bone* 2011; 49:605–12.
17. Dong J, Luo S, Xu R, Chen Y, Xu Y. Clinical characteristics and outcomes of “silent” and “non-silent” peritonitis in patients on peritoneal dialysis. *Perit Dial Int* 2013; 33:28–37.
18. Schaefer F, Klaus G, Müller-Wiefel DE, Mehls O. Intermittent versus continuous intraperitoneal glycopeptide/ceftazidime treatment in children with peritoneal dialysis-associated peritonitis. The Mid-European Pediatric Peritoneal Dialysis Study Group (MEPPS). *J Am Soc Nephrol* 1999; 10:136–45.
19. Li PK, Szeto CC, Piraino B, Bernardini J, Figueiredo AE, Gupta A, et al. Peritoneal dialysis-related infections recommendations: 2010 update. *Perit Dial Int* 2010; 30:393–423.
20. Bikle DD, Pillai S, Gee E, Hincenbergs M. Tumor necrosis factor- α regulation of 1,25-dihydroxyvitamin D production by human keratinocytes. *Endocrinology* 1991; 129:33–8.
21. Rook GA, Steele J, Fraher L, Barker S, Karmali R, O’Riordan J, et al. Vitamin D3, gamma interferon, and control of proliferation of mycobacterium tuberculosis by human monocytes. *Immunology* 1986; 57:159–63.
22. Stubbs JR, Idiculla A, Slusser J, Menard R, Quarles LD. Cholecalciferol supplementation alters calcitriol-responsive monocyte proteins and decreases inflammatory cytokines in ESRD. *J Am Soc Nephrol* 2010; 21:353–61.
23. Jones G. Why dialysis patients need combination therapy with both cholecalciferol and a calcitriol analogs. *Semin Dial* 2010; 23:239–43.
24. Mithal A, Wahl DA, Bonjour JP, Burckhardt P, Dawson-Hughes B, Eisman JA, et al. Global vitamin D status and determinants of hypovitaminosis D. *Osteoporos Int* 2009; 20:1807–20.
25. Wolf M, Shah A, Gutierrez O, Ankers E, Monroy M, Tamez H, et al. Vitamin D levels and early mortality among incident hemodialysis patients. *Kidney Int* 2007; 72:1004–13.
26. Gonzalez EA, Sachdeva A, Oliver DA, Martin KJ. Vitamin D insufficiency and deficiency in chronic kidney disease. A single center observational study. *Am J Nephrol* 2004; 24:503–10.
27. Armas LA, Andukuri R, Barger-Lux J, Heaney RP, Lund R. 25-hydroxyvitamin D response to cholecalciferol supplementation in hemodialysis. *Clin J Am Soc Nephrol* 2012; 7:1428–34.
28. Jean G, Souberbielle JC, Chazot C. Monthly cholecalciferol administration in haemodialysis patients: A simple and efficient strategy for vitamin D supplementation. *Nephrol Dial Transplant* 2009; 24:3799–805.
29. Tokmak F, Quack I, Schieren G, Sellin L, Rattensperger D, Holland-Letz T, et al. High-dose cholecalciferol to correct vitamin D deficiency in haemodialysis patients. *Nephrol Dial Transplant* 2008; 23:4016–20.
30. Yang Z, Xu R, Zhuo M, Dong J. Advanced nursing experience is beneficial for lowering the peritonitis rate in patients on peritoneal dialysis. *Perit Dial Int* 2012; 32:60–6.