Guidelines for the prevention and treatment of hypovitaminosis D with cholecalciferol

Guidelines on prevention and treatment of vitamin D deficiency

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SUMMARY

The Italian Society for Osteoporosis, Mineral Metabolism and Bone Diseases (SIOMMMS) has elaborated the following guidelines about the definition, prevention and treatment of inadequate vitamin D status. The highlights are presented here.

- Daily vitamin D allowance ranges from 1,500 IU (healthy adults) to 2,300 IU (elderly with low calcium intake). Since the average Italian diet includes around 300 IU/day, subjects with no effective sun exposure should be supplemented with 1,200-2,000 IU vitamin D per day.
- The serum 25-hydroxy-vitamin D [25(OH)D] levels represents the most accurate way to assess vitamin D repletion, even though there are still no standardized assay methods.
- Conditions of “deficiency” and “insufficiency” are defined by the following ranges of 25(OH)D levels: less than 20 ng/ml and 20-30 ng/ml, respectively.
- In Italy, approximately 50% of young healthy subjects have vitamin D insufficiency during the winter months. The prevalence of deficiency increases with ageing, affecting almost all elderly subjects not on vitamin D supplements.
- When a condition of deficiency has been identified, a cumulative dose of 300,000-1,000,000 IU, over 1-4 weeks is recommended.
- In subjects recently treated for deficiency-insufficiency, a maintenance dose of 800-2,000 IU/day (or weekly equivalent) is recommended. In patients on daily doses over 1,000 IU, 25(OH)D levels should be checked regularly (e.g. once every two years).
- The highest tolerated daily dose has been identified as 4,000 IU/day.
- Vitamin D supplementation should be carefully monitored in patients at higher risk of vitamin D intoxication (granulomatosis) or with primary hyperparathyroidism.
- In pregnant women, vitamin D supplements should be given as in non-pregnant women, but bolus administration (i.e.: single dose >25,000 IU) should be avoided.

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n PHARMACOLOGY OF VITAMIN D

Production and metabolism The actions of vitamin D are attributable to its active metabolite, i.e. 1,25-dihydroxycholecalciferol [1,25(OH)2 D3] or calcitriol, which is produced through a series of enzymatic steps starting from cholecalciferol or vitamin D3. Ergocalciferol, or vitamin D2 , follows the same metabolic steps. Both vitamin D3 and vitamin D2 can be taken in the diet but the predominant portion of vitamin D3 derives from the conversion of 7-dehydrocholesterol (or provitamin D) (1) following exposure of the skin to ultraviolet rays of specific wavelength (UVB between 290 and 315 nm) (2). Sunlight is characterized by the presence of this radiation only for a limited number of hours, which however varies in relation to the season and latitude. For this reason, in Italy, the production of vitamin D is linked

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sun exposure is negligible in the winter months. Other factors that strongly influence vitamin synthesis are age (for the same amount of sun exposure, elderly subjects produce 30% less) (3, 4), the surface and thickness of the skin exposed to the sun, the irradiation time , as well as the use of protective creams, which can reduce the skin synthesis of vitamin D by 97%.

In temperate latitudes, 80% of the need for vitamin D is guaranteed by solar radiation and the remaining 20% is ensured by nutrition (5). Vitamin D3 is contained almost exclusively in animal fats, while the amount of vitamin D2 present in some vegetable fats is negligible.

Vitamin D comes 4/5 from sun exposure. It is deposited in adipose tissue and the amount that is released is immediately converted in the liver into 25(OH)D, whose serum concentrations represent a precise indicator of vitamin deposits. The 25(OH) D is converted to the active metabolite 1,25(OH)2 D primarily in the kidney as part of a homeostatic mechanism involving serum levels of PTH, calcium and phosphorus.

Vitamin D is highly fat-soluble. It is rapidly absorbed at the duodenal and jejunal level and then distributed through the lymphatic circulation almost entirely to the adipose tissue, from which it is released in small quantities compared to the stored amount. Therefore, a greater adipose mass "dilutes" vitamin D, which explains why the risk of its deficiency is higher in obese subjects.

Vitamin D as such remains in circulation only for a very short time and its blood concentrations are therefore very low (1-2 ng/ml) and, during hepatic transit, are converted into 25-hydroxylecalciferol [25(OH)D3] by the enzyme 25-hydroxylase. This metabolic process can take place even in the presence of a reduction in functioning liver tissue. However, quite recently a high prevalence of hypovitaminosis D has been documented in patients suffering from HCV-related chronic hepatitis; in patients with genotype 1 this condition seems to interfere with the therapeutic response to interferon (6).

25(OH)D is the main circulating metabolite of vitamin D and its serum concentrations constitute the most reliable biochemical index of the state of vitamin repletion (5, 7). The 25(OH) D is a partially hydrophilic metabolite that is deposited only in the liver and muscles; its half-life is shorter than that of vitamin D and such as to satisfy the requirement for no more than 12-18 days (8, 9). After this period, in patients with vitamin D deficiency who have been administered 25(OH)cholecalciferol, serum 25(OH)D levels tend to reduce rapidly after treatment is stopped; the administration of higher monthly doses of calcifediol overcomes this problem (10).

The conversion into 1,25(OH)2 D3 (or calcitriol) by 1-alpha-hydroxylase is carried out at the level of various tissues (see below) although the most relevant portion relating to the control of mineral metabolism occurs in the proximal renal tubules, requires the presence of parathyroid hormone (PTH) and is partly modulated by serum calcium and phosphorus levels. The di-hydroxylated metabolites have a very short half-life and are not deposited at the tissue level.

In subjects suffering from renal failure, the production of 1,25(OH)2 D is progressively compromised. However, alpha-hydroxylase activity is no longer able to ensure normal hormonal levels only in the presence of a significant deterioration in renal function. Consequently, prevention of vitamin D deficiency with hydroxylated metabolites is justified only in patients with advanced renal failure (stage IV-V). However, it must be underlined that, even in these patients, 25(OH)D levels must be maintained within the normal range to ensure an adequate substrate for extra-renal 1-alpha-hydroxylases (11).

On the other hand, treatment with final active metabolites of vitamin D could be irrational and in other conditions – or even potentially risky, since 1-hydroxylation is the limiting step in the synthesis process of active vitamin D; it therefore constitutes a protection mechanism against possible intoxication, which is preserved with other types of supplementation.

Studies conducted in patients with osteoporosis

metasteroidea, would however demonstrate a greater effectiveness of calcitriol compared to cholecalciferol in preventing the risk of fracture.

Mechanism of action

Once activated at 1,25(OH)2 D, vitamin D binds to a specific receptor (VDR) which belongs to the superfamily of steroid receptors. In reality, two types of vitamin D receptors have been identified. The first, located in the nucleus, is able to directly stimulate the transcription of genes and therefore the ex-novo synthesis of proteins (genomic mechanism). The other receptor is located on the cell membrane and acts by inducing the formation of second cellular messengers (such as cAMP, diacylglycerol, inositol triphosphate, arachidonic acid) or by phosphorylating some cellular proteins. This non-genomic mechanism of action is capable of rapidly modulating the cellular response to various stimuli (12).

Vitamin D receptors are practically ubiquitous, demonstrating their important physiological role, not only in mineral metabolism but also in numerous other functions of the organism. The affinity of the vitamin D receptor for 1,25(OH)2 D is one thousand times greater than that for 25(OH)D or other metabolites.

A. *Effects on intestine, bone tissue and muscle*

L'1,25(OH)2 D determines an increase of intestinal calcium absorption through the induction of the synthesis of a protein expressed on the brush border of intestinal epithelial cells, which binds the ion and transports it from the lumen to the cell cytoplasm. Furthermore, 1,25(OH)2 D also facilitates the passive absorption of calcium, increasing the permeability of intercellular tight junctions.
The action of vitamin D on bone tissue occurs through the interaction with 1,25(OH)2 D receptors expressed by osteoblasts; it has been observed that vitamin D promotes the synthesis of some proteins, especially osteocalcin, which are fundamental for the homeostasis of bone tissue. 1,25(OH)2 D also induces the production of RANKL thus promoting the activation of osteoclasts.

1,25(OH)2 D is able to stimulate the production of muscle proteins but above all to activate some calcium transport mechanisms at the level of the sarcoplasmic reticulum, which are essential for muscle contraction. In conditions of hypovitaminosis D, pictures of proximal myopathy (difficulty in getting up from the chair, functional impotence in bringing the arms above the head, etc.), of sarcopenia and reduction of muscular strength, with balance disorders and resulting in an increased risk of falls (13-24).

Vitamin D deficiency, especially if prolonged over time, can lead to real disabilities. The histological counterpart consists of an atrophy of type II muscle fibers, an increase in spaces between muscle fibrils and the replacement of muscle tissue with adipose cells and fibrous tissue (25). In elderly subjects this phenomenon can add to the physiological loss of muscle mass (sarcopenia). A reduction in the risk of falls following the administration of vitamin D in elderly subjects has been reported (26-30).

B. *Extra-skeletal effects*

Vitamin D also performs important functions outside the musculoskeletal tissue. In fact, the presence of receptors for vitamin D has been observed in various cell types and the expression of 1ÿ-hydroxylase has been documented, i.e. the ability to produce 1,25(OH)2 D, in activated macrophages, in osteoblasts, in keratinocytes and at the prostate, colon and breast level.
Local production of 1,25(OH)2 D does not contributes to the maintenance of calcium homeostasis, as demonstrated by the fact that nephrectomized patients or patients with severe renal failure are characterized by practically unwearable levels of 1,25(OH)2 D. The local production of 1,25(OH)2 D appears to be implicated in the mechanisms of paracrine regulation of cell growth, including tumor growth (31-37).

1,25(OH)2D, is to metabolite of neoplasms ***exerts*** colon) (39). ***its actions through The*** and the correlation detected between the levels the renin suggest a ***It exerts a crucial*** phosphorus capacity is ***intestinal calcium absorption, reabsorption of calcium and phosphorus and positively interfering on osteoblastic and osteoclastic activity.***

It also promotes some muscle functions. Finally, 1,25(OH)2D contributes to the control of cell proliferation, with effects on the immune system, the skin and numerous neoplasms.

This documented anti-proliferative activity has promoted the use of 1,25(OH)2 D o of its metabolites in the treatment of psoriasis (38) and has made ***it*** hypothesize its use in some types of ***active*** (prostate, breast and ***vitamin D which*** identification of the VDR in the renal tissue with negative ***specific receptors (VDR)*** should be of 1,25(OH)2 D ***nuclear or located on*** possible ***cell surface.*** role of vitamin D also in the regulation of arterial blood pressure ***(40).*** Calcium- also known , ***increasing the efficiency of increasing renal tubular*** ability of vitamin D to modulate the activity of the immune system, through actions exerted on B and T lymphocytes and on cells of the monocyte-macrophage lineage. In this sense, correcting vitamin D deficiency could contribute to reducing the risk of various autoimmune diseases including type 1 diabetes (41). In relation to the latter pathology, it should be remembered that the cells of the pancreatic islets express VDR receptors and that 1,25(OH)2 D is able to promote the synthesis and secretion of insulin.

However, for some of the extraskeletal effects hypothesized based on association studies, solid demonstration is still lacking scientific, since the few intervention studies have not provided unequivocal results.

n OPTIMAL LEVELS OF 25(OH)D

Although an adequate standardization of dosing methods has not yet been achieved (42), the serum concentration of 25(OH)D is considered the best clinical indicator of the bioavailable vitamin reserve. The definition of normal 25(OH)D levels has changed profoundly over the last decade. The first approach was based on the minimum levels of 25(OH)D necessary to ensure optimal intestinal calcium absorption. This limit was established as 32 ng/ml (80 nmol/l) (43). Others they considered the 25(OH) level to be optimal D associated with PTH values within the normal range in 90-100% of the population studied. This latter approach defines very different estimates of the 25(OH) threshold D, i.e. 32-50 nmol/l (12.8-20 ng/ml) and 68-78 nmol/l (27.2-31 ng/ml), depending on the statistical method used in the construction of the regression (44, 45). Furthermore, the approach linked to PTH levels is also affected by the duration of the deficiency state (until the development of forms similar to tertiary hyperparathyroidism) and the calcium intake in the diet, as a condition of vitamin D deficiency can be masked by a particularly generous dietary calcium intake and vice versa (46). On the other hand, serum PTH levels are significantly affected by vitamin D status for 25(OH)D concentrations lower than 16.3 ng/ml and by calcemia for vitamin levels above this threshold (47).

Theoretically, the most accurate method for estimating minimum vitamin D concentrations would be to detect the appearance of histological signs of osteomalacia (see below); however this approach is impractical and probably also insensitive.
Over the last decade, various observational studies have documented a relationship between 25(OH)D levels and various other parameters: bone mineral density (BMD) values (48, 49), propensity to fall, incidence of fractures, cardiovascular diseases, neoplasms (especially colon, breast and prostate), depressive syndromes, diabetes, multiple sclerosis and numerous other pathological conditions. The relationship between 25(OH)D levels and relative risk for some of these conditions would seem to indicate that optimal 25(OH)D concentrations are around 75-100 nmol/l (or 30-45 ng/ml) (49) [(Fig. 1), modified from 49].
In recent years, several authors, on the basis of meta-analyses and cross-sectional studies, have identified the threshold value of an adequate vitamin D status with a serum concentration of 25(OH)D equal to 32 ng/ml. However, it must be underlined that, in the absence of intervention studies, this threshold value cannot be conclusively defined. Co-

yes, recently these normality thresholds have been called into question by a *report* from the Institute of Medicine (IOM) (51).
In this document, with regards to the threshold levels of 25(OH)D below which negative consequences occur, the following points are developed:

A. *Rickets and osteomalacia*

In the presence of a normal dietary calcium intake, the risk of rickets increases for 25(OH)D levels lower than 25 nmol/l (10 ng/ml) (43) but is still minimal for levels between 25 and 50 nmol/l. Recent autopsy data (52) have documented the presence of osteomalacia for 25(OH)D levels lower than or equal to 50 nmol/l (20 ng/ml). However, when calcium intake is inadequate, levels above 75 nmol/l (30 ng/ml) may be necessary to avoid the risk of osteomalacia.

B. *Risk of fracture*

Results of intervention studies

There is a wide variability of data. In some studies it is concluded that 25(OH)D levels above approximately 40 nmol/l are sufficient to ensure adequate bone protection, but in others this threshold is set at 50 nmol/l (53-57).

Results of observational studies

Numerous observational studies have linked 25(OH)D levels to the risk of fracture (mainly of the femur) (58, 59). In all these studies, low levels of 25(OH)D are associated with a higher risk of fracture. The threshold values above which no further benefits are observed are not very unambiguous and vary from 40 nmol/l (60) to 50 nmol/l (61) to 60-70 nmol/l (62-65).

It should be underlined that with the exception of the study conducted in Sweden (60) where the calcium intake is very high and the concentrations of 25(OH)D rather homogeneous, most of the other studies indicate that a a significant increase in the risk of fracture is observed for 25(OH)D values lower than approximately 60 nmol/l. On the other hand, this last threshold was identified by the IOM as 50 nmol/l, in con-

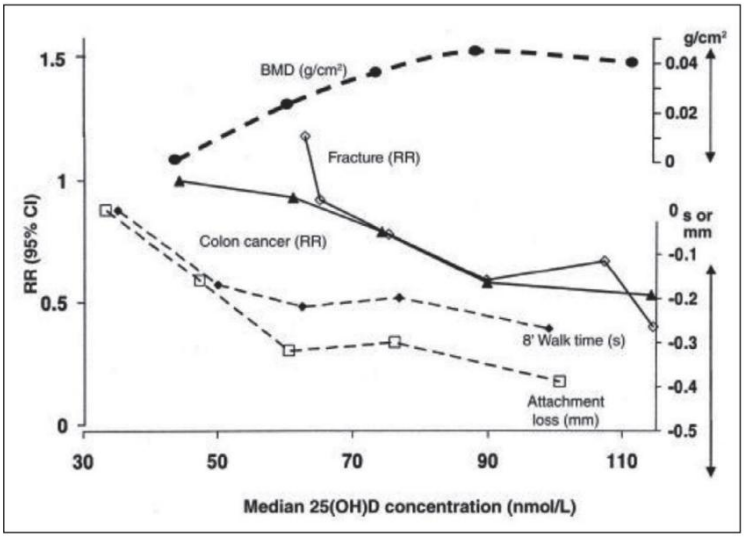

Figure 1 - Relationship between 25(OH)D levels and relative risk for some pathological conditions.

moved to other authoritative positions (66). In particular, it should be remembered that in 2 meta-analyses of randomized controlled *trials* (RCTs) a threshold of 50 nmol/l was found to be inadequate to guarantee a reduction in the risk of fractures and falls (28, 67).
Furthermore, from the NHANES study, the relationship between BMD and 25(OH)D levels demonstrated a clear persistence of the positive correlation for values well above 50 nmol/l (48,49).
The partial inconsistency of these positions and the results of the various studies can be attributed to the interaction between 25(OH)D levels and dietary calcium intake, since in the presence of a particularly rich calcium diet, levels may be sufficient of lower 25(OH)D and vice-versa (46, 68).
It should also be remembered that the expression of vitamin D receptors (and therefore the response to vitamin D) is reduced with advancing age at the level of all tissues, and in particular the intestine, kidney and muscle (69). The mechanism of renal regulation of 1,25(OH)2 D production in response to PTH also appears to attenuate with aging (70, 71). These latter observations would therefore lead us to believe that in the elderly the optimal levels of 25(OH)D must be slightly higher than those identified for young people.
In conclusion considering that:

a) desirable serum 25(OH)D levels

Table I - Interpretation of blood 25(OH)D levels.

Definition	nmol/L	of/ml
Shortage	<50	<20
Insufficiency	50-75	20-30
Excess	>250	>100
Intoxication	>375	>150

they probably increase with age (46);

b) in Italy the calcium intake is generally very modest (46, 72, 73);
c) the risk of vitamin D poisoning (see below) is rather remote;

it appears reasonable to conservatively identify the vitamin deficiency threshold with a serum concentration of 25(OH) D inferior a 75 nmol/l (30 ng/ml).

These guidelines therefore consider the ranges of 25(OH)D levels reported by Holick to be acceptable (74) (Tab. I).

In Table I, deficiency is defined as a marked deficiency and insufficiency as a subtle *(or less severe)* deficiency of vitamin D, while concentrations above 30 ng/ml (75 nmol/l) can be considered optimal (75). However, it must be remembered that according to some authors even serum 25(OH)D levels higher than 20 ng/ml (50 nmol/l) would be associated with a reduction in bone turnover and the risk of fracture (76, 77) and could therefore be considered optimal.

Vitamin D insufficiency affects almost all of the elderly Italian population who do not take vitamin D supplements. This appears to be linked to diet (little animal fats, infrequent addition of vitamin D to foods) and poor sun exposure.

Vitamin D insufficiency affects approximately 50% of young people, at least in the winter months.

n PREVALENCE OF DEFICIT OF VITAMIN D IN ITALY

Vitamin D deficiency is particularly frequent in Italy, especially in the elderly and in the winter months (4, 78-85). The deficiency is so common and of such magnitude that 86% of Italian women over 70 have blood levels of 25(OH)D lower than 10 ng/ml at the end of winter (72). This finding takes on dramatic aspects in institutionalized subjects or with other concomitant pathologies (78, 83, 86), both due to the poor sun exposure and dietary imbalances. Sun exposure, in terms of both exposed body surface and irradiation time, reduces with advancing age, in relation to socio-cultural reasons or ocular or skin pathologies that limit its tolerability. It should also be remembered that the skin production of vitamin D, with the same ultraviolet exposure, decreases with age. Even the intake of foods that contain the greatest quantities of vitamin D (milk and derivatives, animal fats) decreases with age for fear of increasing cardiovascular risk.

A worrying picture has also emerged from international polycentric studies which have unexpectedly documented a greater prevalence of hypovitaminosis D in Southern European countries (Italy, Greece and Spain) (79, 82, 87). In Italy, vitamin D is not added to foods and consequently the correction of the deficit is commonly entrusted to pharmacological supplementation.

In clinical practice, the combination of calcium and vitamin D is usually indicated and used for the treatment of osteopenia and postmenopausal osteoporosis, available in various pharmaceutical formulations in our country. This treatment has been shown in various clinical *trials* to reduce the risk of hip fractures in particular groups of patients (88, 89). Due to the very poor compliance with these formulations (90, 91) it often happens that even in patients being treated with drugs for the treatment of osteoporosis, vitamin D supplementation is infrequent and stopped early (92), despite the recommendation reported in the leaflet. illustrative of drugs for osteoporosis and in AIFA Note 79. It has been documented in Italy that this tends to nullify the therapeutic *outcomes* of the drugs used for osteoporosis (93-95).

The problem of vitamin D deficiency is not exclusive to the elderly.
A recent study aimed at establishing the normal values of mineral metabolism markers in young and healthy subjects has demonstrated that vitamin D deficiency

it affected 30 and 65% of subjects for cut-offs of 25(OH)D <20 ng/ml or <30 ng/ml, respectively (96). The problem worsens in the winter months even in young people and in particular in women (81).

n PREVENTION STRATEGIES AND CURE FOR DEFICIENCY OF VITAMIN D WITH COLECALCIFEROLO

Factors determining serum 25(OH)D levels and their changes in response to oral administration of vitamin D.

Several factors, including the initial 25(OH)D level, influence the increase in serum 25(OH)D in response to a given dose of vitamin D. With a dose of 2.5 ÿg (100 *IU/ day),* the average increase varies from 2.75 nmol/l (1.1 ng/ml) for low initial levels of 25(OH)D to 1.75 nmol/l (0.7 ng/ml) for higher initial levels (near optimal) (97). The increase in 25(OH) levels D in response to a given dose of vitamin D is also a function of body mass, being lower in subjects with a high body mass index (BMI) compared to those with a normal BMI (98, 99). Other factors, which also affect 25(OH)D levels, have no known impact on the response of 25(OH)D to vitamin D supplementation. For example, estrogen use increases measured serum levels of 25(OH)D via increased levels of vitamin D binding protein (100), but does not alter the increase in serum 25(OH)D achieved through supplementation. Furthermore, serum 25(OH)D levels decrease with aging, but the response of serum 25(OH)D to a given dose of vitamin D supplementation is independent of age (101). Finally, dietary calcium intake, within the range normally consumed, does not affect the 25(OH) response

Serum D to vitamin D supplementation. This latter concept must be distinguished from observations that calcium requirements may depend on vitamin D status (46, 102).

Vitamin D requirement

To estimate the need for vitamin D it is necessary to know the quantity normally used by the body. The latter has never been defined and must therefore be estimated empirically, on the basis of the doses necessary to reach and maintain the desirable serum levels of 25(OH)D indicated above.
In subjects with vitamin D deficiency or insufficiency and therefore probably with a very low dietary/solar intake (for example <400 IU/day) a daily dose of less than 2,000 units is not able to allow the achievement of adequate levels of 25(OH)D (Adami S: personal data; 103, 104).
Based on dosages used in various *trials* for the treatment of osteoporosis, which involved the administration of vitamin D to patients with deficiency or insufficiency, it was estimated that to achieve 25(OH)D levels equal to or greater than 75 nmol/l it is necessary to take between 1,800 and 4,000 IU of vitamin D per day (105, 106). We can therefore reasonably hypothesize an average daily requirement of vitamin D of around 1,500-2,300 IU/day, taking into account that it can increase with age, body mass, fat mass and calcium intake.

The problem of defining the daily requirement can also be addressed by relating the vitamin D intake and the 25(OH)D concentrations. The most consistent data in this regard are those collected in the NHANES study, 2005-2006 (107). The results of this investigation in women are summarized in Table II, where the average levels of 25(OH)D detected in Italy for similar age ranges are also reported. Note that, if a 25(OH)D value of 75 nmol/l is assumed as the normal threshold, the prevalence of vitamin D deficiency in Italy appears slightly lower than in the USA in young subjects, but significantly higher among the elderly.
In the rereading of these data by the IOM (51) it is assumed that sun exposure contributes only 30% to the intake of vitamin D, which contradicts the experimental data obtained in the USA and North

America (5, 74), according to which sun exposure contributes 80%.
In Italian epidemiological studies (72, 96) a difference of approximately 40% has been documented between the 25(OH)D values detected in subjects with minimal and in those with medium sun exposure, suggesting that the latter contributes 60%. -90% of vitamin D needs.
These percentages are very relevant for estimating the global daily requirement (sun, food, supplements) of vitamin D to achieve normal 25(OH) levels
D. The consequences on the estimate of vitamin D requirements, using NHA-NES data and assuming the contribution of sun exposure to 25(OH)D levels equal to 30% or 80% are shown in Table III .
The global intake (sun, food, supplements) of vitamin D in the USA in elderly subjects is approximately 600 IU/day if a contribution from sun exposure of 30% is assumed (400 oral +200 sun exposure) which rises to 2,000 IU/day if the share attributed to sun exposure is equal to 80% (400 oral +1,600 sun exposure).
Estimating the daily requirement of vitamin D can also be done following another approach. It has been observed that for every 100 IU of vitamin D taken, 25(OH)D levels increase by 1 ng/ml (2.5 mmol/l). If this relationship remained linear for any initial level of 25(OH)D, the daily administration of 2,000 IU of vitamin D3 would still guarantee serum concentrations equal to or greater than 20 ng/ml even in subjects with negligible daily exposure to sunlight (43).

It must be remembered that the estimate of a daily requirement of vitamin D equal to 2,000 IU (value obtained from the three analyzes set out above) is associated with sub-optimal average levels of 25(OH)D, so it appears reasonable to hypothesize that the optimal global intake (sun exposure, foods, sup-

Table II - Relationship between vitamin D intake and 25(OH)D concentrations: results of the NHANES study.

	Vitamin D intake (IU/day)			Levels of 25OHD nmol/l	
Age years (range)	Food	Supplements	Total	Media deer	Average Italy
<8	250	70	375	71	
9-18	190	70	260	58	
19-30	144	88	232	63	682
31-50	176	132	308	58	682
51-70	156	148	404	57	471
>70	180	120	400	56	401

1 Isaiah et al.; 2 Adami et al.

Table III - Estimate of the daily requirement of vitamin D in healthy subjects assuming that solar irradiation provides 30 or 80% of the average daily requirement.

Age years (range)	Global dietary intake of vitamin D	Daily requirement of vitamin D	
		UV irradiation contribution = 30%	UV irradiation contribution = 80%
<8	375	536	1.875
9-18	260	371	1.300
19-30	232	331	1.160
31-50	308	440	1.540
51-70	404	577	2.020
>70	400	571	2.000

plements) could also be higher than previously suggested.

Sources of vitamin D in Italy

Dietary intake of vitamin D and effective exposure to sunlight are the main factors determining the serum 25(OH)D level.

Vitamin D is present in foods in limited quantities. The main source is animal fats contained mainly in fatty fish (for example salmon) and dairy products.

In order to guarantee a greater intake of vitamin D through dairy products, it would be necessary to add these foods, as is currently the case only in Northern European countries and more widely in North America, where however this supplementation only guarantees an average intake. of 40-80 IU/ day in young subjects and 160-200 IU/day in people over 50 (108). This US estimate on vitamin D supplementation of the most commonly used foods was made a few years ago (109) but now takes on greater importance due to the growing perception of the role of vitamin D in the epidemiology of many diseases (51).

It is likely that the contribution of the diet to 25(OH)D levels in Italy is considerably lower than in the USA, both due to the type of diet (less animal fats) and the infrequent supplementation of vitamin D in high-quality foods. common use. This exposes elderly people with little sun exposure even more to the deficiency.

In past centuries, exposure to the sun during the summer months (the ones effective for the skin synthesis of vitamin D) was the unavoidable necessity of a population mainly dedicated to outdoor work and which included a lower percentage of elderly people. Today sun exposure is mostly discretionary and appreciated only by some young people. The majority of the population, particularly with advancing age, limits exposure to the sun and uses protective creams more and more often, which reduce the photosynthesis of the vitamin. In hiring populations a diet low in vitamin D (as is the Mediterranean diet in general) its deficiency among the elderly inevitably takes on epidemic dimensions.

Use of vitamin D supplements

The recommended dose of vitamin D is expressed as a daily dosage. However, at the same cumulative dose, vitamin D can also be administered through weekly boluses (110). To improve adherence to treatment it is also possible to resort to monthly or quarterly boluses; in this case the equivalent doses, according to some authors, should be greater than the cumulative daily or weekly doses (111). In some cases, an annual bolus was also used for practical reasons (90).

However, it should be noted that, according to a recent study, the administration of an annual bolus of 500,000 IU of cholecalciferol to a group of elderly women induced the paradoxical effect of increasing the risk of falls and fractures (112).

Despite the gross methodological limitations of this research, in which the risk factors for falls and fractures had not been controlled and in which the population studied had initial levels of 25(OH)D that were double compared to the elderly Italian population, the results of this study suggest a community preventive approach with smaller boluses (maximum of 300,000 IU).

This approach would seem at least initially justified by the pharmacokinetics of vitamin D with the aim of "saturating" the body's stores.

Vitamin D is normally administered orally, limiting the use of the intramuscular route to patients with severe malabsorption syndromes.

The recommended dosage schedules must take into account potential interference from other drugs or pathological conditions (Tab. IV).

The dosage of vitamin D to be recommended can therefore also vary depending on the clinical condition treated and the therapeutic objectives that are set.

The need for vitamin D varies from 1,500 IU/ day (healthy adults) to 2,300 IU/ day (elderly). The diet in Italy provides on average around 300 IU/ die, so when sun exposure is virtually absent, supplements of 1,200-2,000 IU/ day must be guaranteed.

Table IV - Clinical conditions that interfere with vitamin D metabolism.

Decreased bio-availability
A. Fat malabsorption
- Cystic fibrosis
- Celiac disease
- Whipple's disease
- Crohn's disease
- Gastro-intestinal by-pass surgery
- Drugs that reduce the absorption of fats
- Other
B. Reduced availability
- Obesity with sequestration of vitamin D in adipose tissue
Increased catabolism/consumption
- Anticonvulsants
- Glucocorticoids
- Drugs for the treatment of AIDS or anti-rejection.
- Breastfeeding and pregnancy
Decreased synthesis of 25(OH)D (also administer calcidiol)
- Severe liver failure
Urinary losses of 25(OH)D
- Nephrotic syndrome
Decreased synthesis of 1,25(OH)2D (also administer calcitriol)
- Chronic renal failure
- Iperfosforemia
- Congenital deficiencies of 1-hydroxylase

Prevention of vitamin D insufficiency

By prevention of insufficiency we mean the dose of vitamin D to be recommended to subjects in whom the vitamin deficiency has already been corrected and the supplementation is aimed at preventing its recurrence.
In many guidelines it has been preferred to give universal recommendations that are easy to understand and adopt.
In particular, a minimum daily intake of 200 IU of vitamin D is recommended from the second month of life until adolescence (113), 400 IU in adulthood and at least 600 IU (15 µg) over the age of 70 (114).
These minimum recommended intakes have recently been revised by the IOM, which proposes 400 IU up to 1 year of life, 600 IU up to 70 years of age (even during pregnancy and breastfeeding) and 800 IU over 70 years of age (51) .
These dosages may be considered acceptable in contexts where prevalence of the deficiency states is considerably lower than in Italy and are capable of guaranteeing serum 25(OH)D levels higher than 50 nmol/l, but lower than those desired by these guidelines (75-80 nmol/l). In fact, supplementation with 600 IU/day in subjects over 70 years of age does not substantially modify the proportion of deficient subjects (110).
Measurement of serum 25(OH) levels D can be considered a good indicator of daily needs. However, the use of this dosage, as part of a generalized screening or even for periodic monitoring during supplementation, does not appear economically acceptable.

In young adults it has been calculated that a summer exposure on both surfaces of the body lasting 20-30 minutes/ per day in the middle hours of the day is equivalent to an intake of 10,000-20,000 IU of vitamin D.
It is therefore reasonable to believe that a young person (<60 years) with a lifestyle characterized by normal sun exposure (one seaside holiday per year, and more than 20 minutes/day of outdoor life in the summer) (72, 96), does not need to carry out checks to verify a possible state of insufficiency and therefore not even supplements.

In elderly subjects aged between 60 and 70, it can be assumed that a state of vitamin D insufficiency does not exist only in the presence of lifestyles characterized for example by prolonged summer holidays, with extensive sun exposure. For this age range (60-70 years) a control of 25(OH)D levels otherwise appears justified. However, a pragmatic approach involving "blind" supplementation with 600-1000 IU/day also appears acceptable, once the conditions in which vitamin D may be contraindicated are excluded (see overdose).

In people over the age of 70 (and increasingly with age) who do not take vitamin D supplements, vitamin D deficiency has a prevalence close to 100%. In these cases, where the control of 25(OH) is not essential

D serum, it is fully justified to start a "blind" treatment, after having excluded the presence of contraindications.
A scheme that still guarantees a wide margin of safety in terms of overdose could be represented by the administration of a single bolus of 300,000 IU in the winter months followed by 1,000 IU/day (or equivalent weekly or monthly).

For all the schemes mentioned above it must be remembered that in the presence of precarious nutritional conditions or intestinal malabsorption syndromes and in case of administration of some drugs (for example anticonvulsants or glucocorticoids) it is advisable to increase the doses or resort to the administration parenteral.
Although few studies have been published on this specific topic, the use of hydroxylated metabolites of vitamin D seems to be indicated only in some particular conditions.

Treatment of vitamin D deficiency and insufficiency

The objective of therapy for vitamin D deficiency and insufficiency is to restore normal serum levels and therefore 25(OH)D deposits in a short time. The cumulative dose to be administered over a few weeks may vary depending on the severity of the deficiency and body mass. The 25(OH)D value detected at the time of identification of the deficiency or insufficiency state can be considered a good indicator of the requirement (Table V).

This dose must be followed by a maintenance dose, to avoid returning to conditions of insufficiency or deficiency (103, 115); if the conditions persist have induced this state of deficiency or insufficiency (Table V), the dosage must be higher.
An accurate estimate of the effects of "therapeutic" doses of vitamin D has never been made, but there are some studies on the effect of vitamin D boluses on 25(OH) levels D in adults. In order to increase 25(OHD) concentrations, both vitamin D2 and vitamin D3 can be used , at adequate doses.

In this regard, it should in fact be noted that the increases in serum concentrations of 25(OH)D achieved through the administration of vitamin D3 are more pronounced than those obtained by administering vitamin D2 (116, 117).
It has been reported that a bolus of 100,000 IU of cholecalciferol raises 25(OH)D levels by 10 ng/ml for 3 months (118), one of 300,000 IU maintains levels above 32 ng/ml for at least two months (116) while 600,000 IU of approximately 40 ng/ml for 3 months (119).
In a study conducted in Australia, annual boluses of 500,000 IU resulted in increases in 25(OH)D of approximately 20 ng/ml within the first month (112).
If the deficiency is defined by the finding of 25(OH)D levels lower than 20 ng/ml, to achieve values certainly higher than 30 ng/ml, adults must be administered, over a few weeks, a cumulative dose including between 600,000 and 900,000 IU, in relation to the initial value of 25(OH)D. Table V shows some possible treatment schemes as a guide.

In subjects deficient or persistently at risk for deficiency, a check of serum 25(OH)D after 3-6 months may be appropriate.

Table V - Estimate of therapeutic and maintenance doses as a function of 25(OH)D levels in subjects who have not received supplements in the last year.

Baseline value of 25(OH)D	Cumulative therapeutic dose of vitamin D	Daily maintenance dose
<10 ng/ml or 25 nmol/l	1.000.000	2.000
10-20 ng/ml or 25- 50 nmol/l	600.000	1.000
20-30 ng/ml or 50-75 nmol/l	300.000	800

n SUPPLEMENTATION VITAMIN D AT AGE PEDIATRIC

In infants, vitamin D supplementation depends on the maternal intake of vitamin D (120). With artificial feeding, the Federal Food, Drug, and Cosmetic Act (121) recommends a dietary intake of 100 IU/100 Kcal and therefore an intake varying between 265 and 660 IU/day. In children under 6 months of age the recommended intake is 300 IU/day and in children over 6 months of age 400 IU/day. The Institute of Medicine has recently recognized that under one year of age the current evidence does not allow it to provide recommendations and it is only possible to hypothesize that an intake of 400 IU/day is adequate; the recommended dose increases to 600 IU after one year of life (51). This intake can increase if children are not exposed to sunlight and in any case in the winter months.

The dose of supplements possibly recommended must also take into account the possible presence of vitamin D in foods (artificial milk or baby food).

n VITAMIN D REQUIREMENT AND PREGNANCY

In the 1960s, some cases of Supravalvular Aortic Stenosis (SAS) associated with hypercalcemia, attributed to an excessive intake of vitamin D, were reported in Great Britain (122, 123). These observations have led to a drastic reduction in the use of supplements during pregnancy and some drug data sheets list pregnancy among the contraindications to the use of vitamin D. In subsequent years it was clarified that SAS actually corresponds to Williams Syndrome (124), condition linked to an alteration of the gene that controls the synthesis of elastin, in which there is an increased conversion of vitamin D into 25(OH)D.

In the following years, numerous experimental works on vitamin D intoxication during pregnancy were conducted, in which the onset of syndromes even vaguely similar to SAS in newborns has never been documented (120). Despite this, the technical data sheets of products containing vitamin D supplements continue to include very strict recommendations on the use of vitamin D in pregnant women.

These recommendations may have a significant negative impact in terms of public health. In fact, many works have clearly demonstrated that vitamin D deficiency during pregnancy has negative effects on both the mother and the newborn and that during pregnancy the prevention of vitamin D deficiency must be done as in non-pregnant women (125). The most recent guidelines recommend the administration of 600 IU/day of vitamin D (51).

n OVERDOSE

Excessive administration of vitamin D can cause intoxication, characterized by hypercalcemia and rapid deterioration of renal function. The few cases described in the past refer to the use of very high doses of vitamin D, used up to 30 years ago for the treatment of hypoparathyroidism.

According to some authors, dosages up to 2,000 IU/day or, more recently, up to 4,000 IU/die (51, 126) are completely safe after 9 years of age.

This dosage should, however, be commensurate with the non-pharmacological intake of vitamin D. In other words, if 2,000 IU/day in elderly subjects with little sun exposure can be considered absolutely safe (and in some cases even insufficient), the same dose may not be as safe in the long term in young, thin subjects with frequent sun exposure.

In some conditions (granulomatosis, primary hyper-parathyroidism) there is an abnormal, substrate-dependent conversion of 25(OH)D into the active metabolite 1,25(OH)2 D; therefore, in patients suffering from these pathologies the levels of 25(OH) D must be strictly maintained

within the normal range. In these cases, lower doses and periodic checks of serum 25(OH)D levels are recommended. Recently the Institute of Medicine identified the maximum tolerated dose as 4,000 IU/ day (51).

n CLINICAL UTILITY OF DOSAGE OF 25(OH)D

For obvious economic reasons it is not possible to propose the measurement of 25(OH)D levels in the entire population. It is therefore desirable to develop algorithms for estimating the risk of vitamin D deficiency, in order to select subjects in whom carrying out this dosage is useful. Quite recently, using the databases of 3 studies conducted on large cohorts of the general Italian population, an algorithm is being developed under the aegis of SIOMMMS which the company then intends to subject to prospective validation.

Deficiency [25(OH)D <20 ng/ ml] and insufficiency [25(OH)D between 20 and 30 ng/ ml] are very common conditions in the Italian population and their prevalence increases with advancing age. The measurement of serum 25(OH)D represents the most appropriate method to estimate the s However, epidemiological data allow us to assume that all elderly people who are not taking supplements are deficient. In the presence of deficiency or insufficiency, cumulative doses of vitamin D ranging between 300,000 and 1,000,000 IU should be administered over 1-4 weeks. The daily maintenance dose (or weekly, monthly or quarterly equivalents), after reaching normal levels of 25(OH)D, varies according to age and sun exposure, with a range between 800 and 2,0 A control of 25(OH)D levels is recommended after treatments with daily doses greater than 1,000 IU, for several years. Vitamin D supplements should be used with caution and with periodic monitoring of 25(OH) levels. D in patients with granulomatous diseases (such as sarcoidosis) and primary hyperparathyroidism. During pregnancy, vitamin D supplements can be used as in non-pregnant women, however avoiding the use of boluses.

SUMMARY

The Italian Society of Osteoporosis, Mineral Metabolism and Skeletal Diseases (SIOMMMS) has decided to develop guidelines relating to the definition of hypovitaminosis D and prevention and treatment strategies, summarized in the following points:

- Vitamin D requirement varies from 1,500 IU/day (healthy adults) to 2,300 IU/day (elderly, with low dietary calcium intake). The diet in Italy provides on average around 300 IU/day, so when sun exposure is virtually absent, supplements of 1,200-2,000 IU/day must be guaranteed.
- The measurement of serum 25-hydroxy-vitamin D [25(OH)D] represents the most accurate method for estimating the state of vitamin D repletion, although the measurement techniques are not yet adequately standardized.
- Thresholds have been identified for a condition of "deficiency" [25(OH)D <20 ng/ml] and "insufficiency" [25(OH)D between 20 and 30 ng/ml] of vitamin D status.
- Vitamin D insufficiency affects approximately 50% of young people in the winter months. The deficiency condition increases with advancing age until it affects almost all of the elderly Italian population who do not take vitamin D supplements.
- In the presence of severe deficiency, cumulative doses of vitamin D varying between 300,000 and 1,000,000 IU, over 1-4 weeks.
- Once the vitamin deficiency has been corrected, the daily prevention - maintenance dose varies according to age and sun exposure, with a range between 800 and 2,000 IU/day or equivalent weekly.
  A check of 25(OH)D levels is recommended approximately every two years for treatments with daily doses greater than 1,000 IU.
- The maximum daily dose beyond which the risk of intoxication is considered high has been identified as 4,000 UI.
- Vitamin D supplements should be used with caution and with periodic monitoring of 25(OH) levels D in patients with granulomatous diseases or primary hyperparathyroidism.
- During pregnancy, vitamin D supplements can be administered as in non-pregnant women pregnant women, avoiding the use of boluses (doses >25,000 IU).

n **Bibliography**

1. Holick MF. Vitamin D: the underappreciated D-lightful hormone that is important for skeletal and cellular health. Curr Opin Endocrinol Diabetes 2002; 9: 87-98.
2. Holick MF. Vitamin D: photobiology, metabolism, mechanism of action, and clinical applications. In: Favus MJ, Editor. Primer on the metabolic bone diseases and disorders of mineral metabolism. 4th ed. Philadelphia: Lippincott Williams & Wilkins, 1999: 92-98.
3. Holick MF, Matsuoka LY, Wortsman J. Age, vitamin D, and solar ultraviolet radiation. Lancet 1989; 2: 1104-5.
4. Maggio D, Cherubini A, Lauretani F, Russo RC, Bartali B, Pierandrei M, et al. 25(OH) D serum levels decline with age earlier in women than in men and less efficiently prevent compensatory hyperparathyroidism in older adults. J Gerontol A Biol Sci Med Sci 2005; 60: 1414-9.
5. Webb AR, Pilbeam C, Hanafin N, Holick MF. An evaluation of the relative contribu-tions of exposure to sunlight and of diet to the circulating concentrations of 25-hydroxyvitamin D in an elderly nursing home population in Boston. Am J Clin Nutr 1990; 51: 1075-81.
6. Petta S, Cammà C, Scazzone C, Tripodo C, Di Marco V, Bono A, et al. Low vitamin D serum level is related to severe fibrosis and low responsiveness to interferon-based therapy in genotype 1 chronic hepatitis C. Hepatology. 2010; 51: 1158-67.
7. Reichel H, Koeffler HP, Norman AW. The role of the vitamin D endocrine system in health and disease. N Engl J Med 1989; 320: 981-91.
8. Smith JE, Goodman DS. The turnover and transport of vitamin D and of a polar metabolite with the properties of 25-hydroxycholecalciferol in human plasma. J Clin Invest 1971; 50: 2159-67.
9. Haddad JG, Rojanasathit S. Acute administration of 25-hydroxycholecalciferol in man. J Clin Endocrinol Metab 1976; 42: 284-90.
10. Russo S, Carlucci L, Cipriani C, Ragno A, Piemonte S, Del Fiacco R, et al. Metabolic changes following 500 mcg monthly administration of calcidiol: a study in normal females. Calcif Tissue Int 2011; 89: 252-7.
11. Jones G. Why dialysis patients need combination therapy with both cholecalciferol and a calcitriol analogs. Semin Dial 2010; 23: 239-43.
12. Norman AW, Nemere I, Zhou LX, Bishop JE, Lowe KE, Maiyar AC. 1,25(OH)2 D3 , a steroid hormone that produces biologic effects via both genomic and nongenomic pathways. J Steroid Biochem Mol Biol 1992; 41: 231-40.
13. Schott GD, Wills MR. Muscle weakness in osteomalacia. Lancet 1976; 1: 626-7.
14. Boland R. Role of vitamin D in skeletal muscle function. Endocr Rev 1986; 7: 434-48.
15. Mowè M, Haug E, Bøhmer T. Low serum calcidiol concentration in older adults with reduced muscolar function. J Am Geriatr Soc 1999; 47: 220-6.
16. Bischoff HA, Stahelin HB, Urscheler N, Ehrsam R, Vonthein R, Perrig-Chiello P, et al. Muscle strength in the elderly: its relation to vitamin D metabolites. Arch Phys Med Rehabil 1999; 80: 54-8.
17. Pfeifer M, Begerow B, Minne HW, Schlotthauer T, Pospeschill M, Scholz M, et al. Vitamin D status, trunk muscle strength, body sway, falls, and fractures among 237 postmenopausal women with osteoporosis. Exp Clin Endocrinol Diabetes 2001; 109: 87-92.
18. Zamboni M, Zoico E, Tosoni P, Zivelonghi A, Bortolani A, Maggi S, et al. Relation between vitamin D, physical performance, and disability in elderly persons. J Gerontol A Biol Sci Med Sci 2002; 57: M7-11.
19. Dhesi JK, Bearne LM, Moniz C, Hurley MV, Jackson SH, Swift CG, et al. Neu-romuscular and psychomotor function in elderly subjects who fall and the relation-ship with vitamin D status. J Bone Miner Res 2002; 17: 891-7.
20. Visser M, Deeg DJ, Lips P. Longitudinal Aging Study Amsterdam. Low vitamin D and high parathyroid hormone levels as determinants of loss of muscle strength and muscle mass (sarcopenia): the Longitudinal Aging Study Amsterdam. J Clin Endocrinol Metab 2003; 88: 5766-72.
21. Venning G. Recent developments in vitamin D deficiency and muscle weakness among elderly people. BMJ 2005; 330: 524-6.
22. Gerdhem P, Ringsberg KAM, Obrant KJ, Akesson K. Association between 25-hydroxy vitamin D levels, physical activity, muscle strength and fractures in the prospective population-based OPRA Study of Elderly Women. Osteoporos Int 2005; 16: 1425-31.
23. Houston DK, Cesari M, Ferrucci L, Cherubini A, Maggio D, Bartali B, et al. Association between vitamin D status and physical performance: the InCHIANTI study. J Gerontol A Biol Sci Med Sci. 2007; 62:440-6.
24. Summary of the Updated American Geriatric Society/British Geriatrics Society Clinical Practice Guidelines for Prevention of Falls in Older Persons. J Am Geriatr Soc 2011; 59: 148-57.
25. Yoshikawa S, Nakamura T, Tanabe H, Ima-

mura T. Osteomalacic myopathy. Endocrinol Jpn 1979; 26: 65-72.
26. Bischoff HA, Stähelin HB, Dick W, Akos R, Knecht M, Salis C, et al. Effects of vitamin D and calcium supplementation on falls: a randomized controlled trial. J Bone Miner Res 2003; 18: 343-51.
27. Bischoff-Ferrari HA, Dawson-Hughes B, Willett WC, Staehelin HB, Bazemore MG, Zee RY, et al. Effect of vitamin D on falls. JAMA 2004; 291: 1999-2006.
28. Bischoff-Ferrari HA, Dawson-Hughes B, Staehelin HB, Orav JE, Stuck AE, Theiler R, et al. Fall prevention with supplemental and active forms of vitamin D: a meta-analysis of randomised controlled trials. BMJ 2009; 339: b3692.
29. Dukas L, Bischoff HA, Lindpaintner LS, Schacht E, Birkner-Binder D, Damm TN, et al. Alfacalcidol reduces the number of fallers in a community-dwelling elderly population with a minimum calcium intake of more than 500 mg daily. J Am Geriatr Soc 2004; 52: 230-6.
30. Kalyani RR, Stein B, Valiyil R, Manno R, Maynard JW, Crews DC. Vitamin D treatment for the prevention of falls in older adults: systematic review and meta-analysis. J Am Geriatr Soc. 2010; 58: 1299-310.
31. McAlindon TE, Felson DT, Zhang Y, Hannan MT, Aliabadi P, Weissman B, et al. Relation of dietary intake and serum levels of vitamin D to progression of osteoarthritis of the knee among partecipants in the Framingham Study. Ann Intern Med 1996; 125: 353-9.
32. Glowacki J, Hurwitz S, Thornhill TS, Kelly M, LeBoff MS. Osteoporosis and vitamin D deficiency among postmenopausal women with osteoarthritis undergoing total hip arthroplasty. J Bone Joint Surg 2003; 85: 2371-7.
33. Merlino LA, Curtis J, Mikuls TR, Cerhan JR, Criswell LA, Saag KG. Vitamin D in-take is inversely associated with rheumatoid arthritis: results from the Iowa Women's Health Study. Arthritis Rheum 2004; 50: 72-7.
34. Munger KL, Zhang SM, O'Reilly E, Hernan MA, Olek MJ, Willett WC, et al. Vitamin D intake and incidence of multiple sclerosis. Neurology 2004; 62: 60-5.
35. Grant WB, Garland CF. A critical review of studies on vitamin D in relation to colorectal cancer. Nutr Cancer 2004; 48: 115-23.
36. Holick MF. Sunlight and vitamin D for bone health and prevention of autoimmune diseases, cancers, and cardiovascular disease. Am J Clin Nutr 2004; 80: 1678S-88S.
37. Holick MF. Vitamin D: importance in the prevention of cancers, type 1 diabetes, heart disease, and osteoporosis. Am J Clin Nutr 2004; 79: 362-71.
38. Holick MF. Clinical efficacy of 1,25- dihydroxyvitamin D3 and analogues in the treatment of psoriasis. Retinoids 1998; 14: 12-7.
39. Guyton KZ, Kensler YW, Posner GH. Cancer chemoprevention using natural vitamin D and synthetic analogs. Annu Rev Pharmacol Toxicol 2001; 41: 421-42.
40. Li Y, Kong J, Wei M, Chen ZF, Liu S, Cao LP. 1,25- dihydroxyvitamin D3 is a negative endocrine regulator of the renin-angiotensin system. J Clin Invest 2002; 110: 229-38.
41. Hypponen E, Laara E, Jarvelin MR, Virtanen SM. Intake of vitamin D and risk of type 1 diabetes: a birth-cohort study. Lancet 2001; 358: 1500-3.
42. Binkley N, Krueger DC, Morgan S, Wiebe D. Current status of clinical 25-hydroxy-vitamin D measurement: an assessment of between-laboratory agreement. Clin Chim Acta 2010; 14: 1976-82.
43. Heaney RP. Functional indices of vitamin D status and ramifications of vitamin D deficiency. Am J Clin Nutr 2004; 80 (Suppl.) 1706S-9S.
44. Chapuy MC, Preziosi P, Maamer M, Arnaud S, Galan P, Hercberg S, et al. Prevalence of vitamin D insufficiency in an adult normal population. Osteoporos Int 1997; 7: 439-43.
45. Durazo-Arvizu RA, Dawson-Hughes B, Sempos CT, Yetley EA, Looker AC, Cao G, et al. Three-phase model harmonizes estimates of the maximal suppression of parathyroid hormone by 25-hydroxyvitamin D in persons 65 years of age and older. J Nutr 2010; 140: 595-9.
46. Adami S, Viapiana O, Gatti D, Idolazzi L, Rossini M. Relationship between serum parathyroid hormone, vitamin D sufficiency, age, and calcium intake. Bone 2008; 42: 267-70.
47. Carnevale V, Nieddu L, Romagnoli E, Battista C, Mascia ML, Chiodini I, et al. Regulation of PTH secretion by 25-hydroxyvitamin D and ionized calcium depends on vitamin D status: a study in a large cohort of healthy subjects. Bone 2010; 47: 626-30.
48. Bischoff- Ferrari HA, Dietrich T, Orav EJ, Dawson-Hughes B. Positive association between 25-hydroxy vitamin D levels and bone mineral density: a population-based study of younger and older adults. Am J Med 2004; 116: 634-9.
49. Bischoff-Ferrari HA, Kiel DP, Dawson-Hughes B, Orav JE, Li R, Spiegelman D, et al. Dietary calcium and serum 25-hydroxy-vitamin D status in relation to BMD among U.S. adults. J Bone Miner Res 2009; 24: 935-42.
50. Bischoff-Ferrari HA, Giovannucci E, Wil-lett WC, Dietrich T, Dawson-Hughes B. Estimation of optimal serum concentrations

of 25-hydroxyvitamin D for multiple health outcomes. Am J Clin Nutr 2006; 84:18-28.
51. Ross AC, Taylor CL, Yaktine AL, Del Valle HB, Editors; Committee to Review Dietary Reference Intakes for Vitamin D and Calcium; Institute of Medicine. ISBN: 0-309-16395-1, 482 pages. National Academics Press 2010.
52. Priemel M, von Domarus C, Klatte TO, Kessler S, Schlie J, Meier S, et al. Bone mineralization defects and vitamin D deficiency: histomorphometric analysis of iliac crest bone biopsies and circulating 25-hydroxyvitamin D in 675 patients. J Bone Miner Res 2010; 25: 305-12.
53. Flicker L, MacInnis RJ, Stein MS, Scherer SC, Mead KE, Nowson CA, et al. Should older people in residential care receive vitamin D to prevent falls? Results of a randomized trial. J Am Geriatr Soc 2005; 53: 1881-8.
54. Grant AM, Avenell A, Campbell MK, McDonald AM, MacLennan GS, McPherson GC, et al; RECORD Trial Group. Oral vitamin D3 and calcium for secondary prevention of low-trauma fractures in elderly people (Randomised Evaluation of Calcium Or vitamin D, RECORD): a randomised placebo-controlled trial. Lancet 2005; 365: 1621-8.
55. Papadimitropoulos E, Wells G, Shea B, Gillespie W, Weaver B, Zytaruk N, et al; Osteoporosis Methodology Group and The Osteoporosis Research Advisory Group. Meta-analysis of the efficacy of vitamin D treatment in preventing osteoporosis in postmenopausal women. Endocr Rev 2002; 23: 560-9.
56. Porthouse J, Cockayne S, King C, Saxon L, Steele E, Aspray T, et al. Randomised controlled trial of calcium and supplementation with cholecalciferol (vitamin D3) for prevention of fractures in primary care. BMJ 2005; 330: 1003-9.
57. Trivedi DP, Doll R, Khaw KT. Effect of four monthly oral vitamin D3 (cholecalciferol) supplementation on fractures and mortality in men and women living in the community: randomised double blind controlled trial. BMJ 2003; 326: 469-72.
58. LeBoff MS, Kohlmeier L, Hurwitz S, Franklin J, Wright J, Glowacki J. Occult vitamin D deficiency in postmenopausal US women with acute hip fracture. JAMA 1999; 281: 1505-11.
59. Nuti R, Martini G, Valenti R, Gambera D, Gennari L, Salvadori S, et al. Vitamin D status and bone turnover in women with acute hip fracture. Clin Orthop Relat Res 2004; 422: 208-13.
60. Melhus H, Snellman G, Gedeborg R, Byberg L, Berglund L, Mallmin H, et al. Plasma 25-hydroxyvitamin D levels and fracture risk in a community-based cohort of elderly men in Sweden. J Clin Endocrinol Metab 2010; 95: 2637-45.
61. Lips P, Obrant KJ. The pathogenesis and treatment of hip fractures. Osteoporos Int 1991: 1: 218-31.
62. Cauley JA, Lacroix AZ, Wu L, Horwitz M, Danielson ME, Bauer DC, et al. Serum 25-hydroxyvitamin D concentrations and risk for hip fractures. Ann Intern Med 2008; 49: 242-50.
63. Looker AC, Mussolino ME. Serum 25-hydroxyvitamin D and hip fracture risk in older U.S. white adults. J Bone Miner Res 2008; 23: 143-50. 64.
van Schoor NM, Visser M, Pluijm SM, Kuchuk N, Smit JH, Lips P. Vitamin D deficiency as a risk factor for osteoporotic fractures. Bone 2008; 42: 260-6.
65. Cauley JA, Parimi N, Ensrud KE, Bauer DC, Cawthon PM, Cummings SR, et al; Osteoporotic Fractures in Men (MrOS) Research Group. Serum 25 hydroxyvitamin D and the risk of hip and non-spine fractures in older men. J Bone Miner Res 2010; 25: 545-53.
66. Heaney RP, Holick MF. Why the IOM recommendations for Vitamin D are deficient. J Bone Miner Res 2011; 26: 455-57.
67. Bischoff-Ferrari HA, Willett WC, Wong JB, Stuck AE, Staehelin HB, Orav EJ, et al. Prevention of nonvertebral fractures with oral vitamin D and dose dependency: a meta-analysis of randomized controlled trials. Arch Intern Med 2009; 169: 551-61.
68. Steingrimsdottir L, Gunnarsson O, Indridason OS, Franzson L, Sigurdsson G. Relationship between serum parathyroid hormone levels, vitamin D sufficiency, and calcium intake. JAMA 2005; 294: 2336-41.
69. Ebeling PR, Sandgren ME, Di Magno EP, Lane AW, De Luca HF, Riggs BL. Evidence of an age-related decrease in intestinal responsiveness to vitamin D: relationship between serum 1,25- dihydroxyvitamin D3 and intestinal vitamin D receptor concentration in normal women. J Clin Endocrinol Metab 1992; 75: 176-82.
70. Slovik DM, Adams JS, Neer RM, Holick MF, Potts JT. Deficient production of 1,25-dihydroxyvitamin D in elderly osteoporotic patients. N Engl J Med 1981; 305: 372-4.
71. Riggs BL, Hamstra A, De Luca HF. Assessment of 25 hydroxyvitamin D 1ÿ-hydroxylase reserve in postmenopausal osteoporosis by administration of parathyroid extract. J Clin Endocrinol Metab 1981; 53: 833-5.
72. Isaia G, Giorgino R, Rini GB, Bevilacqua M, Maugeri D, Adami S. Prevalence of

hypovitaminosis D in elderly women in Italy: clinical consequences and risk factors. Osteoporosis Int 2003; 14: 577-82.
73. Rossini M, Mattarei A, Braga V, Viapiana O, Zambarda C, Benini C, et al. Risk factors for hip fracture in elderly persons. Rheumatism. 2010; 62: 273-82.
74. Holick MF. Vitamin D deficiency. N Engl J With 2007; 357: 266–81.
75. Bilke DD. Vitamin D: newly discovered actions require reconsideration of physiologic requirements. Trends Endocrinol Metab 2010; 21: 375-84.
76. Gallagher JC, Sai AJ. Vitamin D insufficiency, deficiency, and bone health. J Clin Endocrinol Metab 2010; 95: 2630-3.
77. Sai AJ, Walters RW, Fang X, Gallagher JC. Relationship between vitamin D, par-athyroid hormone, and bone health. J Clin Endocrinol Metab 2011; 96: E436-46.
78. Rossini M, Perbellini S, Lazzarin M, Adami S, Bertoldo F, Lo Cascio V. Incidence of hypovitaminosis D in Northern Italy. It J Min Elect Metab 1990; 4:13-7.
79. Van der Wielen RPJ, Lowik MRH, Van den Berg H, De Groot LCPGM, Haller J, Moreiras O, et al. Serum vitamin D concentrations among elderly people in Europe The Lancet, 1995; 345: 207-10.
80. Bettica P, Bevilacqua M, Vago T, Norbiato G. High prevalence of hypovitaminosis D among free-living postmenopausal women referred to an osteoporosis outpatient clinic in northern Italy for initial screening. Oste-oporos Int 1999; 9: 226-9.
81. Carnevale V, Modoni S, Pileri M, Di Giorgio A, Chiodini I, Minisola S, et al. Longitudinal evaluation of vitamin D status in healthy subjects from southern Italy: seasonal and gender differences. Osteoporosis Int 2001; 12: 1026-30.
82. Lips P, Duong T, Oleksik A, Black D, Cummings S, Cox D, et al. A global study of vitamin D status and parathyroid function in postmenopausal women with osteoporosis: baseline data from the Multiple Outcomes of Raloxifene Evaluation clinical trial. J Clin Endocrinol Metab 2001; 86: 1212-21.
83. Romagnoli E, Caravella P, Scarnecchia L, Martinez P, Minisola S. Hypovitaminosis D in an Italian population of healthy subjects and hospitalized patients. Br J Nutr 1999; 81: 133-7.
84. Giusti A, Barone A, Razzano M, Pizzonia M, Oliveri M, Palummeri E, et al. High prevalence of secondary hyperparathyroidism due to hypovitaminosis D in hospitalized elderly with and without hip fracture. J Endocrinol Invest 2006; 29: 809-13.
85. Passeri G, Pini G, Troiano L, Vescovini R, Sansoni P, Passeri M, et al. Low vitamin D status, high bone turnover, and bone fractures in centenarians. J Clin Endocrinol Metab 2003; 88: 5109-15.
86. Muscarella S, Filabozzi P, D'Amico G, Mascia ML, Annese MA, Scillitani A, et al. Vitamin D status in inpatients admitted to an internal medicine department. Horm Res 2006; 66: 216-20.
87. Kuchuk NO, van Schoor NM, Pluijm SM, Chines A, Lips P. Vitamin D status, parathyroid function, bone turnover, and BMD in postmenopausal women with osteoporo-sis: global perspective. J Bone Miner Res. 2009; 24: 693-701.
88. Chapuy MC, Arlot ME, Duboeuf F, Brun J, Crouzet B, Arnaud S, et al. Vitamin D3 and calcium to prevent hip fractures in the elderly women. N Engl J Med 1992; 327: 1637-42.
89. Chapuy MC, Arlot ME, Delmas PD, Meuni-er PJ. Effect of calcium and cholecalciferol treatment for three years on hip fractures in elderly women. BMJ 1994; 308: 1081-2.
90. Rossini M, Alberti V, Flor L, Masiero L, Giannini S, Gatti D, et al. Effect of oral vitamin D2 yearly bolus on hip fracture risk in elderly women: a community primary prevention study. Aging Clin Exp Res 2004; 16: 432-6.
91. Giusti A, Barone A, Razzano M, Oliveri M, Pizzonia M, Palummeri E, et al. Persistence with calcium and vitamin D in elderly patients after hip fracture. J Bone Miner Metab 2009; 27: 95-100.
92. Carnevale V, Nieddu L, Romagnoli E, Bona E, Piemonte S, Scillitani A, et al. Osteoporosis intervention in ambulatory patients with previous hip fracture: a multicentric, nationwide Italian survey. Osteoporos Int 2006; 17: 478-83.
93. Adami S, Isaia G, Luisetto G, Minisola S, Sinigaglia L, Gentilella R, et al; ICARO Study Group. Fracture incidence and characterization in patients on osteoporosis treatment: the ICARO study. J Bone Miner Res 2006; 21: 1565-70.
94. Adami S, Isaia G, Luisetto G, Minisola S, Sinigaglia L, Silvestri S, et al; ICARO Study Group. Osteoporosis treatment and fracture incidence: the ICARO longitudinal study. Osteoporosis Int 2008; 19: 1219-23.
95. Adami S, Giannini S, Bianchi G, Sinigaglia L, Di Munno O, Fiore CE, et al. Vitamin D status and response to treatment in postmenopausal osteoporosis. Osteoporos Int 2009; 20: 239-44.
96. Adami S, Bertoldo F, Braga V, Fracassi E, Gatti D, Gandolini G, et al. 25-hydroxy vitamin D levels in healthy premenopausal women: association with bone turnover markers and bone mineral density. Bone 2009; 45: 423-6.
97. Heaney RP, Davies KM, Chen TC, Holick

MF, Barger-Lux MJ. Human serum 25-hydroxycholecalciferol response to extended oral dosing with cholecalciferol. Am J Clin Nutr 2003; 77: 204-10.
98. Giusti A, Barone A, Pioli G, Girasole G, Razzano M, Pizzonia M, et al. Heterogeneity in serum 25-hydroxy-vitamin D response to cholecalciferol in elderly women with secondary hyperparathyroidism and vitamin D deficiency. J Am Geriatr Soc. 2010; 58: 1489-95.
99. Wortsman J, Matsuoka LY, Chen TC, Lu Z, Holick MF. Decreased bioavailability of vitamin D in obesity. Am J Clin Nutr 2000; 72: 690-3.
100. Harris SS, Dawson-Hughes B. The association of oral contraceptive use with plasma 25-hydroxyvitamin D levels. J Am Coll Nutr 1998; 17: 282-4.
101. Harris SS, Dawson-Hughes B. Plasma vitamin D and 25OHD responses of young and old men to supplementation with vitamin D3. J Am Coll Nutr 2002; 21: 357-62.
102. Boonen S, Lips P, Bouillon R, Bischoff-Ferrari HA, Vanderschueren D, Haentjens P. Need for additional calcium to reduce the risk of hip fracture with vitamin d supplementation: evidence from a comparative metaanalysis of randomized controlled trials. J Clin Endocrinol Metab 2007; 92: 1415-23.
103. Premaor MO, Scalco R, da Silva MJ, Froehlich PE, Furlanetto TW. The effect of a single dose versus a daily dose of cholecal-ciferol on the serum 25-hydroxycholecalcif-erol and parathyroid hormone levels in the elderly with secondary hyperparathyroidism living in a low-income housing unit. J Bone Miner Metab 2008; 26: 603-8.
104. Leidig-Bruckner G, Roth HJ, Bruckner T, Lorenz A, Raue F, Frank-Raue K. Are commonly recommended dosages for vitamin D supplementation too low? Vitamin D status and effects of supplementation on serum 25-hydroxyvitamin D levels—an observa-tional study during clinical practice condi-tions. Osteoporos Int 2011; 22: 231-40.
105. Bischoff-Ferrari HA, Shao A, Dawson-Hughes B, Hathcock J, Giovannucci E, Willett WC. Benefit-risk assessment of vitamin D supplementation. Osteoporos Int 2010; 21: 1121-32.
106. Vieth R. Why the optimal requirement for vitamin D3 is probably much higher than what is officially recommended for adults. J Ster Biochem Mol Bio 2004; 89-90: 575-9.
107. Bailey RL, Dodd KW, Goldman JA, Gahche JJ, Dwyer JT, Moshfegh AJ, et al. Estimation of total usual calcium and vitamin D intake in the United States. J Nutr 2010; 140: 817-22.
108. Calvo MS, Whiting SJ. Determinants of vitamin D intake. From: Nutrition and Health: vitamin D. Holick MF ed. Springer Science + Business Media, LLC 2010.
109. Whiting SJ, Green TJ, Calvo MS. Vitamin D intakes in North America and Asia-Pacific countries are not sufficient to prevent vitamin D insufficiency. J Steroid Biochem Mol Biol 2007; 103: 626-30.
110. Rossini M, Viapiana O, Gatti D, James G, Girardello S, Adami S. Correction of hypovitaminosis D in the long term: comparison between different methods of vitamin D administration in clinical practice. Minerva Med 2005; 96 (Suppl 1): 1-7.
111. Chel V, Wijnhoven HA, Smit JH, Ooms M, Lips P. Efficacy of different doses and time intervals of oral vitamin D supplementation with or without calcium in elderly nursing home residents. Osteoporos Int 2008; 19: 663-71.
112. Sanders KM, Stuart AL, Williamson EJ, Simpson JA, Kotowicz MA, Young D, et al. Annual high-dose oral vitamin D and falls and fractures in older women: a randomized controlled trial. JAMA 2010; 303: 1815-22.
113. Gartner LM, Greer FR; Section on Breastfeeding and Committee on Nutrition. American Academy of Pediatrics. Prevention of rickets and vitamin D deficiency: new guidelines for vitamin D intake. Pediatrics 2003; 111: 908-10.
114. Standing Committee on the Scientific Evaluation of Dietary Reference Intakes, Food and Nutrition Board, Institute of Medicine, National Academy of Sciences. Dietary reference intakes for calcium, phosphorus, magnesium, vitamin D, and fluoride. Washington, DC: National Academy Press 2000.
115. Bacon CJ, Gamble GD, Horne AM, Scott MA, Reid IR. High-dose oral vitamin D3 supplementation in the elderly. Osteoporos Int 2009; 20: 1407-15.
116. Romagnoli E, Mascia ML, Cipriani C, Fassino V, Mazzei F, D'Erasmo E, et al. Short and long-term variations in serum calciotropic hormones after a single very large dose of ergocalciferol (vitamin D2) or cholecalciferol (vitamin D3) in the elderly. J Clin Endocrinol Metab 2008; 93: 3015-20.
117. Heaney RP, Recker RR, Grote J, Horst RL, Armas LAG. Vitamin D3 is more potent than vitamin D2 in humans. J Clin Endocrinol Metab 2011; 96: E447-52.
118. Ilahi M, Armas LAG, Heaney RP. Pharmacokinetics of a single, large dose of vitamin D3. Am J Clin Nutr 2008; 87: 688-91.
119. Cipriani C, Romagnoli E, Scillitani A, Chiodini I, Clerico R, Carnevale V, et al. Effect of a single oral dose of 600,000 IU of cholecalciferol on serum calciotropic hormones in young subjects with vitamin D deficiency: a prospective intervention study.

J Clin Endocrinol Metab 2010; 95: 4771-7.
120. Hollis BW, Wagner CL. Vitamin D requirements during lactation: high-dose maternal supplementation as therapy to prevent hypovitaminosis D for both the mother and the nursing infant. Am J Clin Nutr 2004; 80 (Suppl): 1752S- 8S.
121. Federal Food, Drug, and Cosmetic Act of 1980. 21 USC §321.
[PubMed] 122. Antia AU, Wiltse HE, Rowe RD, Pitt EL, Levin S, Ottesen OE, et al. Pathogenesis of the supravalvular aortic stenosis syndrome. J Pediatr 1967; 71: 431-41.
123. Seelig M. Vitamin D and cardiovascular, renal and brain damage in infancy and child-hood. Ann N Y Acad Sci 1969; 147: 537-82.
124. Aravena T, Castillo S, Carrasco X, Mena I, López J, Rojas JP, et al. Williams syndrome: clinical, cytogenetic, neurophysiological and neuroanatomic study. Rev Med Child 2002; 130: 631-7.
125. Specker B. Vitamin D requirements dur-ing pregnancy. Am J Clin Nutr 2004; 80 (6 suppl): 1740S-7S.
126. Rosen CJ. Vitamin D insufficiency. N Engl J Med 2011; 364: 248-54.