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Vitamin D: clinical implications beyond musculoskeletal diseases

Vitamin D: Klinische Bedeutung bei nicht muskuloskelettalen Erkrankungen

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Abstract

Vitamin D has historically been known to play a significant role in the regulation of mineral and bone metabolism. It is, however, increasingly recognized that vitamin D deficiency may also be relevant to various extraskeletal diseases. This may be of clinical significance when considering that the majority of the general population has 25-hydroxyvitamin D (25[OH]D) levels below the target concentration of at least 30 ng/mL (75 nmol/L). In this review, we briefly summarize available data on the association of vitamin D with cancer, cardiovascular, renal, and autoimmune diseases and we discuss further emerging fields of vitamin D research. Finally, we give some practical guidance for vitamin D treatment. In summary, the available evidence is still insufficient for general compelling recommendations for vitamin D treatment of extraskeletal diseases. However, in our opinion the high prevalence of vitamin D deficiency, the gradually accumulating knowledge on multiple health benefits of vitamin D, and the safety of vitamin D supplementation suggest that public health strategies should aim to improve the vitamin D status in the general population.

Keywords: epidemiology; guidelines; recommendations; review; vitamin D.

Zusammenfassung

Vitamin D ist historisch gesehen für seine Rolle in der Regulation des Mineral- und Knochenstoffwechsels bekannt. Es wird jedoch zunehmend erkannt, dass ein Mangel an Vitamin D auch von Relevanz für extraskelettale Erkrankungen ist. Das könnte von klinischer Bedeutung sein, wenn man bedenkt, dass der Großteil der Allgemeinbevölkerung 25-hydroxyvitamin D (25[OH]D)-Konzentrationen unter dem Zielwert von zumindest 30 ng/mL (75 nmol/L) hat. In diesem Übersichtsartikel fassen wir kurz die bekannten Zusammenhänge von Vitamin D mit Krebs, kardiovaskulären und renalen Erkrankungen sowie Autoimmunerkrankungen zusammen und wir diskutieren neu aufkommende Gebiete der Vitamin D-Forschung. Schließlich geben wir auch praktische Ratschläge für die Vitamin D-Behandlung. Zusammenfassend ist die derzeitige Evidenzlage zwar noch unzureichend für generelle und zwingende Empfehlungen zur Vitamin D-Behandlung von extraskelettalen Erkrankungen. Wir sind jedoch davon überzeugt, dass die hohe Prävalenz des Vitamin D-Mangels, die allmählich zunehmenden Erkenntnisse über multiple gesundheitsfördernde Effekte von Vitamin D und die Sicherheit einer Vitamin D-Behandlung daraufhin deuten, Public-Health-Strategien darauf auszurichten, den Vitamin D-Status in der Allgemeinbevölkerung zu verbessern.

Schlüsselwörter: empfehlungen; epidemiologie; richtlinien; übersichtsartikel; vitamin D.

Introduction

Vitamin D deficiency is considered a public health problem, because beyond its classic association with musculoskeletal disorders such as osteoporosis, fractures, and falls, a poor vitamin D status has also been associated with various other chronic diseases [1–4]. This is of particular concern when

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considering that the majority of the general population has an insufficient or deficient vitamin D status [1, 2, 5]. In this brief review, we summarize and discuss the current knowledge and the clinical implications of vitamin D with regard to its associations with extraskeletal diseases. We start with a short overview on the assessment and classification of the vitamin D status and outline the role of vitamin D in cancer, cardiovascular, and renal diseases, as well as in autoimmune disorders and further emerging fields of vitamin D research. Finally, we give some practical advice for vitamin D treatment.

Determination and classification of vitamin D status

Vitamin D is mainly generated by endogenous, sunlight [ultraviolet B (UV-B)] induced vitamin D synthesis in the skin. Vitamin D intake by natural foods usually only plays a minor role as a source of vitamin D [1, 2, 4]. Vitamin D3 (cholecalciferol) is the major form of vitamin D derived from skin synthesis and non-plant sources, and vitamin D2 (ergocalciferol) is produced and derived from plants and yeast. Considering that these two main forms of vitamin D are generally comparable regarding their biological effects and metabolism, we refer to vitamin D without differentiating D2 and D3 in this review.

Vitamin D itself is biologically hardly active and is hydroxylated to 25-hydroxyvitamin D (25[OH]D) in the liver. Then, 25(OH)D is further hydroxylated to 1,25-dihydroxyvitamin D (1,25[OH]2D), which is the most active vitamin D metabolite since it possesses the highest affinity for the almost ubiquitously expressed vitamin D receptor (VDR) [2]. For a long-time, it was believed that the conversion of 25(OH)D to 1,25(OH)2D can only be done by the kidney, but recent studies demonstrated that many extra-renal tissues express the enzyme 1α-hydroxylase and are thus able to produce 1,25(OH)2D. Compared to 1,25(OH)2D, serum levels of 25(OH)D are up to 1000-fold higher in their concentrations, have a much longer half life (3–4 weeks vs. 6–8 h) and serve as a rate limiting precursor for the local tissue production of 1,25(OH)2D. Hence, serum levels of 25(OH)D are measured to determine the vitamin D status.

There is still no general consensus on 25(OH)D cut-offs for the clinical assessment of the vitamin D status. The most widely accepted definition of a normal vitamin D status (vitamin D sufficiency) is based on the consideration that disturbances in mineral metabolism emerge at 25(OH)D levels below 30 ng/mL (75 nmol/L, multiply by 2.496 to convert ng/mL to nmol/L) [1]. These are characterized by reduced intestinal calcium absorption and subsequently low serum calcium levels which, in turn, stimulate the release of parathyroid hormone (PTH) to maintain a normal calcium homeostasis. Vitamin D insufficiency is commonly diagnosed if 25(OH)D serum levels range between 20 and <30 ng/mL, whereas levels below 20 ng/mL indicate vitamin D deficiency. There is, however, an ongoing debate on the threshold for vitamin D deficiency with proposed cut-offs ranging from 10 to 20 ng/mL [6]. Optimal target levels for 25(OH)D seem to range from 30 to 40 ng/mL [3]. A 25(OH)D level of 100 ng/mL can be considered as a safe upper limit of normal, and vitamin D toxicity, which is characterized by hypercalcemia, does not occur until levels of 150 ng/mL [1, 3, 4, 6].

Differences between 25(OH)D assays should be considered when determining vitamin D status, but this issue extends the scope of the present review. It should, however, be acknowledged that previous epidemiologic data on vitamin D status were generally based on the DiaSorin radioimmunoassay [7]. Increasing interest in vitamin D during the last few years has prompted the development of various automated immunoassays which should be critically evaluated in clinical routine, with particular attention to the relatively high inter-laboratory variation of 25(OH)D measurements [4, 7].

The majority of the general population has 25(OH)D levels below the normal range of 30 ng/mL and a significant part is even vitamin D deficient. This has recently been shown by Mithal et al. who presented an overview of the global vitamin D status [5]. This high prevalence of vitamin D deficiency and its related adverse health outcomes have already prompted some national health authorities to initiate food fortification with vitamin D (e.g., in the US or Finland) in order to reduce the burden of vitamin D deficiency.

Vitamin D and cancer

Observations at the beginning of the last century showed that high sunlight exposure is associated with reduced risk of cancer mortality [8, 9]. It has meanwhile been established that UV-B exposure, which induces vitamin D synthesis in the skin, is associated with reduced risk of overall cancer incidence and cancer mortality [10]. Studies in humans have partially, but not consistently, confirmed that higher serum concentrations of 25(OH)D are associated with reduced risk of cancer mortality [11, 12]. There are, in addition, various studies which have almost consistently shown that in cancer patients, low 25(OH)D levels are an independent risk factor for increased mortality [11]. Whereas there are partially inconsistent data for many specific cancer sites, recent systematic reviews and meta-analyses support the notion that vitamin D may protect against colorectal cancer [13]. This is in line with various molecular effects of vitamin D which suggest anti-carcinogenic properties of vitamin D [2]. These include induction of differentiation and cancer cell apoptosis, inhibition of metastasis, proliferation and angiogenesis, anti-inflammatory effects as well as increased sensitivity to radiation therapy and chemotherapy [2]. Interventional trials in this field are sparse and are often limited by the fact that vitamin D doses were too low to produce relevant increases of 25(OH)D. Importantly, one study among 1179 postmenopausal women has already shown that vitamin D (1100 International Units [IU] per day) plus calcium (1500 mg/day) supplementation is associated with a significantly reduced risk of cancer incidence when compared to placebo (relative risk RR: 0.40; 95% CI: 0.20–0.82) [14]. Hence, there are promising data on vitamin D and cancer which have already lead to some

recommendations for vitamin D supplementation (i.e., Canadian Cancer Society) to protect against cancer. Other large organisations such as the International Agency for Research on Cancer (IARC) have concluded that there is still insufficient evidence for the use of vitamin D in the prevention and treatment of cancer and recommended further studies in this field [15]. Interestingly, data on cancer and vitamin D, which may even protect against melanoma, have also gained much interest in the field of dermatology and have lead to initiatives to reconsider the "no sun policy" of dermatologists and related societies towards moderate sunlight exposure [16].

Vitamin D and cardiovascular disease

Vitamin D deficiency has been associated with cardiovascular diseases (CVD) and its risk factors [1–3]. This is supported by the fact that VDR knockout mice suffer from CVD, and in particular from arterial hypertension and myocardial hypertrophy [17, 18]. Almost all classic cardiovascular risk factors are associated with low levels of 25(OH)D, but evidence from interventional trials are still sparse and partially controversial. Strong evidence exists that vitamin D exerts antihypertensive properties, which may be partially mediated by suppression of the renin angiotensin aldosterone system (RAAS) [17–21]. Interestingly, three meta-analyses of randomized controlled trials (RCTs) have shown that vitamin D supplementation reduces systolic blood pressure by 2–6 mm Hg [22]. PTH levels, which increase as a result of vitamin D deficiency, may also exert deleterious effects on heart and vessels. In this context, it is important to note that PTH levels are an independent risk factor for adverse cardiovascular outcomes [23, 24]. In addition, data from experimental and clinical studies suggest that vitamin D exerts direct beneficial effects on the cardiovascular system and may protect against myocardial damage, atherosclerosis, and endothelial dysfunction. In epidemiological studies, it has been largely but not consistently shown that low 25(OH)D levels are an independent risk factor for cardiovascular events and mortality [25–30]. Systematic reviews followed by meta-analyses support the hypothesis that a poor vitamin D status is associated with CVD [31–33]. By contrast, there is only insufficient evidence from RCTs. One major problem is that many studies used combined calcium plus vitamin D supplementation which limits the ability to differentiate the separate effects of vitamin D and calcium [34]. This may be of clinical significance because calcium intake has been partially linked to higher cardiovascular risk. A recent systematic review identified two RCTs with exclusive vitamin D supplementation (without calcium) that reported on CVD events [34]. It turned out that there was a moderate but statistically non-significant reduction in CVD risk (pooled RR: 0.90; 95% CI: 0.77–1.05) in the vitamin D group [34]. Further large studies are currently evaluating the effect of vitamin D on CVD, but these trials will still take some years before they will be finished (e.g., the VITAL study among 20,000 healthy US men and women; see www.vitalstudy.org).

Vitamin D and chronic kidney disease

The prevalence of vitamin D deficiency is extraordinary high in patients suffering from chronic kidney diseases (CKD) [35]. This can be attributed to limited sunlight access, impaired vitamin D synthesis in the skin, and disturbed vitamin D metabolism including increased loss of vitamin D metabolites via the urine [35]. Beyond low levels of 25(OH)D, there is also a significant decline of circulating 1,25(OH)2D levels in renal failure, because the kidney is the major site for production of circulating (serum) levels of 1,25(OH)2D [35]. Interestingly, various experimental and clinical studies indicate that vitamin D may protect against a decline of renal function and may reduce glomerular damage and proteinuria [35]. Moreover, it has been documented that low 25(OH)D, as well as low 1,25(OH)2D, are associated with an increased risk of CVD and mortality in CKD patients [36–39]. Considering this, the 2009 Kidney Disease: Improving Global Outcomes (KDIGO) clinical practice guidelines include a recommendation for measuring 25(OH)D levels and treating patients with reduced 25(OH)D levels with vitamin D supplementation [40]. Treatment with 1,25(OH)2D or its analogues (e.g., paricalcitol) is also recommended for patients with advanced CKD and uncontrolled secondary hyperparathyroidism [40]. According to large epidemiological studies, this latter therapy has been associated with an improved overall survival [41].

Vitamin D and autoimmune disease

Vitamin D deficiency, as well as low UV-B radiation, have both been linked to an increased incidence of autoimmune diseases, in particular to multiple sclerosis and type 1 diabetes mellitus [42]. There exist data from small interventional studies and from retrospective evaluations of vitamin D intake (i.e., in children) which indicate that vitamin D supplementation may indeed protect against these autoimmune diseases [42, 43]. Autoimmunity in general is a very complex phenomenon, but mounting evidence suggests that vitamin D supplementation may increase regulatory T cells (Tregs) [44]. These Tregs suppress autoimmunological processes and vitamin D may therefore be a promising agent for the prevention and treatment of autoimmune diseases like type 1 diabetes mellitus.

Further emerging fields of vitamin D

Vitamin D deficiency has been identified as an independent risk factor for strokes [45, 46]. Considering that stroke patients are prone to osteoporosis, fractures and falls, it is important to maintain a sufficient vitamin D status in these patients because vitamin D has been shown to significantly reduce the incidence of these musculoskeletal disorders [46]. Furthermore, mood disorders and depression, epilepsy as well as cognitive function may be modulated by vitamin D, but

further data from RCTs are warranted before drawing final conclusions [46, 47].

Long before the discovery of vitamin D by McCollum in 1922, it was known that patients with tuberculosis have a better recovery if they are exposed to sunlight [48, 49]. Meanwhile, it is known that vitamin D exerts various anti-infectious properties [49]. Regarding tuberculosis it has been shown that VDR activation induces the synthesis of the antimicrobial peptide cathelicidin and there is convincing evidence that vitamin D may be useful for the treatment and probably also for the prevention of tuberculosis [49, 50]. In addition, there are data on vitamin D effects on various other infectious diseases and it has been shown in a RCT that vitamin D supplementation was associated with a significantly reduced incidence of influenza A infections in Japanese schoolchildren [49, 51].

Apart from this, vitamin D may also play a role in human reproduction [2]. In this context, some studies suggest that a sufficient vitamin D status may be important for spermatogenesis [52]. Interestingly, a significant correlation between 25(OH)D and testosterone levels was observed in older men, and a small RCT has shown an increase in testosterone levels in obese men who were supplemented with vitamin D [53, 54]. Vitamin D may also be important for fertility in females, because vitamin D deficiency is particularly prevalent in women with polycystic ovary syndrome (PCOS) [55, 56]. Importantly, there is also mounting evidence that a poor vitamin D status is related to pregnancy complications including preterm delivery [56]. Towards this, it is important to underline that vitamin D requirements seem to increase during pregnancy and lactation [56].

Guidance for vitamin D treatment

Vitamin D treatment can be either done by oral supplementation of vitamin D or by increasing sunlight (UV-B) exposure. It is hardly possible to significantly raise the vitamin D status by increasing the intake of natural vitamin D containing foods (e.g., fish). In general, it can be expected that a daily intake of 1000 IU vitamin D will increase 25(OH)D levels by 10 ng/mL (6–10 ng/mL) but individual variations (e.g., higher vitamin D requirements in obese individuals) should be considered [1, 3]. Re-testing of 25(OH)D levels may therefore be done, also to ensure a better compliance, but this should not be performed earlier than 3 months after starting vitamin D therapy in order to allow for a steady state to be reached [3]. Interestingly, it has been shown that daily, weekly or monthly doses of vitamin D produce similar increases in 25(OH)D levels so that dosing intervals can be adapted to individual choices [3, 57]. Some data indicate that for the improvement of vitamin D status, slightly higher supplementation doses are required for vitamin D2 compared to vitamin D3 supplementation. It should also be considered that there exists a significant seasonal variation of 25(OH)D with, in the UK, 50% higher levels in summer compared to winter [58]. Vitamin D therapy is very safe and there are no known adverse effects at doses up to 10,000 IU/day, which corresponds to the maximal increase of 25(OH)D produced by endogenous vitamin D synthesis in response to sunlight (UV-B) exposure [59]. Towards this, the recent 2011 report from the Institute Of Medicine (IOM) on dietary reference intakes for calcium and vitamin D, suggested a safe tolerable upper intake level (UL) of 4000 IU vitamin D per day [60]. In that report, it was stated that there is sufficient evidence on beneficial vitamin D effects on "bone health" with still insufficient evidence for other health outcomes. In detail the IOM report recommends a 25(OH)D level of at least 20 ng/mL and a daily vitamin D intake of 600 IU for individuals until the age of 70 years and 800 IU for older adults as the recommended daily allowance (RDA, covering requirements of ≥97.5% of the population). This should be viewed in light of the fact that in general populations, the current daily vitamin D intake is usually not higher than 100–200 IU, which may suggest that public health authorities should aim to improve vitamin D intake. In some countries such as Finland or in North America, there exists food fortification with vitamin D, but this could not prevent hypovitaminosis D in large parts of the general population [61, 62]. In this context, it is also important to underline that a meta-analysis of RCTs among frail elderly patients has already shown that vitamin D supplementation is associated with a significant 7% reduction in total mortality when compared to placebo [63].

Conclusions

Beyond its classic role in bone and mineral metabolism, vitamin D may also be relevant for various other chronic diseases. Accumulating evidence, including RCTs, suggests that vitamin D supplementation may be useful for the prevention and treatment of adverse health outcomes. However, currently available evidence is for most extraskeletal diseases still insufficient to raise general recommendations for vitamin D treatment. On the other hand, it should be acknowledged that the majority of the general population has an insufficient or deficient vitamin D status and that vitamin D has been proven to significantly prevent common musculoskeletal diseases. Furthermore, it should be considered that vitamin D supplementation is safe, simple and affordable and may improve survival. We therefore believe that public health strategies should aim to improve the vitamin D status in the general population.

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References

1. Holick MF. Vitamin D deficiency. N Engl J Med 2007;357:266–81.
2. Bouillon R, Carmeliet G, Verlinden L, van Etten E, Verstuyf A, Luderer HF, et al. Vitamin D and human health: lessons from vitamin D receptor null mice. Endocr Rev 2008;29:726–76.

3. Souberbielle JC, Body JJ, Lappe JM, Plebani M, Shoenfeld Y, Wang TJ, et al. Vitamin D and musculoskeletal health, cardiovascular disease, autoimmunity and cancer: recommendations for clinical practice. Autoimmun Rev 2010;9:709–15.
4. Pilz S, Dobnig H, Fahrleitner-Pammer A, Polt G, März W. Vitamin D-Mangel: Ein globales Gesundheitsproblem. J Lab Med 2008;32:200–8.
5. 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.
6. Rosen CJ. Clinical practice. Vitamin D insufficiency. N Engl J Med 2011;364:248–54.
7. Carter GD. Accuracy of 25-hydroxyvitamin D assays: confronting the issues. Curr Drug Targets 2011;12:19–28.
8. Apperly FL. The relation of solar radiation to cancer mortality in North America. Cancer Res 1941;1:191–5.
9. Garland CF, Garland FC. Do sunlight and vitamin D reduce the likelihood of colon cancer? Int J Epidemiol 1980;9:227–31.
10. Grant WB, Mohr SB. Ecological studies of ultraviolet B, vitamin D and cancer since 2000. Ann Epidemiol 2009;19:446–54.
11. Pilz S, Tomaschitz A, Obermayer-Pietsch B, Dobnig H, Pieber TR. Epidemiology of vitamin D insufficiency and cancer mortality. Anticancer Res 2009;29:3699–704.
12. Pilz S, Dobnig H, Winklhofer-Roob B, Riedmüller G, Fischer JE, Seelhorst U, et al. Low serum levels of 25-hydroxyvitamin D predict fatal cancer in patients referred to coronary angiography. Cancer Epidemiol Biomarkers Prev 2008;17:1228–33.
13. Touvier M, Chan DS, Lau RN, Aune D, Vieira R, Greenwood DC, et al. Meta-analyses of vitamin D intake, 25-hydroxyvitamin D status, vitamin D receptor polymorphisms and colorectal cancer risk. Cancer Epidemiol Biomarkers Prev 2011;20: 1003–16.
14. Lappe JM, Travers-Gustafson D, Davies KM, Recker RR, Heaney RP. Vitamin D and calcium supplementation reduces cancer risk: results of a randomized trial. Am J Clin Nutr 2007;85: 1586–91.
15. Grant WB. A critical review of vitamin D and cancer: a report of the IARC Working Group. Dermatoendocrinol 2009;1:25–33.
16. Reichrath J, Nürnberg B. Cutaneous vitamin D synthesis versus skin cancer development: The Janus faces of solar UV-radiation. Dermatoendocrinol 2009;1:253–61.
17. Pilz S, Tomaschitz A, Ritz E, Pieber TR. Vitamin D status and arterial hypertension: a systematic review. Nat Rev Cardiol 2009;6:621–30.
18. Pilz S, Tomaschitz A, Drechsler C, Dekker JM, März W. Vitamin D deficiency and myocardial diseases. Mol Nutr Food Res 2010; 54:1103–13.
19. Tomaschitz A, Pilz S, Ritz E, Grammer T, Drechsler C, Boehm BO, et al. Independent association between 1,25-dihydroxyvitamin D, 25-hydroxyvitamin D and the renin angiotensin system: the Ludwigshafen Risk and Cardiovascular Health (LURIC) study. Clin Chim Acta 2010;411:1354–60.
20. Pilz S, Tomaschitz A, März W. Diagnostic procedures for primary aldosteronism. J Lab Med 2009;33:202–9.
21. Tomaschitz A, Pilz S, Ritz E, Obermayer-Pietsch B, Pieber TR. Aldosterone and arterial hypertension. Nat Rev Endocrinol 2010; 6:83–93.
22. Pilz S, Tomaschitz A. Role of vitamin D in arterial hypertension. Expert Rev Cardiovasc Ther 2010;8:1599–608.
23. Pilz S, Tomaschitz A, Drechsler C, Ritz E, Boehm BO, Grammer TB, et al. Parathyroid hormone level is associated with mortality and cardiovascular events in patients undergoing coronary angiography. Eur Heart J 2010;31:1591–8.
24. Hagström E, Hellman P, Larsson TE, Ingelsson E, Berglund L, Sundström J, et al. Plasma parathyroid hormone and the risk of cardiovascular mortality in the community. Circulation 2009;119:2765–71.
25. Anderson JL, May HT, Horne BD, Bair TL, Hall NL, Carlquist JF, et al. Relation of vitamin D deficiency to cardiovascular risk factors, disease status, and incident events in a general healthcare population. Am J Cardiol 2010;106:963–8.
26. Pilz S, Dobnig H, Nijpels G, Heine RJ, Stehouwer CD, Snijder MB, et al. Vitamin D and mortality in older men and women. Clin Endocrinol 2009;71:666–72.
27. Cawthon PM, Parimi N, Barrett-Connor E, Laughlin GA, Ensrud KE, Hoffman AR, et al. Serum 25-hydroxyvitamin D, parathyroid hormone, and mortality in older men. J Clin Endocrinol Metab 2010;95:4625–34.
28. Pilz S, März W, Wellnitz B, Seelhorst U, Fahrleitner-Pammer A, Dimai HP, et al. Association of vitamin D deficiency with heart failure and sudden cardiac death in a large cross-sectional study of patients referred for coronary angiography. J Clin Endocrinol Metab 2008;93:3927–35.
29. Wang TJ, Pencina MJ, Booth SL, Jacques PF, Ingelsson E, Lanier K, et al. Vitamin D deficiency and risk of cardiovascular disease. Circulation 2008;117:503–11.
30. Melamed ML, Michos ED, Post W, Astor B. 25-Hydroxyvitamin D levels and the risk of mortality in the general population. Arch Intern Med 2008;168:1629–37.
31. Grandi NC, Breitling LP, Brenner H. Vitamin D and cardiovascular disease: systematic review and meta-analysis of prospective studies. Prev Med 2010;51:228–33.
32. Pittas AG, Chung M, Trikalinos T, Mitri J, Brendel M, Patel K, et al. Systematic review: vitamin D and cardiometabolic outcomes. Ann Intern Med 2010;152:307–14.
33. Parker J, Hashmi O, Dutton D, Mavrodaris A, Stranges S, Kandala NB, et al. Levels of vitamin D and cardiometabolic disorders: systematic review and meta-analysis. Maturitas 2010;65: 225–36.
34. Wang L, Manson JE, Song Y, Sesso HD. Systematic review: vitamin D and calcium supplementation in prevention of cardiovascular events. Ann Intern Med 2010;152:315–23.
35. Doorenbos CR, von den Born J, Navis G, de Borst MH. Possible renoprotection by vitamin D in renal disease: beyond mineral metabolism. Nat Rev Nephrol 2009;5:691–700.
36. Mehrotra R, Kermah DA, Salusky IB, Wolf MS, Thadhani RI, Chiu YW, et al. Chronic kidney disease, hypovitaminosis D, and mortality in the United States. Kidney Int 2009;76:977–83.
37. Drechsler C, Pilz S, Obermayer-Pietsch B, Verduijn M, Tomaschitz A, Krane V, et al. Vitamin D deficiency is associated with sudden cardiac death, combined cardiovascular events, and mortality in haemodialysis patients. Eur Heart J 2010;31:2253–61.
38. Pilz S, Tomaschitz A, Friedl C, Amrein K, Drechsler C, Ritz E, et al. Vitamin D status and mortality in chronic kidney disease. Nephrol Dial Transplant 2011; DOI: 10.1093/ndt/gfr076.
39. 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.
40. Kidney-Disease: Improving Global Outcomes (KDIGO) CKD-MBD Work Group. KDIGO clinical practice guideline for the diagnosis, evaluation, prevention, and treatment of chronic kidney disease-mineral and bone disorder (CKD-MBD). Kidney Int Suppl 2009;113:S1–130.

41. Kalantar-Zadeh K, Kovesdy CP. Clinical outcomes with active versus nutritional vitamin D compounds in chronic kidney disease. Clin J Am Soc Nephrol 2009;4:1529–39.
42. Shoenfeld N, Amital H, Shoenfeld Y. The effect of melanism and vitamin D synthesis on the incidence of autoimmune disease. Nat Clin Pract Rheumatol 2009;5:99–105.
43. Hyppönen E, Läärä E, Reunanen A, Järvelin MR, Virtanen SM. Intake of vitamin D and risk of type 1 diabetes: a birth-cohort study. Lancet 2001;358:1500–3.
44. Prietl B, Pilz S, Wolf M, Tomaschitz A, Obermayer-Pietsch B, Graninger W, et al. Vitamin D supplementation and regulatory T cells in apparently healthy subjects: vitamin D treatment for autoimmune diseases? Isr Med Assoc J 2010;12:136–9.
45. Pilz S, Dobnig H, Fischer JE, Wellnitz B, Seelhorst U, Boehm BO, et al. Low vitamin D levels predict stroke in patients referred to coronary angiography. Stroke 2008;39:2611–3.
46. Pilz S, Tomaschitz A, Drechsler C, Zittermann A, Dekker JM, März W. Vitamin D supplementation: a promising approach for the prevention and treatment of strokes. Curr Drug Targets 2011;12:88–96.
47. Stewart A, Wong K, Cachat J, Elegante M, Gilder T, Mohnot S, et al. Neurosteroid vitamin D system as a nontraditional drug target in neuropsychopharmacology. Behav Pharmacol 2010;21: 420–6.
48. Holick MF. McCollum Award Lecture, 1994: vitamin D–new horizons for the 21st century. Am J Clin Nutr 1994;60:619–30.
49. Schwalfenberg GK. A review of the critical role of vitamin D in the functioning of the immune system and the clinical implications of vitamin D deficiency. Mol Nutr Food Res 2011;55:96–108.
50. Martineau AR, Timms PM, Bothamley GH, Hanifa Y, Islam K, Claxton AP, et al. High-dose vitamin D(3) during intensive-phase antimicrobial treatment of pulmonary tuberculosis: a double-blind randomised controlled trial. Lancet 2011;377:242–50.
51. Urashima M, Segawa T, Okazaki M, Kurihara M, Wada Y, Ida H. Randomized trial of vitamin D supplementation to prevent seasonal influenza A in schoolchildren. Am J Clin Nutr 2010;91:1255–60.
52. Blomberg Jensen M, Nielsen JE, Jørgensen A, Rajpert-De Meyts E, Kristensen DM, Jørgensen N, et al. Vitamin D receptor and vitamin D metabolizing enzymes are expressed in the human male reproductive tract. Hum Reprod 2010;25:1303–11.
53. Pilz S, Frisch S, Koertke H, Kuhn J, Dreier J, Obermayer-Pietsch B, et al. Effect of vitamin D supplementation on testosterone levels in men. Horm Metab Res 2011;43:223–5.
54. Wehr E, Pilz S, Boehm BO, März W, Obermayer-Pietsch B. Association of vitamin D status with serum androgen levels in men. Clin Endocrinol (Oxf) 2010;73:243–8.
55. Wehr E, Pilz S, Schweighofer N, Giuliani A, Kopera D, Pieber TR, et al. Association of hypovitaminosis D with metabolic disturbances in polycystic ovary syndrome. Eur J Endocrinol 2009;161:575–82.
56. Lewis S, Lucas RM, Halliday J, Ponsonby AL. Vitamin D deficiency and pregnancy: from preconception to birth. Mol Nutr Food Res 2010;54:1092–102.
57. Ish-Shalom S, Segal E, Salganik T, Raz B, Bromberg IL, Vieth R. Comparison of daily, weekly, and monthly vitamin D3 in ethanol dosing protocols for two months in elderly hip fracture patients. J Clin Endocrinol Metab 2008;93:3430–5.
58. Macdonald HM, Mavroeidi A, Fraser WD, Darling AL, Black AJ, Aucott L, et al. Sunlight and dietary contributions to the seasonal vitamin D status of cohorts of healthy postmenopausal women living at northerly latitudes: a major cause for concern? Osteoporos Int 2010; DOI: 10.1007/s00198-010-1467-z.
59. Hathcock JN, Shao A, Vieth R, Heaney RP. Risk assessment for vitamin D. Am J Clin Nutr 2007;85:6–18.
60. Ross AC, Manson JE, Abrams SA, Aloia JF, Brannon PM, Clinton SK, et al. The 2011 report on dietary reference intakes for calcium and vitamin D from the Institute of Medicine: what clinicians need to know. J Clin Endocrinol Metab 2011;96:53–8.
61. Schwalfenberg G. Not enough vitamin D: health consequences for Canadians. Can Fam Physician 2007;53:841–54.
62. Lehtonen-Veromaa M, Möttönen T, Leino A, Heinonen OJ, Rautava E, Viikari J. Prospective study on food fortification with vitamin D among adolescent females in Finland: minor effects. Br J Nutr 2008;100:418–23.
63. Autier P, Gandini S. Vitamin D supplementation and total mortality: a meta-analysis of randomized controlled trials. Arch Intern Med 2007;167:1730–7.