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Which circulating level of 25-hydroxyvitamin D is appropriate?☆

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Abstract

Moderate Vitamin D deficiency causes secondary hyperparathyroidism and bone loss, leading to osteoporosis and fractures. Controversy exists which circulating level of 25-hydroxyvitamin D (25OH)D is appropriate. The high incidence of hip fractures at northern latitudes suggest a relationship with Vitamin D deficiency. However, international studies show lower serum 25(OH)D levels in southern than in northern Europe. Serum 25(OH)D was not a risk factor for hip fractures in several epidemiological studies. The required serum 25(OH)D is usually established by assessing the point where serum parathyroid hormone (PTH) starts to rise. This point varied in several studies between 30 and 78 nmol/l. However, interlaboratory variation may also influence the apparent required serum 25(OH)D level. Dietary calcium intake influences serum PTH and serum PTH may influence the turnover of Vitamin D metabolites. A low calcium intake causes an increase of serum PTH and serum 1,25(OH)2D thereby decreasing the half life of serum 25(OH)D. While a low calcium intake may aggravate Vitamin D deficiency, a high calcium intake may have a Vitamin D sparing effect. With current knowledge, a global estimate for the appropriate serum 25(OH)D is 50 nmol/l.

Keywords: Vitamin D deficiency; Threshold serum 25(OH)D; Calcium intake; Vitamin D replete states; Classification of Vitamin D status

1. Introduction

Vitamin D deficiency causes rickets in children and osteomalacia in adults. Both diseases are relatively uncommon. Moderate Vitamin D deficiency is more frequent and has been associated with the occurrence of hip fractures in the elderly [1]. Moderate Vitamin D deficiency causes secondary hyperparathyroidism, high bone turnover, and may lead to osteoporosis and fractures [2]. Risk groups for Vitamin D deficiency are the elderly, persons with low sunshine exposure, persons with a dark skin in northern climates and patients with diseases interfering with Vitamin D and calcium absorption. Randomized placebo-controlled trials on the effects of Vitamin D supplementation with fracture as outcome criterion have led to discordant results [2]. The different outcomes of these studies and the varying results of epidemiological studies have led to an ongoing discussion on the required serum concentration of 25-hydroxyVitamin D (25(OH)D) in adults. These differences may also be due to differences in assays for serum 25(OH)D and the influence of calcium intake. These subjects will be discussed in this paper.

2. Is Vitamin D deficiency a risk factor for hip fractures?

The association between the occurrence of hip fractures and Vitamin D deficiency has been known for more than 30 years [1]. Studies in patients with hip fractures showed signs of osteomalacia or high bone turnover in 5–30% of these patients [2,3]. In addition, serum 25(OH)D levels were lower in these patients than in age-matched controls. Most of these studies are cross-sectional or case-control and these studies may not always be appropriate to establish causal relationships.

In Europe, the incidence of hip fractures is highest at the most northern latitudes in the countries Norway, Sweden and Denmark [4]. The incidence is intermediate in western Europe and hip fracture incidence is lowest in southern Europe. This geographical variation would suggest a possible relationship with sunshine exposure and Vitamin D synthesis in the skin. However, international studies on serum 25(OH)D concentration show an inverse gradient with the lowest levels in southern Europe, intermediate levels in western Europe and the highest levels in northern Europe [5,6]. This is probably due to cultural habits like sunbathing and dietary intake of fish and supplements which may be much higher in northern Europe.

A more rigorous prospective epidemiological study collects baseline data and continues fracture follow-up for

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Table 1
Methods to assess the required serum 25(OH)D levels

Substrate-dependent synthesis of 1,25(OH)2D in a Vitamin D deficient state
Correlation between serum 25(OH)D and 1,25(OH)2D
Below threshold serum 25(OH)D level, increase of serum 1,25(OH)2D after Vitamin D supplementation
Negative relationship between serum 25(OH)D and PTH
Below threshold serum 25(OH)D, serum PTH starts to increase
Below threshold serum 25(OH)D, a Vitamin D supplement decreases serum PTH
Positive relationship between serum 25(OH)D and bone mineral density
Effects of Vitamin D supplementation on fracture incidence

several years. The largest of these studies, the study on osteoporotic fractures performed in the USA, did not confirm that low serum 25(OH)D was a risk factor for hip fracture [7]. However, a low serum 1,25(OH)2D ($\leq$57 pmol/l) was associated with an increased risk for hip fracture (RR 2.1).

3. Assessment of the required serum 25(OH)D level

The metabolite 25(OH)D is the main circulating Vitamin D metabolite, but it is not the most active metabolite. Conclusions on the required serum 25(OH)D concentration might be based on homeostatic mechanisms, such as the synthesis of 1,25(OH)2D, the increase of the serum parathyroid hormone (PTH) level in case of Vitamin D deficiency, or the effects of Vitamin D on bone. Several methods to assess the required serum 25(OH)D concentration are summarized in Table 1. These methods are based on correlations found in cross-sectional studies or on dynamic changes of 1,25(OH)2D or PTH in intervention studies with a Vitamin D supplement. The most commonly used way to assess the required serum 25(OH)D is based on the negative relationship between serum 25(OH)D and serum PTH. The inverse seasonal relationship of serum PTH and serum 25(OH)D has been known for many years [8]. Serum 25(OH)D is maximal at the end of summer and serum PTH reaches its maximum at the end of winter. Whereas mild and moderate Vitamin D deficiency only causes small increases in serum PTH still within normal limits, these increases might not be advantageous as they stimulate bone resorption. However, it is not known which increase of serum PTH should be considered physiological and at what stage it becomes pathological. In other words, a compensatory increase of serum PTH of about 10% with the decrease of serum 25(OH)D in winter might be considered either physiological or pathological. Other ways to assess the minimally required level of serum 25(OH)D might be the relationship between serum 25(OH)D and bone mineral density and between serum 25(OH)D and fractures.

4. Evidence from epidemiological and intervention studies

In case of Vitamin D deficiency, the synthesis of 1,25(OH)2D becomes dependent on the availability of the substrate 25(OH)D. In that case, a positive correlation has been observed between serum 25(OH)D and serum 1,25(OH)2D. Positive correlations were found in elderly patients with hip fracture [3] and in institutionalized elderly in a study from Belgium [9]. A threshold serum 25(OH)D level was not established in these studies. Vitamin D supplementation may increase serum 1,25(OH)2D in case of a low serum 25(OH)D. This was shown in a Vitamin D supplementation study in institutionalized elderly in The Netherlands comparing the effects of 400 and 800 IU per day with a control group [10]. An increase of serum 1,25(OH)2D was seen when serum 25(OH)D at baseline was lower than 30 nmol/l. It was concluded from this study that the threshold serum 25(OH)D was 30 nmol/l. Another supplementation study done in nursing home residents in the USA did not show an increase of serum 1,25(OH)2D, but the baseline 25(OH)D was over 40 nmol/l [11]. The negative relationship between serum 25(OH)D and serum PTH has been used more often to establish the appropriate serum 25(OH)D level. This relationship was studied in the Amsterdam Vitamin D study, a double-blind placebo-controlled study on the effect of Vitamin D3 400 IU per day or placebo on the incidence of hip fractures and other peripheral fractures. This study showed at baseline a negative correlation between serum PTH and serum 25(OH)D. However, this relationship only existed when serum 25(OH)D was lower than 25 nmol/l [12]. The Suvimax study performed in France in postmenopausal women also showed a negative relationship between serum PTH and serum 25(OH)D. The deflection point in this study where serum PTH started to increase was at serum 25(OH)D 78 nmol/l [13]. A study done in patients admitted to a general ward of a hospital in the USA showed similar data: serum PTH started to increase when serum 25(OH)D was lower than 75 nmol/l or 30 ng/ml [14]. Vitamin D status was also studied in the MORE Study, a global study on the effects of raloxifene on postmenopausal osteoporosis. The baseline data showed a small increase of serum PTH when serum 25(OH)D was between 25 and 50 nmol/l compared to >50 nmol/l [15]. A larger elevation of serum PTH of about 30% was seen when serum 25(OH)D was lower than 25 nmol/l. Dynamic studies on the decrease of serum PTH following Vitamin D supplementation show similar data. In the MORE Study serum PTH did not decrease when baseline serum 25(OH)D was above 50 nmol/l. Serum PTH decreased 12% when serum 25(OH)D was between 25 and 50 nmol/l and 17% when serum 25(OH)D was below 25 nmol/l [6]. According to this study the appropriate serum 25(OH)D level should be 50 nmol/l or higher.

In the Amsterdam Vitamin D study, a positive relationship between serum 25(OH)D and bone mineral density of the femoral neck was observed [12]. However, this relationship was only significant when serum 25(OH)D was lower than 30 nmol/l. Two Vitamin D supplementation studies demonstrated a decrease of fracture incidence either departing from low baseline serum 25(OH)D in the DECALYOS study in

Table 2
Classification of Vitamin D replete and deficient states

	Serum 25(OH)D (nmol/l)	(ng/ml)	Serum PTH	Bone
Vitamin D replete	>50	>20	No increase	Normal
Mild Vitamin D deficiency	25–50	10–20	<15% increase	Normal or high turnover
Moderate Vitamin D deficiency	12.5–25	5–10	15–30% increase	High turnover
Severe Vitamin D deficiency	<12.5 nmol/l	<5	>30% increase	High turnover or osteomalacia

Lyon [15], or from a higher baseline serum 25(OH)D in a study from Boston [16], but in both studies Vitamin D and calcium supplementation was combined, so that no conclusions on the appropriate serum 25(OH)D can be drawn. The Amsterdam Vitamin D study did not show an effect of Vitamin D supplementation on fracture incidence [17].

5. Comparability of assays for serum 25(OH)D

When comparing the results from large epidemiological and intervention studies, it became clear that the results obtained with different assays for serum 25(OH)D may be very different. An interlaboratory comparison of serum 25(OH)D assays was done between laboratories in Lyon, Boston, Rotterdam and Amsterdam [18]. It appeared from this study that a difference between an older competitive protein binding assay and HPLC analysis differed a factor 2, while radioimmunoassays were in between. When comparing the results in the large Vitamin D supplementation studies in Lyon and Amsterdam it appeared that after correction by applying a correction factor, the patients in Lyon were more Vitamin D deficient than those in Amsterdam [18]. The comparability of assays has important implications for the discussion on the minimally required serum 25(OH)D level.

6. The influence of calcium intake on the required serum 25(OH)D

The dietary calcium intake influences the serum PTH level. An oral calcium supplement of 1000 mg decreases serum PTH within one hour [19]. Calcium supplementation can reduce serum PTH during 24 h [20]. Serum PTH influences the turnover of Vitamin D metabolites. A low calcium intake due to low dairy intake after gastrectomy caused an increase of serum PTH and serum 1,25(OH)2D, and decreased the half life of serum 25(OH)D thus leading to Vitamin D deficiency [21]. Metabolic studies in rats with a low calcium intake showed that the increased serum PTH and serum 1,25(OH)2D were associated with increased metabolic clearance of 25(OH)D [22]. So, the increase of serum PTH (secondary hyperparathyroidism) may aggravate Vitamin D deficiency by increasing the turnover of Vitamin D metabolites. On the other hand, a high calcium intake may have a Vitamin D sparing effect. Intervention studies with Vitamin D and calcium supplementation combined are more effective in decreasing fracture incidence than Vitamin D supplementation alone as was demonstrated by the DECALYOS study in Lyon and the supplementation study in Boston compared with the effects in the Amsterdam Vitamin D study [15–17]. Another supplementation study in Boston with Vitamin D2 15,000 IU per week and calcium 1000–1500 mg per day showed a decrease of serum PTH of 40% when serum 25(OH)D was lower than 40 nmol/l, a decrease of 20% when serum 25(OH)D was between 40 and 50 nmol/l and no change when serum 25(OH)D was higher than 50 nmol/l [23]. This study also suggests that the minimally required serum 25(OH)D is 50 nmol/l.

7. Classification of Vitamin D replete and deficient states

A safe lower reference limit for serum 25(OH)D applicable under most circumstances is 50 nmol/l. A serum 25(OH)D between 25 and 50 nmol/l may indicate mild Vitamin D deficiency, while moderate Vitamin D deficiency is a level lower than 25 nmol/l and severe deficiency is defined as a serum 25(OH)D lower than 12.5 nmol/l. This classification is summarized in Table 2.

The assessment of the appropriate circulating level of 25(OH)D is hampered by the moderate comparability of assays for 25(OH)D. The dietary intake of calcium may influence the minimally required serum 25(OH)D level. Other determinants of serum PTH are renal function, estrogen status and the use of loop diuretics. These may indirectly influence the circulating level of 25(OH)D [2].

8. Conclusion

A serum 25(OH)D level of 50 nmol/l or higher can be considered appropriate. This level may be influenced by dietary calcium intake. The required level may be somewhat lower with a high calcium intake, or higher with a very low calcium intake.

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