RESEARCH

Low serum 1,25(OH)₂D₃ in end-stage renal disease: is reduced 1 α -hydroxylase the only problem?

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Abstract

Low serum 1,25-dihydroxyvitamin D (1,25(OH)₂D) in end-stage renal disease (ESRD) is considered a consequence of elevated fibroblast growth factor 23 (FGF23) and concomitant reduced activity of renal 1 α -hydroxylase (*CYP27B1*). Current ESRD treatment strategies to increase serum calcium and suppress secondary hyperparathyroidism involve supplementation with vitamin D analogues that circumvent 1 α -hydroxylase. This overlooks the potential importance of 25-hydroxyvitamin D (25(OH)D) deficiency as a contributor to low serum 1,25(OH)₂D. We investigated the effects of vitamin D (cholecalciferol) supplementation (40,000 IU for 12 weeks and maintenance dose of 20,000 IU fortnightly), on multiple serum vitamin D metabolites (25(OH)D, 1,25(OH)₂D₃ and 24,25(OH)₂D₃) in 55 haemodialysis patients. Baseline and 12 month data were compared using related-samples Wilcoxon signed rank test. All patients remained on active vitamin D analogues as part of routine ESRD care. 1,25(OH)₂D₃ levels were low at baseline (normal range: 60–120 pmol/L). Cholecalciferol supplementation normalised both serum 25(OH)D and 1,25(OH)₂D₃. Median serum 25(OH)D increased from 35.1 nmol/L (IQR: 23.0–47.5 nmol/L) to 119.9 nmol/L (IQR: 99.5–143.3 nmol/L) ($P < 0.001$). Median serum 1,25(OH)₂D₃ and 24,25(OH)₂D₃ increased from 48.3 pmol/L (IQR: 35.9–57.9 pmol/L) and 3.8 nmol/L (IQR: 2.3–6.0 nmol/L) to 96.2 pmol/L (IQR: 77.1–130.6 pmol/L) and 12.3 nmol/L (IQR: 9–16.4 nmol/L), respectively ($P < 0.001$). A non-significant reduction in daily active vitamin D analogue dose occurred, 0.94 μ mcg at baseline to 0.77 μ mcg at 12 months ($P = 0.73$). The ability to synthesise 1,25(OH)₂D₃ in ESRD is maintained but is substrate dependent, and serum 25(OH)D was a limiting factor at baseline. Therefore, 1,25(OH)₂D₃ deficiency in ESRD is partly a consequence of 25(OH)D deficiency, rather than solely due to reduced 1 α -hydroxylase activity as suggested by current treatment strategies.

Key Words

- ▶ vitamin D
- ▶ mineral metabolism
- ▶ haemodialysis
- ▶ secondary hyperparathyroidism

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Introduction

The kidney is the major site for synthesis of the active form of vitamin D, 1,25-dihydroxyvitamin D (1,25(OH)₂D). The enzyme that synthesises 1,25(OH)₂D from precursor 25-hydroxyvitamin D (25(OH)D), 1 α -hydroxylase

(*CYP27B1*), is expressed primarily in the renal proximal tubule (1, 2), and is positively and negatively regulated by parathyroid hormone (PTH) and fibroblast growth factor 23 (FGF23), respectively (3). While the kidney has

a central role in $1,25(\text{OH})_2\text{D}$ production, 1α -hydroxylase activity has been found in several other cell types throughout the body including the parathyroid glands, testes, skin, placenta, decidua and macrophages (4, 5). The precise contribution of these extra-renal sites of 1α -hydroxylase activity to circulating levels of $1,25(\text{OH})_2\text{D}$ remains unclear.

Serum $1,25(\text{OH})_2\text{D}$ concentration decreases as chronic kidney disease (CKD) progresses, as a consequence of reduced renal $1,25(\text{OH})_2\text{D}$ synthesis (6, 7). This, in turn, contributes to the lack of feedback regulation of PTH that can lead to secondary hyperparathyroidism in CKD, notably in end-stage renal disease (ESRD). To address this, patients with ESRD are routinely prescribed either $1,25(\text{OH})_2\text{D}$ (calcitriol), synthetic analogues of $1,25(\text{OH})_2\text{D}$ (e.g. paracalcitol) or vitamin D analogues that do not require the action of 1α -hydroxylase (e.g. alfacalcidol). However, patients with ESRD also have low serum levels of the substrate for 1α -hydroxylase ($25(\text{OH})\text{D}$), with reported prevalence rates of $25(\text{OH})\text{D}$ deficiency of up to 95% (8, 9). The significance of this finding is unclear but low serum $25(\text{OH})\text{D}$ may also contribute to impaired feedback regulation of PTH (10). $25(\text{OH})\text{D}$ is a substrate for both activating and catabolic pathways; thus in addition to low serum levels of $25(\text{OH})\text{D}$ and $1,25(\text{OH})_2\text{D}$, CKD patients may also have dysregulated levels of $24,25$ -dihydroxyvitamin D ($24,25(\text{OH})_2\text{D}$). $24,25(\text{OH})_2\text{D}$ is the most abundant product of vitamin D catabolism and is produced by the enzyme 24-hydroxylase (*CYP24A1*) (11). The relative abundance of catabolic vitamin D metabolites such as $24,25(\text{OH})_2\text{D}$ in patients with CKD has yet to be fully defined, but it is important to recognise that decreased availability of substrate $25(\text{OH})\text{D}$ may be exacerbated by the stimulatory effect of FGF23 on *CYP24A1* (12). With this in mind, recent research in healthy populations suggest that ratios between vitamin D metabolites may provide a more pathophysiologically relevant insight into the metabolism and function of vitamin D (13). An overview of the chemical structures and hydroxylation steps is shown in Fig. 1.

In this study, we hypothesised that ESRD patients undergoing haemodialysis may benefit from supplementation with vitamin D_3 (cholecalciferol) to elevate serum $25(\text{OH})\text{D}$ levels in addition to conventional active vitamin D analogues. Based on insight into pathways involved in vitamin D metabolism, we postulated that patients with ESRD, when given sufficient substrate, retain the capacity to generate a significant rise in serum levels of $1,25(\text{OH})_2\text{D}_3$.

Materials and methods

All aspects of the study received National Health Service ethical approval (reference 14/EE/10 and 14/NS/1012). Patients were included if they were established on haemodialysis for ≥ 1 month (three sessions per week of 3.5–4 h), had no hospital admissions within the past 4 weeks and had no active malignancy. Patients were excluded if they were already taking cholecalciferol or ergocalciferol. All 202 patients having dialysis at University Hospitals Coventry and Warwickshire (UHCW) satellite dialysis centres meeting the inclusion criteria were invited for recruitment; 81 agreed to participate. Cholecalciferol supplementation was given by the nursing staff, during routine dialysis visits, based on serum $25(\text{OH})\text{D}$ levels. Patients received cholecalciferol, 40,000 IU weekly for 12 weeks if serum $25(\text{OH})\text{D}$ was < 50 nmol/L, 20,000 IU fortnightly if serum $25(\text{OH})\text{D}$ was 75–150 nmol/L, and cholecalciferol was stopped if serum $25(\text{OH})\text{D}$ was ≥ 150 nmol/L. Serum $25(\text{OH})\text{D}$ was measured 3 monthly. The aim was to maintain serum $25(\text{OH})\text{D}$ levels between 75 and 150 nmol/L; the lower target of 75 nmol/L is based on the level defined as sufficient by current Endocrine Society guidelines (14). Active analogue dose was recorded at baseline and study end. Serum calcium was measured monthly and PTH concentration 3 monthly as part of usual care. Blood samples were taken by renal research nurses at routine dialysis sessions, processed and stored at -80°C , for subsequent measurement of serum $25(\text{OH})\text{D}$, $1,25(\text{OH})_2\text{D}_3$ and $24,25(\text{OH})_2\text{D}_3$ using liquid chromatography-tandem mass spectrometry (LC-MS/MS) at baseline (T0) and after 12 months supplementation (T1) (using previously described, quality controlled methods) (15). All samples (T0 and T1) were processed for the measurement of vitamin D metabolites at study end; clinicians were blinded to serum $1,25(\text{OH})_2\text{D}_3$ levels during the study. Doses of active vitamin D analogues were modified according to calcium and PTH levels as part of usual practice. Vitamin D metabolite ratios (VMRs): $1,25(\text{OH})_2\text{D}_3:25(\text{OH})\text{D}_3$, $25(\text{OH})\text{D}_3:24,25(\text{OH})_2\text{D}_3$ and $1,25(\text{OH})_2\text{D}_3:24,25(\text{OH})_2\text{D}_3$, were calculated. Analysis was carried out using the related-samples Wilcoxon signed rank test and Spearman's rank correlation coefficient to compare data pre- and post-cholecalciferol supplementation.

Results

Complete data for all parameters at T0 and T1 was obtained for 55 of 81 participants. Reasons for

Figure 1

Vitamin D metabolism – the chemical structures and hydroxylation steps. 25-hydroxylase (25-hydroxylase, *CYP2R1*) converts vitamin D to 25(OH)D in the liver. 1α-hydroxylase (1α-hydroxylase, *CYP27B1*) converts 25(OH)D to 1,25(OH)₂D in the kidney. Other tissues contain these enzymes, but the liver is the main source of 25-hydroxylase, and the kidney is considered the main source for 1α-hydroxylase. 1,25(OH)₂D is further metabolised by 24-hydroxylase (24-hydroxylase, *CYP24A1*) to 1,24,25(OH)₃D. 24-hydroxylase also acts on 25(OH)D to produce 24,25(OH)₂D. The production of these metabolites is considered degradation; expression of 24-hydroxylase and 1α-hydroxylase is reciprocal in order to control 1,25(OH)₂D levels.

incomplete data were death ($n = 15$), transplantation ($n = 4$), change in modality to peritoneal dialysis ($n = 1$) and haemolysed samples ($n = 6$). Participants had a median age of 69 (range: 27–76), 50% were male and 93% White. The study population was representative of the local haemodialysis population for all characteristics except for ethnicity; 74% of UHCW haemodialysis patients were White.

Serum levels of all three vitamin D₃ metabolites (25(OH)D₃, 1,25(OH)₂D₃ and 24,25(OH)₂D₃) showed a

significant increase from baseline to 12 months following supplementation with cholecalciferol, $P < 0.001$ (Fig. 2 and Table 1). This elevation of individual metabolites was associated with a significant reduction in the ratio of 1,25(OH)₂D₃:25(OH)D₃ and 1,25(OH)₂D₃:24,25(OH)₂D₃, $P = 0.01$ and $P = 0.05$, respectively (Fig. 2 and Table 1). There was no significant change in the ratio of 25(OH)D₃:24,25(OH)₂D₃, $P = 0.70$. A significant increase in serum calcium occurred, $P = 0.001$. There was no change in serum PTH (Table 1).

Figure 2

Serum 25(OH)D₃, 24,25(OH)₂D₃ and 1,25(OH)₂D₃, and the metabolite ratios 25(OH)D₃:24,25(OH)₂D₃, 1,25(OH)₂D₃:24,25(OH)₂D₃ and 1,25(OH)₂D₃:25(OH)D₃ at baseline and 12 months. Serum 25(OH)D₃, 24,25(OH)₂D₃ and 1,25(OH)₂D₃ were measured using LC-MS/MS and the ratios 25(OH)D₃:24,25(OH)₂D₃, 1,25(OH)₂D₃:24,25(OH)₂D₃ and 1,25(OH)₂D₃:25(OH)D₃ calculated, in patients at baseline, pre-cholecalciferol supplementation (T0), and again at 12 months (T1). (A) Serum 25(OH)D₃ increased: 35.1 nmol/L (23.0–47.5) vs 130.0 nmol/L (99.5–143.3), $P < 0.001$. (B) Serum 24,25(OH)₂D₃ increased: 3.8 nmol/L (2.3–6.0) vs 12.3 nmol (9.0–16.4), $P < 0.001$. (C) No significant change in 25(OH)D₃:24,25(OH)₂D₃: 9.1 (7.0–12.4) vs 10.3 (8.0–12.9) respectively, $P = 0.70$. (D) Serum 1,25(OH)₂D₃ increased: 48.3 pmol/L (35.9–57.9) vs 96.2 pmol/L (77.1–130.6), $P < 0.001$. (E) 1,25(OH)₂D₃:24,25(OH)₂D₃ reduced: 10.4 (5.8–19.4) vs 7.9 (5.6–11.9) $P = 0.05$. (F) 1,25(OH)₂D₃:25(OH)D₃ also reduced: 1.2 (0.8–2.1) vs 0.9 (0.6–1.1), $P = 0.01$. Wilcoxon signed rank test. Data represent median (IQR), $n = 55$.

There was no correlation between serum 25(OH)D₃ and 1,25(OH)₂D₃ at T0 or T1, $\rho = 0.165$, $P = 0.23$, and $\rho = 0.180$, $P = 0.19$, respectively. There was also no correlation between serum calcium and serum 25(OH)D₃ at T0 or T1, $\rho = 0.092$, $P = 0.10$, and $\rho = 0.106$, $P = 0.11$. Serum 1,25(OH)₂D₃ did not correlate with calcium at baseline but there was a weak, significant correlation at study end (T1), $\rho = 0.094$, $P = 0.50$ and $\rho = 0.288$, $P = 0.033$, respectively. Mean prescribed daily active

vitamin D analogue dose reduced from 0.94 at T0 to 0.77 μg at T1 ($P = 0.73$). Forty-four of 55 patients (80%) were prescribed an active vitamin D analogue at baseline and 41 of 55 (75%) were prescribed an active vitamin D analogue at 12 months. The prescribed dose of active vitamin D analogue reduced during study follow-up in 14 patients, did not change in 25 patients, and increased (or was commenced) in 7 patients. 1-Alfacalcidol accounted 91% of active analogue prescriptions (Table 2).

Table 1 Calcium, parathyroid hormone, vitamin D metabolites and metabolite ratios at baseline and 12 months.

Serum level/vitamin D metabolite ratio	Baseline (T0) median (IQR)	12 months (T1) median (IQR)	Wilcoxon signed-rank test (P)
Corrected calcium (mmol/L)	2.35 (2.23–2.44)	2.39 (2.32–2.52)	0.001
Parathyroid hormone (pmol/L)	27.0 (16.0–43.5)	29.0 (14.0–42.5)	0.81
25(OH)D ₃ (nmol/L)	35.1 (23.0–47.5)	119.9 (99.5–143.3)	<0.001
1,25(OH) ₂ D ₃ (pmol/L)	48.3 (35.9–57.9)	96.2 (77.1–130.6)	<0.001
24,25(OH) ₂ D ₃ (nmol/L)	3.8 (2.3–6.0)	12.3 (9–16.4)	<0.001
1,25(OH) ₂ D ₃ :25(OH)D ₃	1.2 (0.8–2.1)	0.9 (0.6–1.1)	0.01
25(OH)D ₃ :24,25(OH) ₂ D ₃	9.1 (7.0–12.4)	10.3 (8–12.9)	0.70
1,25(OH) ₂ D ₃ :24,25(OH) ₂ D ₃	10.4 (5.8–19.4)	7.9 (5.6–11.9)	0.05

Serum corrected calcium, parathyroid hormone, 25(OH)D₃, 1,25(OH)₂D₃ and 24,25(OH)₂D₃ were measured at baseline, pre-cholecalciferol supplementation (T0) and again at 12 months (T1), $n = 55$. A significant increase was seen from T0 to T1 in calcium and all three vitamin D metabolites. Median calcium remained within target range. There was no change in parathyroid hormone levels. A significant reduction in 1,25(OH)₂D₃:25(OH)D₃ and 1,25(OH)₂D₃:24,25(OH)₂D₃ occurred but no significant change was seen from baseline to 12 months in 25(OH)D₃:24,25(OH)₂D₃.

Table 2 Active analogue type and dose at baseline and 12 months.

Active analogue	Baseline (μmcg) mean $\pm$ s.d. (n)	12 month (μmcg) mean $\pm$ s.d. (n)	Wilcoxon signed-rank test (P)
1-Alfacalcidol	0.76 $\pm$ 1.30 (n = 40)	0.58 $\pm$ 0.95 (n = 37)	0.103
Paricalcitol	8.00 (n = 1)	8.00 (n = 1)	N/A
Calcitriol	0.92 $\pm$ 0.95 (n = 3)	0.79 $\pm$ 1.05 (n = 3)	0.180
Total combined	0.94 $\pm$ 1.64 (n = 44)	0.77 $\pm$ 1.43 (n = 41)	0.73

Forty-four of 55 (80%) subjects were prescribed an active analogue at the start of the study. 1-Alfacalcidol accounted for 91% of active analogue prescriptions (40 of 44), three patients were prescribed calcitriol and only one patient was prescribed paricalcitol. There was no significant change in active analogue use during the 12-month study period, $n = 55$.

Discussion

This is the first study to date, to assess the impact of vitamin D supplementation on the serum vitamin D multi-metabolite profile and vitamin D metabolite ratios in ESRD. One of the pathophysiological consequences of CKD is the reduced renal capacity to synthesise $1,25(\text{OH})_2\text{D}$, reportedly due to reduced expression of renal *CYP27B* and concomitant reduction of 1α -hydroxylase activity in the face of elevated levels of FGF23 (6, 12). In the current study, serum $1,25(\text{OH})_2\text{D}_3$ concentrations were low at baseline (median 48.3 pmol/L, IQR: 35.9–57.9, normal reference range 60–108 pmol/L) (16) despite 80% of patients receiving vitamin D analogues to promote $1,25(\text{OH})_2\text{D}$ activity in the context of elevated PTH levels. This suggests that vitamin D analogues may be utilised immediately rather than being stored in a serum $1,25(\text{OH})_2\text{D}$ pool. A possible explanation for this, yet to be demonstrated in the literature, is that a sudden increase in $1,25(\text{OH})_2\text{D}_3$ following active analogue administration is met with a catabolic response (by increased 24-hydroxylase activity) in a bid to minimise the risk of hypercalcaemia. This may, in turn, cause $1,25(\text{OH})_2\text{D}_3$ to be metabolised to its less-active 24-hydroxylated catabolite $1,24,25(\text{OH})_3\text{D}_3$. This metabolite was not measured in the current study but levels of $24,25(\text{OH})_2\text{D}_3$ are frequently used as a general marker of 24-hydroxylase activity. In the current study, the circulating levels of $24,25(\text{OH})_2\text{D}_3$ (3.8 nmol/L, 2.3–6.0 nmol/L) were similar to previously reported serum values for healthy individuals (17). However, this level increased three-fold following vitamin D supplementation highlighting further capacity for 24-hydroxylation of vitamin D metabolites that may rapidly counteract elevation of $1,25(\text{OH})_2\text{D}_3$ levels following administration of active analogues.

A lack of consensus on optimal serum levels of $25(\text{OH})\text{D}$ has hindered the management of vitamin D deficiency (18, 19). The UK Scientific Advisory Committee on Nutrition (SACN) guidance recommends serum levels of $25(\text{OH})\text{D} \geq 25$ nmol/L, with supplementation advised

for those below this level (20). Most of the participants in the current study would, therefore, not have met the SACN criteria for supplementation at baseline. However, prevention of different chronic disease states may require different serum $25(\text{OH})\text{D}$ concentrations (21). Data from the current study suggest that patients with ESRD may need higher serum $25(\text{OH})\text{D}$ to facilitate optimal 1α -hydroxylase activity; in this study, most patients achieved normal serum $1,25(\text{OH})_2\text{D}_3$ concentration with serum $25(\text{OH})\text{D} \geq 75$ nmol/L (the level defined as sufficient by the Endocrine Society) (14).

Data presented here also indicate that ESRD patients retain a significant capacity for 1α -hydroxylase activity, with synthesis of $1,25(\text{OH})_2\text{D}_3$ occurring in a substrate ($25(\text{OH})\text{D}_3$)-dependent fashion. Previous studies have also described the potential for synthesis of $1,25(\text{OH})_2\text{D}_3$ in ESRD patients. Jean *et al.* demonstrated a rise in serum $1,25(\text{OH})_2\text{D}_3$ in response to cholecalciferol supplementation but not within the normal reference range (45 ± 13 pmol/L at study end) (22). Patients had lower baseline serum $1,25(\text{OH})_2\text{D}_3$ than the current study and active vitamin D analogue use was an exclusion criteria (22). Other studies also demonstrated increased serum $1,25(\text{OH})_2\text{D}_3$ in response to cholecalciferol supplementation, with larger increases seen in patients receiving concomitant alfacalcidol (23). Again, the increased serum $1,25(\text{OH})_2\text{D}_3$ in these patients did not reach normal range but this could have been due to the short intervention period of 8 weeks (23). Whether cholecalciferol as a sole therapy is sufficient remains unknown; concurrent active analogue therapy is shown to be safe (not associated with increased risk of hypercalcaemia) (24, 25, 26), and may be required to maintain target serum $1,25(\text{OH})_2\text{D}_3$.

Cholecalciferol supplementation increases the inactive substrate $25(\text{OH})\text{D}$ rather than directly increasing $1,25(\text{OH})_2\text{D}$. Increase in serum $25(\text{OH})\text{D}$ and $24,25(\text{OH})_2\text{D}_3$ are disproportionate to $1,25(\text{OH})_2\text{D}_3$ increase, demonstrating the known tight regulation of $1,25(\text{OH})_2\text{D}_3$ synthesis and secretion (in response to calcium and parathyroid hormone) (27). Therefore optimising

25(OH)D through cholecalciferol supplementation will not result in unlimited 1,25(OH)₂D and severe hypercalcaemia. An increase in median calcium was seen in this study, but this was not clinically relevant. In addition, the absence of correlation between calcium and 25(OH)D, and 1,25(OH)₂D₃ and 25(OH)D, supports the documented safety of cholecalciferol supplementation in ESRD (28, 29). Other studies have reported no change in serum calcium in response to cholecalciferol supplementation (23, 24, 25, 26, 30).

Metabolite ratios have been shown to be useful markers of altered vitamin D metabolism. For example a high 1,25(OH)₂D:25(OH)D ratio can help diagnose sarcoidosis and a high 25(OH)D:24,25(OH)₂D₃ ratio can be used to diagnose loss of *CYP24A1* (24-hydroxylase) function (31, 32, 33, 34). The metabolite ratio offers insight above that of a single metabolite measurement; a low 24,25(OH)₂D₃ could simply reflect low 25(OH)D whereas a high 25(OH)D:24,25(OH)₂D₃ ratio is a viable marker of altered 24-hydroxylase activity (31). A moderate elevation of 25(OH)D:24,25(OH)₂D due to reduced activity of 24-hydroxylase is seen in CKD and bone disorders (35, 36, 37). Evidence suggests the value of using 25(OH)D:24,25(OH)₂D in assessment and management of both fracture risk and CKD risk (38, 39). Serum 1,25(OH)₂D₃ is not routinely measured clinically (40, 41); yet in ESRD 1,25(OH)₂D₃ deficiency is routinely assumed, and active analogue treatment is routinely prescribed (42). A low serum 1,25(OH)₂D₃ in the presence of a high 1,25(OH)₂D₃:25(OH)D ratio would indicate that 25(OH)D is limiting synthesis and secretion of 1,25(OH)₂D₃, suggesting that treatment could include serum 25(OH)D repletion. Results here support the findings of Tang and colleagues who reported an inverse correlation between 1,25(OH)₂D:24,25(OH)₂D and serum 25(OH)D in healthy young adults (13). Data suggest sufficient serum 25(OH)D provides for maintenance of 1,25(OH)₂D and 24,25(OH)₂D in relative proportion (within the normal range). In contrast, when 25(OH)D is lacking, 1,25(OH)₂D:24,25(OH)₂D increases as the production of serum 1,25(OH)₂D is prioritised over 24,25(OH)₂D. Therefore, it appears that, in the presence of low 25(OH)D, the 24,25(OH)₂D pathway is partially inactivated to conserve 25(OH)D for production of 1,25(OH)₂D. The same study also demonstrated that low 25(OH)D (<50 nmol/L), normal 1,25(OH)₂D and high 1,25(OH)₂D:24,25(OH)₂D was associated with a significantly higher serum PTH (13). It is anticipated that treatment with active analogues, for suppression of PTH, would increase the 1,25(OH)₂D:24,25(OH)₂D and, therefore, may be counterproductive. Whereas

treatment focusing on optimising 25(OH)D would induce 1 α -hydroxylase and 24-hydroxylase, increasing 1,25(OH)₂D and 24,25(OH)₂D in a regulated fashion, in turn reducing the 1,25(OH)₂D:24,25(OH)₂D ratio; as shown here. Measurement of vitamin D metabolites and calculation of the metabolite ratios could, therefore, offer insight into the prevention and management of SHPT in CKD. Serum PTH did not change in this study but this may be due to established secondary hyperparathyroidism in this patient cohort which results in parathyroid hyperplasia reducing sensitivity to calcium and 1,25(OH)₂D through downregulation of vitamin D receptors and calcium sensing receptors (43, 44). Optimisation of serum 25(OH)D earlier in the progression of CKD may delay the onset, and minimise the severity, of secondary hyperparathyroidism (10). The recognition for the need to treat 25(OH)D deficiency in the management of secondary hyperparathyroidism has grown in recent years, yet in ESRD the emphasis has remained on active analogue treatment (42).

Prescribers have historically promoted active vitamin D analogues to circumvent apparent lack of 1 α -hydroxylase activity (and associated 1,25(OH)₂D levels) in ESRD yet results here demonstrate otherwise. By their very nature, active vitamin D analogues promote hypercalcaemia. Cholecalciferol is safer and may provide for management of both 25(OH)D and 1,25(OH)₂D deficiencies. Is it time to approach vitamin D deficiency in CKD differently by supplementing the substrate and, if indicated by suboptimal serum PTH response, adding in the active analogue? For example, analogous to the management of anaemia where it is routine to administer iron loading initially before concluding a lack of erythropoietin. The focus on active vitamin D analogues for the treatment of 1,25(OH)₂D deficiency in ESRD not only overlooks 25(OH)D deficiency, but also vitamin D metabolism as a whole. This risks patients' missing out on potential non-bone, as well as bone related benefits of serum 25(OH)D (45, 46, 47, 48).

This study is limited by the absence of a control group, so a randomised controlled trial is required to: (i) confirm findings; (ii) explore potential clinical benefits of cholecalciferol repletion; (iii) investigate whether concurrent active analogue therapy is required or whether cholecalciferol is effective as a sole treatment (can calcium and parathyroid hormone levels remain stable without the need for an active vitamin analogue in ESRD?). Whilst research is steering towards more comprehensive analysis of metabolites (49, 50), at present the clinical usefulness of the multi-metabolite assay in ESRD requires further research

and measurement of serum 25(OH)D concentration remains the sole marker of vitamin D status (14). The results presented here offer new insight into vitamin D metabolism in ESRD, specifically demonstrating that haemodialysis patients retain the capacity to significantly increase serum 1,25(OH)₂D. Conventional oral vitamin D (cholecalciferol) supplementation may, therefore, provide a cheap and safe strategy for the management of vitamin D homeostasis in ESRD patients.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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Consent

Consent has been obtained from each patient or subject after full explanation of the purpose and nature of all procedures.

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