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Resurrection of vitamin D deficiency and rickets

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The epidemic scourge of rickets in the 19th century was caused by vitamin D deficiency due to inadequate sun exposure and resulted in growth retardation, muscle weakness, skeletal deformities, hypocalcemia, tetany, and seizures. The encouragement of sensible sun exposure and the fortification of milk with vitamin D resulted in almost complete eradication of the disease. Vitamin D (where D represents D_2 or D_3) is biologically inert and metabolized in the liver to 25-hydroxyvitamin D [25(OH)D], the major circulating form of vitamin D that is used to determine vitamin D status. 25(OH)D is activated in the kidneys to 1,25-dihydroxyvitamin D [$1,25(OH)_2D$], which regulates calcium, phosphorus, and bone metabolism. Vitamin D deficiency has again become an epidemic in children, and rickets has become a global health issue. In addition to vitamin D deficiency, calcium deficiency and acquired and inherited disorders of vitamin D, calcium, and phosphorus metabolism cause rickets. This review summarizes the role of vitamin D in the prevention of rickets and its importance in the overall health and welfare of infants and children.

Historical perspective

In the mid-1600s, most children who lived in the crowded and polluted industrialized cities of northern Europe developed a severe bone-deforming disease that was characterized by growth retardation, enlargement of the epiphyses of the long bones, deformities of the legs, bending of the spine, knobby projections of the ribcage, and weak and toneless muscles (1, 2) (Figure 1). In the latter part of the 19th century, autopsy studies done in Boston and Leiden, The Netherlands, showed that 80–90% of children had rickets.

In 1822, Sniadecki (3) recognized the importance of sun exposure for the prevention and cure of rickets. Palm (4) extended these observations in 1890 and promoted systemic use of sun baths to prevent rickets. In 1919, Huldschinski (5, 6) found that exposing children to radiation from a sun quartz lamp (mercury arc lamp) or carbon arc lamp for one hour 3 times a week was effective in treating rickets, as demonstrated by a marked increase in the mineralization of the skeleton, especially the ends of the long bones, evident in the child's x-ray (Figure 2). A similar group of children not exposed to UV radiation showed no cure or only a slight improvement (6). He concluded that exposure to UV radiation was an "infallible remedy" against all forms of rickets in children. Two years later, Hess and Unger (7) exposed 7 rachitic children in New York City to varying periods of sunshine and reported marked improvement in the rickets of each child as evidenced by calcification of the epiphyses.

In 1918, Mellanby et al. (8) prevented rickets in puppies with cod liver oil. McCollum et al. (9) called this new nutritional factor vitamin D. Hess and Weinstock (10) and Steenbock and Black (11) observed that UV irradiation of various foods and oils imparted antirachitic activity. This led to enhancement of the antirachitic activity of milk by exposing milk to UV radiation or feeding cows UV-irradiated yeast. Once vitamin D was structurally identified and chemically synthesized inexpensively from yeast, it was directly added to milk at a standard of 400 IU (1 IU = 25 ng) per quart (12, 13). It was thought that the vitamin D obtained from irradiated yeast was the same vitamin D that was produced in the skin. However, when it was observed that vitamin D from irradiated yeast had little antirachitic activity in chickens, whereas cod liver oil was effective, it was concluded that the vitamin D produced in the skin must be different (14). Vitamin D was isolated and identified from pig skin and shown to originate from 7-dehydrocholesterol (1, 2, 14). To distinguish the two vitamin Ds, the vitamin D from yeast was called vitamin D_2 and the one from pig and human skin vitamin D_3 (1, 2).

Photobiology of vitamin D_3

Sunlight was recommended as a therapeutic method to prevent rickets in infants, and a detailed description was published in the United States Children's Bureau Folder in 1931 (13, 14). It was recognized that in the temperate zone, sunlight was feeble in its antirachitic properties in the winter, and thus, it was recommended that children be exposed to UV radiation from a mercury arc or carbon arc lamp in the winter (5, 6, 13, 14) (Figure 2). During exposure to sunlight, the ultraviolet B (UVB) radiation (290–315 nm) is absorbed by 7-dehydrocholesterol in the skin to form previtamin D_3 (1, 15). Previtamin D_3 (1, 15) is inherently unstable and rapidly converts by a temperature-dependent process to vitamin D_3 (Figure 3). Once formed, it is ejected out of the skin cell into the extracellular space, where it is drawn into the dermal capillary bed by the vitamin D–binding protein (DBP) (1).

The efficiency of vitamin D_3 synthesis in the skin is dependent on the number of UVB photons that penetrate into the epidermis. An increase in skin melanin pigmentation (16) and the topical application of a sunscreen (17), both of which efficiently absorb UVB photons, can markedly diminish by more than 90% the production of vitamin D_3. Excessive exposure to sunlight cannot cause vitamin D intoxication because sunlight destroys any excess vitamin D_3 produced in the skin (18, 19). Most UVB photons from the sun are absorbed by stratospheric ozone. An increase in the sun's zenith angle results in an increased path length for the UVB photons to travel, and this explains why at higher latitudes (above

Nonstandard abbreviations used: 1-OHase, 25-hydroxyvitamin D-1α-hydroxylase; $1,25(OH)_2D$, 1,25-dihydroxyvitamin D; 25(OH)D, 25-hydroxyvitamin D; DBP, vitamin D–binding protein; FGF23, fibroblast growth factor 23; PHEX, phosphate-regulating endopeptidase homolog, X-linked; PTH, parathyroid hormone; RANKL, receptor activator of NF-κB ligand; RXR, retinoic acid X receptor; SPF, sun protection factor; UVB, ultraviolet B; VDR, vitamin D receptor; VDRE, vitamin D–responsive element.

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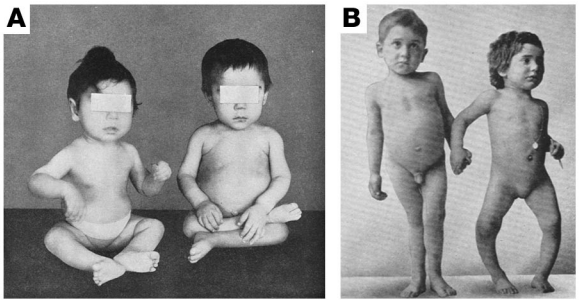

Figure 1
Skeletal deformities observed in rickets. (**A**) Photograph from the 1930s of a sister (left) and brother (right), aged 10 months and 2.5 years, respectively, showing enlargement of the ends of the bones at the wrist, carpopedal spasm, and a typical "Taylorwise" posture of rickets. (**B**) The same brother and sister 4 years later, with classic knock-knees and bow legs, growth retardation, and other skeletal deformities. Reproduced from ref. 14.

~35° latitude), very little, if any, vitamin D_3 is produced in the skin from November through March (19, 20).

Vitamin D metabolism and its role in calcium and phosphorus metabolism

Vitamin D_2 and vitamin D_3 (D represents either D_2 or D_3) derived from supplements, fortified foods, and fish ingested from the diet (Table 1) are incorporated into chylomicrons and absorbed into the lymphatic system. From here they enter the circulation, where they are bound to the DBP and lipoproteins (1, 20–22). Vitamin D is released from DBP to the liver and undergoes a hydroxylation on C-25 by the vitamin D-25-hydroxylases (25-OHase; also known as CYP27A1, CYP3A4, CYP2R1, CYP2J3) to 25-hydroxyvitamin D [25(OH)D] (20–22) (Figure 3). 25(OH)D is the major circulating form of vitamin D that is measured to determine a person's vitamin D status because it has a half-life in the circulation of 2 weeks and it correlates with secondary hyperparathyroidism, rickets, and osteomalacia (20, 22–24). 25(OH)D is bound to DBP, and this complex binds to megalin on the plasma membrane of the renal tubule cell and is transported into the cell (20, 22). Once inside, 25(OH)D is released and is converted in the mitochondria by the 25-hydroxyvitamin D-1α-hydroxylase [1-OHase; also known as CYP27B1] to form 1,25-dihydroxyvitamin D [$1,25(OH)_2D$]. $1,25(OH)_2D$ is the biologically active form of vitamin D responsible for maintaining calcium and phosphorus homeostasis. It accomplishes this by interacting with its nuclear receptor, the vitamin D receptor (VDR) in the small intestinal cells (22, 25). The $1,25(OH)_2D$–VDR structure complexes with retinoic acid X receptor (RXR) in the nucleus. The $1,25(OH)_2D$-VDR-RXR complex binds to the vitamin D–responsive element (VDRE) for the epithelial calcium channel (22, 25). The increased expression of the calcium channel permits more calcium to enter the cell, where the vitamin D–dependent calcium-binding protein calbindin 9K helps calcium's translocation into the bloodstream. $1,25(OH)_2D$ also enhances phosphorus absorption in the small intestine (1, 22, 25).

When dietary calcium is inadequate, vitamin D helps maintain calcium homeostasis by interacting with the VDR in osteoblasts to induce the expression of the plasma membrane protein receptor activator of NF-κB ligand (RANKL). The RANK on the plasma membrane of preosteoclasts binds RANKL, which induces the preosteoclast to become a mature osteoclast (20, 22, 26). The mature osteoclast releases hydrochloric acid and collagenases to dissolve bone and release its precious calcium and phosphorus stores into the circulation. Thus, the major physiologic function of vitamin D is to maintain serum calcium and phosphorus levels within the normal physiologic range to support most metabolic functions, neuromuscular transmission, and bone mineralization (1, 20, 22, 24) (Figure 3).

Vitamin D and calcium deficiencies as a cause of rickets

Vitamin D deficiency is the most common cause of rickets. Vitamin D deficiency prevents the efficient absorption of dietary calcium and phosphorus. In a vitamin D–deficient state, only 10–15% of dietary calcium and 50–60% of dietary phosphorus are absorbed. The poor absorption of calcium causes a decrease in serum-ionized calcium levels. This is immediately recognized by the calcium sensor in the parathyroid glands, resulting in an increase in the expression, synthesis, and secretion of parathyroid hormone (PTH) (1, 20, 22, 27). PTH conserves calcium by increasing tubular reabsorption of calcium in both the proximal and distal convoluted tubules. PTH, like $1,25(OH)_2D$, enhances the expression of RANKL on osteoblasts to increase the production of mature osteoclasts to mobilize calcium stores from the skeleton. PTH also decreases phosphorus reabsorption in the kidney, causing loss of phosphorus into the urine (Figure 4). The serum calcium level is usually normal in a vitamin D–deficient infant or child. However, the serum phosphorus level is low, and thus there is inadequate calcium-phosphorus product, which is necessary to mineralize the osteoid laid down by osteoblasts (1, 20, 22, 24, 28) (Figure 4). Thus, typically, infants with vitamin D–deficiency rickets have a normal serum calcium level, low normal or low fasting serum phosphorus levels, elevated alkaline phosphatase levels, and low 25(OH)D levels (<15 ng/ml) (23, 28–31) (Table 2). The secondary hyperparathyroidism stimulates the kidneys to produce $1,25(OH)_2D$, and thus, $1,25(OH)_2D$ levels are normal or often elevated, which is why the measurement of $1,25(OH)_2D$ is of no value in determining a state of vitamin D deficiency (24). Only when the calcium stores in the skeleton are totally depleted will the infant or child become hypocalcemic.

Vitamin D deficiency causes global poor mineralization of the skeleton. Clinical and radiological bone manifestations predominate in areas of rapid bone growth, including the long bone epiphyses and the costochondral junctions (5, 6, 12–14, 30–32). This is why rickets is mostly observed before 18 months of age, with maximum frequency between the ages of 4 and 12 months. Skeletal deformities are usually a result of long-standing rickets.

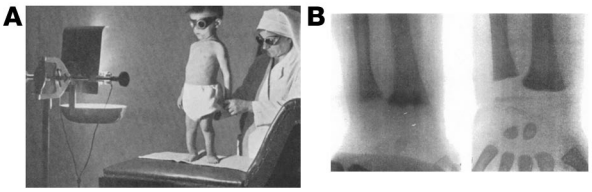

Figure 2
UV radiation therapy for rickets. (**A**) Photograph from the 1920s of a child with rickets being exposed to UV radiation. (**B**) Radiographs demonstrating florid rickets of the hand and wrist (left) and the same wrist and hand taken after treatment with 1 hour UV radiation 2 times a week for 8 weeks. Note mineralization of the carpal bones and epiphyseal plates (right). Reproduced from ref. 126.

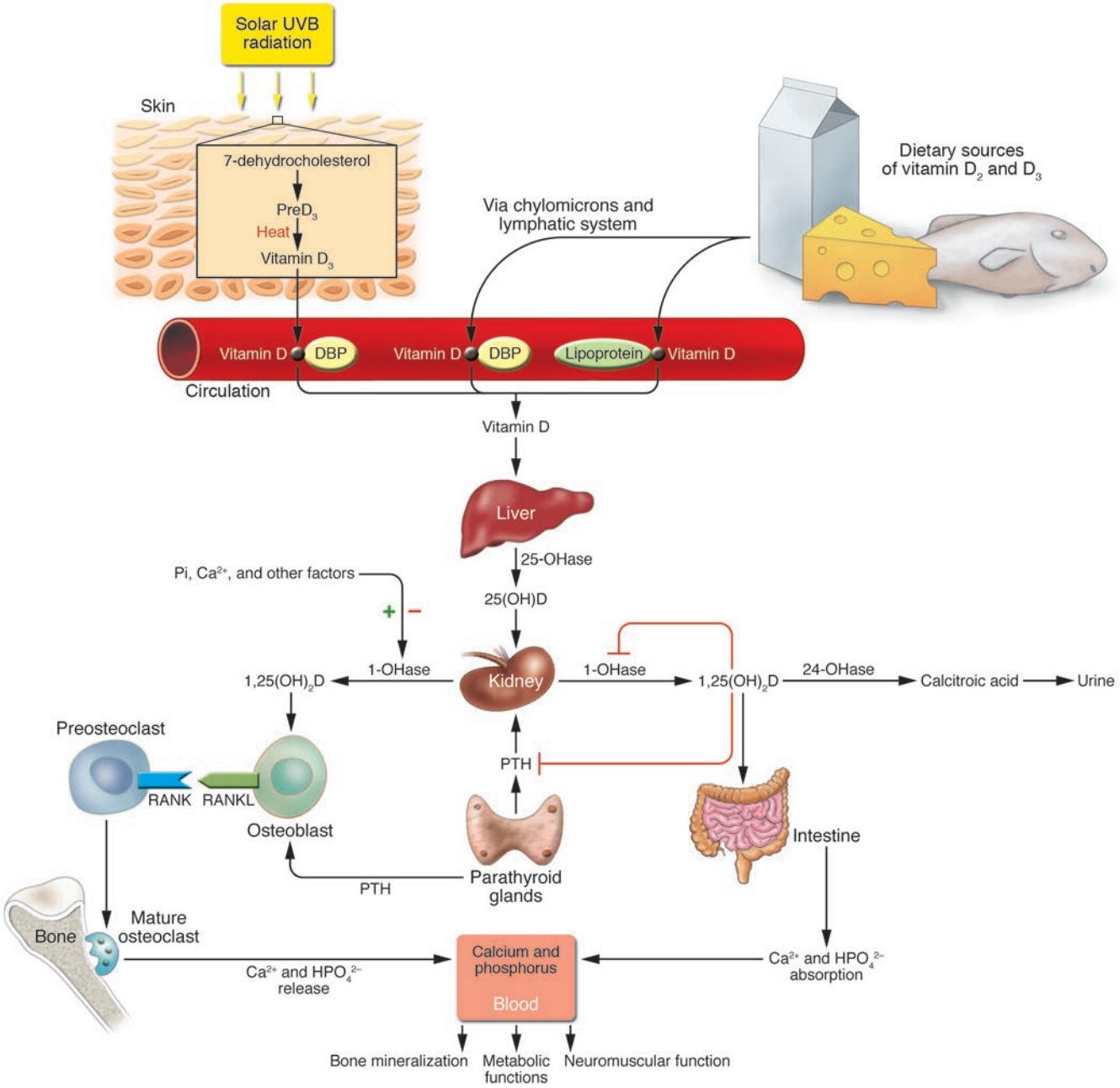

Figure 3
The photoproduction and metabolism of vitamin D and the various biologic effects of $1,25(OH)_2D$ on calcium, phosphorus, and bone metabolism. Vitamin D is either produced in the skin by exposure to UVB radiation or is ingested in the diet. Vitamin D (D represents vitamin D_2 or vitamin D_3) is converted by the vitamin D-25-hydroxylase (25-OHase) in the liver to 25(OH)D. 25(OH)D is converted in the kidneys by 1-OHase to $1,25(OH)_2D$. Once formed, $1,25(OH)_2D$ enhances intestinal calcium and phosphorus absorption and stimulates the expression of RANKL on the osteoblasts to interact with its receptor RANK on preosteoclasts to induce mature osteoclastic activity, which releases calcium and phosphorus (HPO_4^{2-}). In addition, $1,25(OH)_2D$ inhibits the renal 1-OHase and stimulates the expression of the renal 25(OH)D-24-hydroxylase (24-OHase). The induction of the 24-OHase results in the destruction of $1,25(OH)_2D$ into a water-soluble inactive metabolite calcitroic acid. $PreD_3$, previtamin D.

Hypertrophy of the costochondral junctions leads to beading and the classic rachitic rosary that progresses with involution of the ribs and protrusion of the sternum (pigeon chest) and recession of the costochondral junctions and traverse depressions causing Harrison's groove. Once the child begins to stand, gravity pushing on the lower limbs results in either inward (genu valgum) or outward (genu varum) tibial and femoral bowing. Muscle pull can also cause bone deformities in both upper and lower limbs even before the infant begins to walk. Muscle traction on the softened ribcage is responsible for the chest deformation, leading to pectus carinatum, thoracic asymmetry, and widening of the thoracic base. Softening of the occipital area (rachitic craniotabes), enlarged sutures and

Table 1
Dietary sources of vitamin D

Source	Vitamin D content
Fortified milk	100 IU/8 oz
Fortified orange juice	100 IU/8 oz
Infant formulas	100 IU/8 oz
Fortified yogurts	100 IU/8 oz
Fortified butter	56 IU/3.5 oz
Fortified margarine	429 IU/3.5 oz
Fortified cheeses	100 IU/3 oz
Fortified breakfast cereals	~100 IU/serving
Egg yolk	~20 IU/yolk
Shiitake mushrooms, fresh	100 IU/3.5 oz
Tuna, canned	236 IU/3.5 oz
Mackerel, canned	~250 IU/3.5 oz
Sardines, canned	~300 IU/3.5 oz
Salmon, canned	~300–600 IU/3.5 oz
Salmon, fresh	~400–500 IU/3.5 oz
Shiitake mushrooms, sun-dried	1,600 IU/3.5 oz
Drisdol (vitamin D_2) liquid	8,000 IU/cc
Cod liver oil	400 IU/tsp

fontanelles, delayed closing of fontanelles, and occipital or parietal flattening can be observed (5, 6, 12, 13, 30–32) (Figure 1). Tooth development is impaired, with delayed eruption, enamel hypoplasia, and early dental caries (12–14). The pelvic bone structure is flattened in rachitic children. Because of the high incidence of infant and maternal morbidity and mortality in rachitic women, children were often delivered by Caesarian section (1, 2).

Extraskeletal manifestations associated with hypocalcemia lead to tetany, seizures, laryngospasm, and hypocalcemic myocardiopathy and death (6, 12–14, 32). Often there is delayed motor development with hypotonia in the absence of hypocalcemia. Weakness of the thoracic muscles together with softening of the ribcage results in defective ventilation with respiratory obstruction and infection. In older children and adolescents, symptoms similar to those observed in adult osteomalacia, including bone pain, waddling gait, and fatigue, may be present (12–14, 24, 30–32). Hematologic disorders are often observed in common rickets, including hypochromic anemia and the rare Von Jacksch–Luzet syndrome. This syndrome is associated with severe anemia and a profile of chronic myeloid leukemia with erythroblastosis, leukocytosis, myelocytosis, and possible myeloblastosis. The spleen and liver can be enlarged as a result of extramedullary hematopoiesis. The bone marrow is hypoplastic. This syndrome is often cured with simple vitamin D therapy (31).

Severe calcium deficiency can lead to rickets in much the same way as vitamin D deficiency (32–35). Very low dietary calcium intake leads to decreased ionized calcium and secondary hyperparathyroidism. This causes a mineralization defect in the skeleton that results in growth retardation and many of the skeletal manifestations seen in vitamin D deficiency, but these are of greater severity due to the hypocalcemia (31–35) (Figure 4).

Inadequate calcium intake during the 3rd trimester of pregnancy can cause a serious calcium deficit in the fetal skeleton that is rapidly being mineralized during the last 7 weeks in utero. Typically at 28 weeks, 100 mg/d of calcium is being deposited in the skeleton, whereas at 35 weeks 350 mg/d is being deposited (30, 31, 36). Therefore, mother's milk containing 240–340 mg/l of calcium is unable to meet the demands of postnatal accretion rates of a preterm infant (30, 31).

Young children and adolescents, especially non-white individuals, on a strict vegetarian diet or a diet that is high in phytate, which binds calcium, can also be calcium deficient, which leads to rickets (33–35). This, in combination with vitamin D deficiency, is often the precipitating cause of rickets in children of Middle Eastern descent living in Great Britain and African American children in the United States (32–35).

The calcium deficiency and associated secondary hyperparathyroidism increase the requirement for vitamin D, since the vitamin D is rapidly metabolized to $1,25(OH)_2D$. The combination of calcium deficiency and vitamin D deficiency accelerates and makes more severe the skeletal abnormalities and hypocalcemia.

Prevalence of subclinical vitamin D deficiency

Severe chronic vitamin D deficiency [25(OH)D level less than 15 ng/ml] leads to overt skeletal abnormalities in children that is typically defined as rickets (23, 30–32). However, there is a large number of infants, children, and adolescents who are vitamin D insufficient but have no apparent skeletal or calcium metabolism abnormalities (Table 2). We observed that of 40 "healthy" mother-infant pairs that were predominantly non-white, 73% of mothers and 80% infants had 25(OH)D levels of less than 20 ng/ml despite the fact that 80% of the mothers took a daily prenatal multivitamin that contained 400 IU of vitamin D (37). Sullivan et al. (38) reported that 48% of white girls aged 9–11 years in Maine had 25(OH)D levels less than 20 ng/ml at the end of the winter and 17% remained vitamin D deficient at the end of the summer due either to avoiding sun exposure or always wearing sun protection. Forty-two percent of adolescent African American and Hispanic children had 25(OH)D levels less than 20 ng/ml in Boston (39), which is consistent with the observation by the Centers for Disease Control that 48% of African American women aged 15–49 years throughout the entire United States had 25(OH)D levels of less than 15 ng/ml at the end of the winter (40). Similar observations have been made in Canada and Europe, where few foods are fortified with vitamin D and the high latitude limits vitamin D production in the skin (41–45). Remarkably, in the sunniest areas of the world, rickets is a major health problem. Because of the practice of purdah or wearing a burka (45, 46), avoidance of exposure of any skin to sunlight, and the fact that few foods are fortified with vitamin D, upward of 35–80% of children in Saudi Arabia (46, 47), India (48), Turkey (29), New Zealand (49), Israel (50), Egypt (51), Hong Kong (52), China (53), Libya (54), Lebanon (55), Spain (56), Australia (57), San Diego, California (58), and the southeastern United States (59) are vitamin D deficient. When the deficiency occurs during fetal life, there is data to suggest that this may cause an increased risk of hip fractures and bone loss later in life (56, 60, 61). Subclinical vitamin D deficiency in neonates is associated with a normal serum calcium level, low 25(OH)D concentration (typically between 10 and 20 ng/ml), and elevated serum PTH, $1,25(OH)_2D$ and alkaline phosphatase levels (30–32).

Inherited causes of rickets

Once it was recognized that vitamin D must be metabolized in the liver and kidneys before it can carry out its biologic effects on calcium, phosphorus, and bone metabolism, it was hypothesized that a defect in the hepatic 25-hydroxylation or renal 1α-hydroxylation steps would lead to an inability to activate vitamin D, thus causing

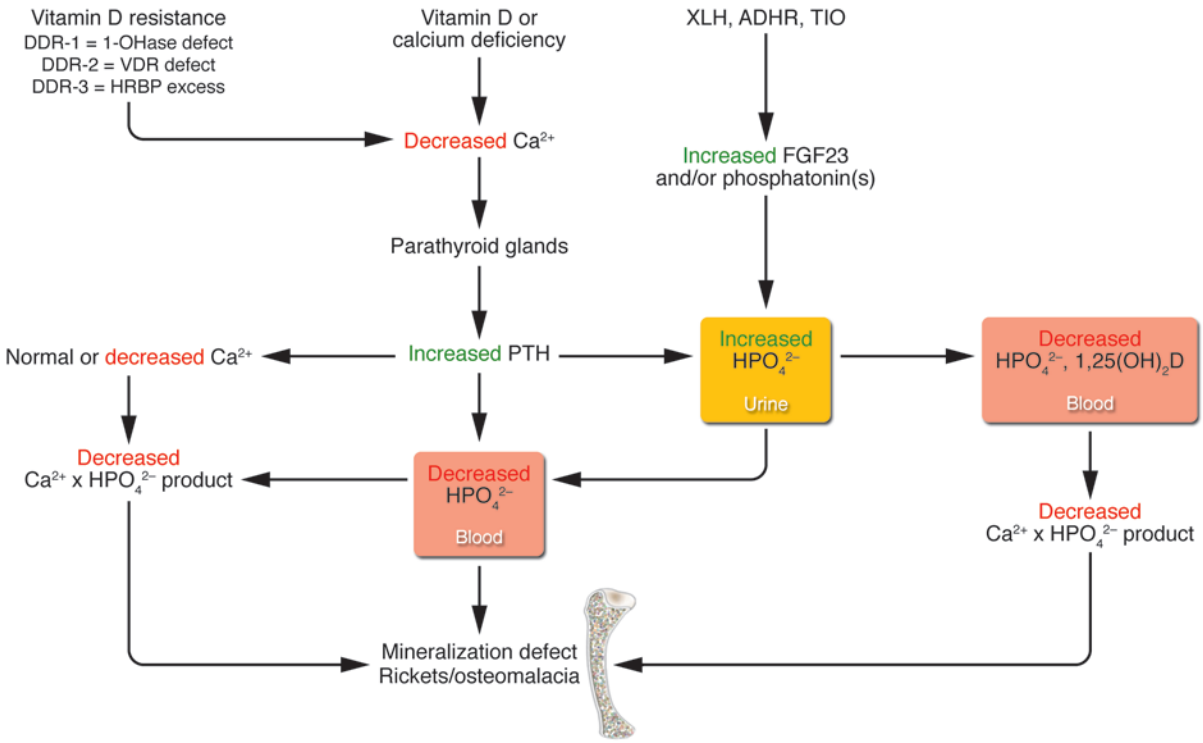

Figure 4
Biochemical changes in calcium and phosphorus metabolism due to vitamin D or calcium deficiency, vitamin D–resistant syndromes, or hypophosphatemic syndromes that cause rickets or osteomalacia. Vitamin D and/or calcium deficiency leads to a decrease in the level of ionized calcium (Ca^{2+}), resulting in an increase in PTH. PTH increases tubular reabsorption of calcium to correct the serum calcium into the normal range. However, in severe calcium and vitamin D deficiency, the serum calcium is below normal. In addition, PTH causes phosphorus loss via the urine, resulting in a decrease in serum HPO_4^{2-}. An inadequate calcium-phosphorus product ($Ca^{+2} \times HPO_4^{2-}$) leads to a defect in bone mineralization that causes rickets in children and osteomalacia in adults. There are various inherited and acquired disorders that can disrupt calcium and phosphorus metabolism that can also result in defective mineralization of the skeleton. There are 3 inherited syndromes that cause vitamin D resistance. Vitamin D–dependent rickets type 1 (DDR-1) is due to a mutation of the 1-OHase. A mutation of the *VDR* gene results in an ineffective recognition of $1,25(OH)_2D$, causing DDR-2. A genetic defect that results in the overproduction of hormone response element–binding protein (HRBP) eliminates the interaction of $1,25(OH)_2D$ with its VDR, resulting in DDR-3. There are also inherited and acquired disorders that cause severe hypophosphatemia and decrease renal production of $1,25(OH)_2D$. The acquired disorders X-linked hypophosphatemic rickets (XLH) and autosomal dominant hypophosphatemic rickets (ADHR) are caused by the increased production or decreased destruction, respectively, of phosphatonins that include FGF23. Tumor-induced osteomalacia (TIO) is caused by the tumor's production of FGF23, which results in phosphaturia and a decrease in the renal production of $1,25(OH)_2D$.

a vitamin D deficiency–like state that was resistant to physiologic doses of vitamin D (Figure 4). There is only one documented case of 25-OHase–deficiency rickets (62). The most likely reason why more cases are not reported is that there are at least 4 different enzymes that have the ability to convert vitamin D to 25(OH)D (63).

Since vitamin D undergoes its final activation in the kidneys, several studies of individuals with "vitamin D–resistant" diseases causing rickets in which patients were evaluated for a defect in the metabolism of 25(OH)D to $1,25(OH)_2D$ have been reported. Pseudovitamin D–deficiency rickets (also known as hereditary, vitamin D–dependent rickets type 1), a rare hereditary disorder, was found to be associated with very low or undetectable levels of $1,25(OH)_2D$ in the circulation (64). These children responded to orally administered $1,25(OH)_2D_3$ (64). The cloning of the renal 1-OHase enzyme led to the identification of a multitude of point mutations of the *CYP27B1* gene, which result in either a poorly functional 1-OHase or the complete absence of 1-OHase activity (65).

Several investigators reported children with severe rickets who often had alopecia and extremely elevated levels of $1,25(OH)_2D$ (65, 66). Some children with this disease, vitamin D–resistant rickets (hereditary, vitamin D–dependent rickets type 2), responded to pharmacologic doses of vitamin D or $1,25(OH)_2D_3$, while others did not (66, 67). Point mutations in the *VDR* gene are responsible for the vitamin D resistance. Chen et al. (68) reported a new form of vitamin D resistance, hereditary vitamin D–dependent rickets type 3, caused by the abnormal expression of a hormone response element–binding protein (HRBP) that binds to the VDRE and therefore prevents the $1,25(OH)_2D$-VDR-RXR complex from binding to its responsive element. This patient had normal VDR expression and was completely resistant to $1,25(OH)_2D_3$ action. Children with these vitamin D–resistance syndromes often suffer from severe bone deformities and more marked hypocalcemia than children with vitamin D–deficiency rickets. Treatment depends on the cause and severity of the vitamin D resistance. Children have responded to pharmacologic doses of vitamin D, physiologic and pharmacologic doses of $1,25(OH)_2D_3$ and its analog 1α-hydroxyvitamin D_3, as well as intravenous infusions of calcium and phosphorus (30, 64, 66–69).

Table 2
Vitamin D status and associated biochemistries: serum levels of 25(OH)D, $1,25(OH)_2D$, Ca, HPO_4^{2-}, alkaline phosphatase (Alk. phos.), PTH, and FGF23

	25(OH) D, ng/ml	1,25(OH)2D	Ca	HPO_4^{2-}	Alk. phos.	PTH	FGF23	Skeletal disease
Vitamin D deficiency	<20	↑	↓ NL	↓	↑	↑	NL	Rickets/osteomalacia
Vitamin D insufficiency	21–29	↑ or NL	NL	NL	↑ or NL	↑ or NL	NL	↓ BMD
Vitamin D sufficiency	>30	NL	NL	NL	NL	NL	NL	None
XLH	NL	↓	NL	↓↓	↑	NL	↑ or NL	Rickets
ADHR	NL	↓	NL	↓↓	↑	NL	↑↑	Rickets
TIO	NL	↓	NL	↓↓	↑	NL	↑↑	Rickets

The upward-pointing arrows (↑ and ↑↑) indicate that the level is moderately or markedly above the normal range, respectively, and the downward-pointing arrows (↓ and ↓↓) indicate that the serum level is moderately or markedly below the normal range, respectively. NL represents levels within the normal range. BMD, bone mineral density; XLH, X-linked hypophosphatemic rickets; ADHR, autosomal dominant hypophosphatemic rickets; TIO, tumor-induced osteomalacia.

Inherited and acquired hypophosphatemic rickets

These disorders are characterized by hypophosphatemia, decreased reabsorption of phosphorus by the renal tubule, decreased absorption of calcium and phosphorus from the gastrointestinal tract, and varying degrees of rickets or osteomalacia (70, 71). Patients often have normal or reduced serum levels of $1,25(OH)_2D$, which is considered to be abnormal, since hypophosphatemia causes an increase in serum $1,25(OH)_2D$ levels because it enhances the renal production of $1,25(OH)_2D$ (70, 72). Originally it was thought that hypophosphatemic disorders were caused by a defect in a renal phosphate transport protein. However, recent evidence suggests that other factors of bone origin participate in maintaining phosphorus homeostasis, including fibroblast growth factor 23 (FGF23), matrix extracellular phosphoglycoprotein, and frizzled-related protein 4. These factors are collectively known as phosphatonins (73). When FGF23 is elevated, it causes an internalization of the sodium phosphate cotransporter in both the kidneys and intestine, thereby causing phosphaturia and decreased intestinal phosphate absorption (74). It also inhibits CYP27B1 activity. Patients with autosomal dominant hypophosphatemic rickets (ADHR) have a mutation in the *FGF23* gene that prevents or reduces FGF23 metabolic breakdown, leading to elevated FGF23 levels (70, 75). Tumor-induced osteomalacia is caused by a small tumor that is often benign and secretes FGF23 (70, 71, 73, 74). The exact cause of X-linked hypophosphatemic rickets is less well understood. It has been linked to a mutation of the *phosphate regulating endopeptidase homolog, X-linked* (*PHEX*) gene. Loss of its expression causes overexpression of FGF23 and possibly other phosphatonins in bone, leading to increased levels of circulating FGF23 (74).

Thus, hypophosphatemic rickets is caused by an inability to metabolize FGF23 or the excessive production of FGF23 (70, 71, 73, 74) (Figure 4). Intravenous phosphate delivery has been effective in treating rickets and osteomalacia (70, 71, 76), and removal of the tumor is curative (77, 78). Treatment includes frequent oral phosphate administration, typically 250–500 mg up to 5 times a day, as well as twice daily oral delivery of 0.5–1.0 μg of $1,25(OH)_2D_3$ (70, 71, 76). Less frequent administration of higher doses of phosphate is to be discouraged, since the transient increase in serum phosphate causes a decrease in ionized calcium and an increase in PTH level and causes parathyroid glands to become hyperplastic and autonomous, which leads to tertiary hyperparathyroidism.

Prevention and treatment of vitamin D– and calcium-deficiency rickets

In the 1940s, the recommended intake of vitamin D for infants was 100 IU/d to prevent rickets (14). However, the current accepted recommendation to prevent rickets is a daily 400 IU dose of vitamin D and adequate calcium intake (32–35, 79). Clinical trials in preterm infants (16 days of age) were randomized to daily vitamin D intakes of 200 IU (90 IU/kg), 400 IU (180 IU/kg), or 800 IU (360 IU/kg) for up to one month. No radiological differences were observed between groups (80). The 25(OH)D levels in the group receiving 200 IU vitamin D for 24–29 days did not change, whereas the groups receiving 400 IU and 800 IU for the same period of time showed an increase in 25(OH)D levels of approximately 30%. Similar studies also suggested that preterm infants' plasma 25(OH)D levels were maintained from early neonatal life to 3 months with administration of supplemental vitamin D of 400 IU/d. No benefit in vitamin D status or forearm bone mineral density was observed at a higher dose of 900 IU/d (30, 81). In Europe, the Nutrition Committee for the European Society of Pediatric Gastroenterology, Hepatology and Nutrition recommended a vitamin D intake of 800–1,600 IU/d (36). Mawer et al. (36) gave either 1,000 or 3,000 IU of vitamin D_2 daily to low-birth-weight infants with a mean weight of 1.36 kg and found that the 25(OH)D level increased from a baseline of 6–10 ng/ml to a mean of 33 ng/ml in both dose groups after 7 weeks. They also observed that the 25(OH)D levels rapidly rose during the first week and began to plateau at 7 weeks at approximately 33 ng/ml. Markestad et al. (23) treated children with 1,700–4,000 IU vitamin D_2/day for up to 10 weeks and showed an average 25(OH)D level of approximately 30 ng/ml and correction of their biochemical and skeletal abnormalities. Premature neonates treated with 1,200 IU vitamin D_3/day for 7 days raised their 25(OH)D level from 8 ng/ml to 18 ng/ml (81). Although it is not known what the minimum normal level of 25(OH)D should be for infants and neonates based on these observations, it is not unreasonable for the blood level to be at least 20 ng/ml. However, since those studies showed that 25(OH)D levels reached a plateau at approximately 33 ng/ml, this is likely the ideal healthy level for infants and children and is similar for adults (23, 24, 30, 36, 81–84). To achieve a healthy 25(OH)D level of greater than 30 ng/ml, infants require at least 400–1,000 IU of vitamin D/day depending on their 25(OH)D levels at birth.

Infants who are vitamin D deficient should not simply receive what is the recommended US adequate intake (200 IU/d) (85) or

even 400 IU/d but rather should be aggressively treated with pharmacologic doses of vitamin D in order to build up the body stores of vitamin D and quickly correct the vitamin D deficiency. The best method to effectively treat and cure rickets is to give a total of 5–15 mg (200,000–600,000 IU) of vitamin D_2 or vitamin D_3 orally with adequate dietary calcium (86). These doses can be given safely either as a single-day therapy or as daily doses of 2,000–4,000 IU/d (50–100 μg/d) for 3–6 months (30, 86, 87). Typically there is rapid correction of both serum calcium and phosphorus levels within 6–10 days and normalization of PTH levels within 1–2 months. Alkaline phosphatase decline and healing of radiologic signs of rickets are observed within 3–6 months depending on the severity of the deficiency (30, 86). For those who may not comply with this regime, it is recommended that 5 mg (200,000 IU) of vitamin D be given as a single oral dose, with a follow-up dose of 5 mg 3 months later. It is imperative to initiate therapy with large doses of vitamin D, since giving small daily doses of 200–400 IU/d will not restore adequate stores of vitamin D as rapidly as either a single large dose or daily doses that are 10- to 20-fold higher than the recommended adequate intake (AI) (30, 85, 86, 87). For infants and children who have fat malabsorption, including cystic fibrosis patients (88), it is recommended that subcutaneous or intramuscular administration be used. Alternatively, controlled exposure to sunlight or UV radiation from a commercial lamp is advisable (5, 6, 12–14, 30, 33, 88).

Sunlight, UV irradiation, and neonatal and maternal vitamin D supplementation

Regular and sensible sun exposure during the months of the year when vitamin D production is promoted is still the most physiologic way to prevent vitamin D deficiency in infants and young children (1, 24, 80, 89). Seasonal variations in serum 25(OH)D in children and adults is well documented, with levels reaching a peak in the middle of the summer and nadir at the end of the winter in both the Northern and Southern Hemispheres (1, 19, 24, 57, 89). Since breast milk has very little, if any, vitamin D (usually no more than 25 IU/l), it is usually inadequate in satisfying the infant's requirement (85, 87, 90). Thus, if the infant is receiving nutrition solely from breast-feeding and if the mother is vitamin D deficient, the infant will become vitamin D deficient and will likely develop rickets (30, 37, 90).

Neonates and young children who do not receive adequate vitamin D from their diet respond well to oral doses of 1,000–1,500 IU/d all year up to the age of 2 and during the winter up to age 5 years without any sign of vitamin D intoxication (30). This prevention scheme has been very effective in Europe for at-risk infants who are exclusively breast-fed or who are too old to take formula (30). Prevention with lower daily doses of 400–500 IU/d is recommended for other breast-fed infants and for at-risk neonates and young infants receiving formula. The effect of providing mother or infant during lactation with supplements of 400 IU of vitamin D showed that it is most effective to give the infant vitamin D supplementation daily. However, Hollis et al. (90) reported that giving lactating females 4,000 IU vitamin D_3 daily provides adequate vitamin D in breast milk to satisfy the infant's requirement.

Noncalcemic and nonskeletal consequences of vitamin D deficiency in children

Children with vitamin D deficiency often suffer from severe muscle weakness with toneless and flabby legs (1, 2, 12–14, 30–32). It is now recognized that skeletal muscle has a VDR and that $1,25(OH)_2D$ improves muscle function (91). Serum 25(OH)D levels above 30 ng/ml maximize proximal muscle leg function in adults (92). It was observed in healthy adults that if the serum 25(OH)D was greater than 20 ng/ml, there was a significant increase in lung function, with an average increase in forced expiratory volume of 176 ml (93). Camargo et al. (94) reported a prospective study of maternal intake of vitamin D during pregnancy and observed that vitamin D deficiency was highly predictive of increased risk for asthma.

The VDR is present not only in tissues that regulate serum calcium, including the small intestine, bone cells, and kidney, but also in essentially all tissues and cells in the body, including brain, colon, breast, prostate, pancreas, heart, skin, skeletal muscle, monocytes, and activated T and B lymphocytes (1, 20–22, 24). The first insight into the noncalcemic role of $1,25(OH)_2D_3$ was observed when $1,25(OH)_2D_3$ was incubated with mouse and human leukemic cells. $1,25(OH)_2D_3$ inhibited leukemic cell proliferation and induced the cells to mature (20–22, 95, 96). Many cancer cell lines and primary cancer cell cultures that posses a VDR demonstrate marked inhibition of growth and induction of maturation when exposed to $1,25(OH)_2D_3$ or its active analogues (1, 20–22, 24, 96) (Figure 5). 1,25(OH)D does this by inducing cellular maturation, regulating the expression of p21 and p27 and apoptosis, and acting as an antiangiogenic factor (20–22, 24, 95, 96).

Living at higher latitudes and being prone to vitamin D deficiency increase risk of cancers of the colon, prostate, breast, ovary, esophagus, and several other tissues (1, 24, 97–99). It has been suggested that maintenance of a 25(OH)D level greater than 20 ng/ml reduces risk of colon, prostate, breast, and ovarian cancer by as much as 30–50% (24, 97–99). Although it is unknown whether vitamin D deficiency in utero and during infancy and childhood would imprint on the child for the rest of his or her life an increased risk of these deadly cancers, the recent observation that children exposed to the most sunlight had a 40% reduced risk of developing non-Hodgkin lymphoma (100) and increased survival from malignant melanoma (101) suggests that maintenance of adequate 25(OH)D levels throughout life may help reduce risk of many deadly cancers (24, 97–99, 102).

Living above the 35° latitude for the first 10 years of life imprints on a child for the rest of his or her life a 100% increased risk of developing multiple sclerosis no matter where they live thereafter (24, 103, 104). Living at higher latitude and being prone to vitamin D deficiency increases risk of several other autoimmune diseases including type 1 diabetes and Crohn disease (105, 106). Children in Finland in the 1960s who received the recommended 2,000 IU of vitamin D/day at least during the first year of life and followed for the next 31 years demonstrated a reduced risk of developing type 1 diabetes by 80% (105). Furthermore, children from the same cohort who were vitamin D deficient at one year of age had a 2.4-fold increased risk of developing type 1 diabetes. Vitamin D deficiency in utero and during the first year of life has also been linked to increased risk of type 1 diabetes (107). $1,25(OH)_2D$ affects the immune system (106, 108), and, as pancreatic islet β cells have a VDR, it also stimulates insulin secretion (20–22, 24) (Figure 5). Thus, hypovitaminosis D in children may increase their risk not only of type 2 diabetes but also insulin resistance and islet β cell dysfunction (109).

Living at higher latitude and vitamin D deficiency are also associated with hypertension and cardiovascular heart disease (24, 110, 111). Li et al. (112) reported that $1,25(OH)_2D_3$ is an effective regulator of renin production, which controls blood pressure. A

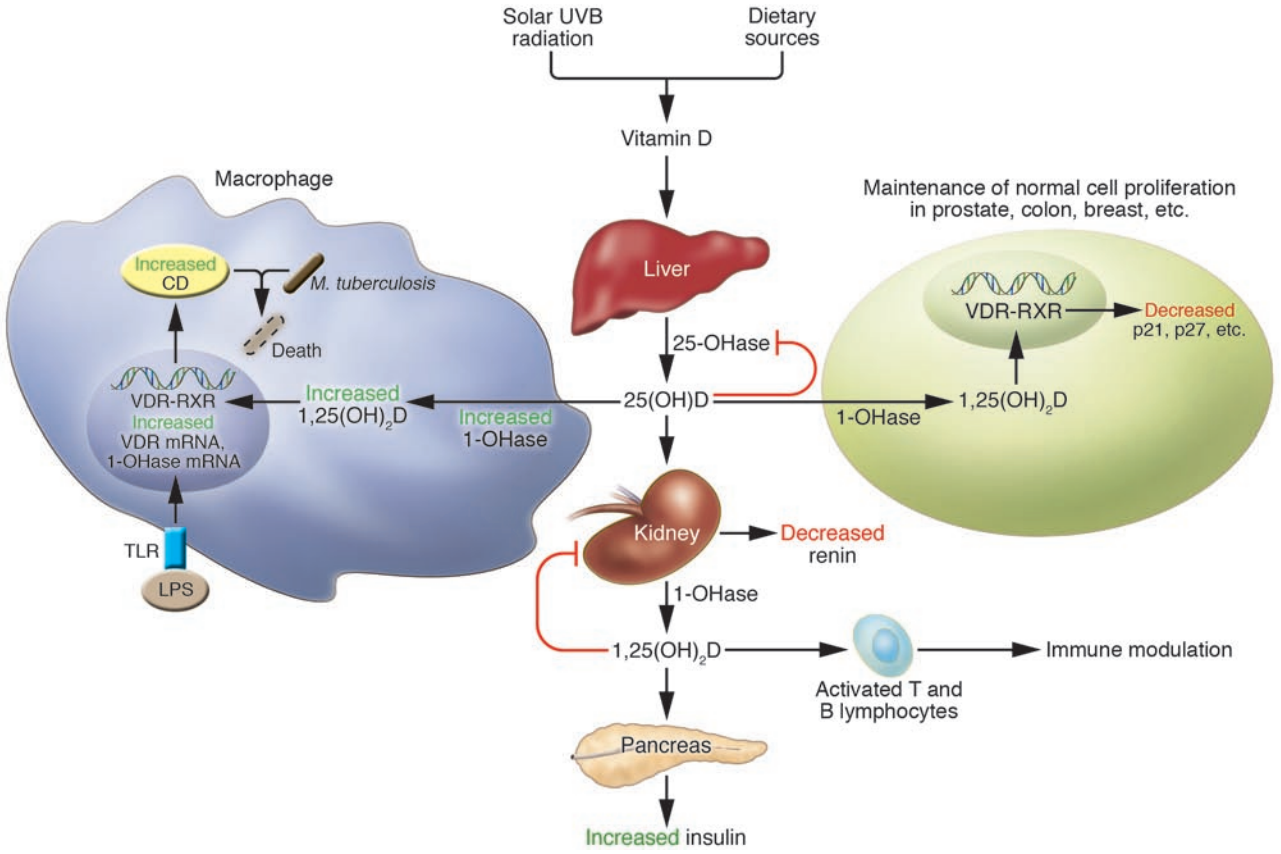

Figure 5
Noncalcemic functions of $1,25(OH)_2D$. Vitamin D coming from the photoproduction of previtamin D or coming from the diet is converted in the liver to 25(OH)D by the vitamin 25-OHase. 25(OH)D is converted in the kidneys by 1-OHase. $1,25(OH)_2D$ not only regulates calcium and phosphorus metabolism but can stimulate the pancreas to produce insulin and to downregulate the renal production of renin. $1,25(OH)_2D$ also interacts with its nuclear receptor (VDR) in a wide variety of tissues and cells and helps maintain normal cell proliferation and differentiation. 25(OH)D can also be converted to $1,25(OH)_2D$ in a wide variety of cells, including colon, prostate, and breast, for the autocrine production of $1,25(OH)_2D$. It is believed that the autocrine production of $1,25(OH)_2D$ is important for regulating cell growth and maturation, which decreases risk of the cell becoming malignant. 25(OH)D also is metabolized in macrophages by the 1-OHase to produce $1,25(OH)_2D$. The expression of the VDR and 1-OHase is upregulated when TLR2/1 is stimulated by LPS. This results in an increase in the expression of the VDR and the 1-OHase. The increase production of $1,25(OH)_2D$ increases the nuclear expression of cathelicidin (CD) in the macrophage, which is a cationic peptide that causes the destruction of infective agents including *M. tuberculosis*.

study in hypertensive adults exposed to simulated sunlight 3 times a week for 3 months resulted in an increase in their 25(OH)D levels by more than 150% and a significant (6 mmHg) reduction in both systolic and diastolic blood pressure (113).

Autocrine production and function of $1,25(OH)_2D$

Circulating levels of $1,25(OH)_2D$ are very low or undetectable in patients with chronic kidney disease (114). It has been assumed that the kidneys are the sole source of $1,25(OH)_2D$. However, just as most tissues and cells in the body have a VDR, so too do these tissues and cells possess the ability to express CYP27B1. Thus, the skin, prostate, breast, colon, lung, brain, and placenta not only express the VDR but also have the capacity to produce $1,25(OH)_2D$ (22, 24, 115, 116). It is now recognized that $1,25(OH)_2D$ helps control the expression of more than 200 genes (20–22, 24, 117). It is thought that $1,25(OH)_2D$ maintains cellular health by acting as a sentinel for preventing malignancy (24, 95, 96) (Figure 5).

Activated macrophages also express CYP27B1 and thus produce $1,25(OH)_2D$. This is the mechanism by which patients with chronic granulomatous diseases such as sarcoidosis and tuberculosis develop a disorder in calcium metabolism that causes hypercalciuria and hypercalcemia (1, 22, 24). Why macrophages produce $1,25(OH)_2D$ was unknown until Liu et al. (118) reported that activation of TLRs with LPS resulted in the upregulation of the expression of not only VDR but also the *CYP27B1* gene. The local production of $1,25(OH)_2D$ induced the expression of the antimicrobial peptide cathelicidin (LL-37), which is thought to be a key factor in the innate immune response when TLR is activated by an infective agent such as *Mycobacterium tuberculosis* (Figure 5). This remarkable observation explains why patients with TB often do better when placed in a solarium and exposed to sunlight or taken to higher altitudes where the vitamin D_3 production in the skin is more efficient (14). This also is the likely reason why African Americans, who are often vitamin D deficient, and children with vitamin D deficiency have increased susceptibility to TB infection (118). This also may explain why it was widely reported that children with rickets often are more prone to infectious diseases, including the common cold virus (12–14, 119).

Conclusion

Vitamin D–deficiency rickets is a sunlight deficiency disease. The inability to appreciate the beneficial effect of sunlight for health had devastating consequences for both children and adults for more than 300 years. When it was finally realized that exposure to sunlight could prevent and treat rickets, this led to the recommendation that all children be exposed to sensible sunlight to maximize bone health. The fortification of milk with vitamin D eradicated rickets as a major health problem, and, therefore, it was thought to have been conquered.

Rickets has, however, made an unfortunate comeback (120). The major cause of rickets in the United States is a lack of appreciation that human milk contains very little if any vitamin D to satisfy the infant's requirement. African American women are often vitamin D deficient, and women who always wear sun protection and only take a prenatal multivitamin are also at a high risk of vitamin D insufficiency. If they provide breast milk to their infant as the sole source of nutrition, the infant will become vitamin D deficient. If the infant is not exposed to sunlight or does not receive a vitamin D supplement, the infant will inevitably develop rickets. However, the skeletal manifestations of rickets represent only the tip of the vitamin D deficiency iceberg. Vitamin D deficiency in utero and during the first year of life has devastating consequences and may imprint on the child's life chronic diseases that will shorten his/her life span (24, 57). In utero, vitamin D deficiency results in reduced intrauterine long bone growth and slightly shorter gestation (121). This has been linked to increased risk of osteoporosis and fractures later in life (24, 60, 61, 82, 122). Children born and raised at latitudes below 35° for the first 10 years have a 50% reduced risk of developing multiple sclerosis later in life (103, 104). Neonates who are vitamin D deficient during the first year of life are 2.4-fold more likely to develop type 1 diabetes compared with children who received 2,000 IU of vitamin D_3/day (105). It has been suggested that the increased risk of developing schizophrenia may be initiated in utero and during childhood due to vitamin D deficiency (102). Muscle function, innate immunity, cellular growth and maturation, immunomodulation, insulin secretion, as well as regulation of calcium, phosphorus, and bone metabolism are all affected or controlled by vitamin D. Thus, ensuring that women during pregnancy are vitamin D sufficient and that newborns either be immediately evaluated for their vitamin D status by measuring 25(OH)D levels in cord blood or given vitamin D prophylactically should be a high priority. Vitamin D deficiency should be immediately treated with at least 1,000 IU of vitamin D_2 or vitamin D_3/day for the first week of life. Alternatively, a single dose of 200,000 IU of vitamin D should suffice for the first few months of life.

There has been a great fear about causing vitamin D intoxication in neonates. This resulted from the poorly described outbreak of neonatal hypercalcemia in the 1950s in Great Britain (123), which led to the enactment of laws in Europe forbidding the fortification of dairy products as well as all other products with vitamin D. In 1997 the Institute of Medicine recommended that the AI for infants and children of all ages be 200 IU/d. The same recommendation was made for pregnant and lactating women. The safe upper limit for infants ages 0–12 months was 1,000 IU/d and for children older than 1 year of age, 2,000 IU/d. However, it is now obvious based on the historical literature (14–16) as well as the recent literature (23, 24, 30, 36, 81, 86, 87) that these recommendations are inadequate without sensible sun exposure. It is well documented that neonates and children can tolerate a single dose of 200,000 IU of vitamin D_2 or vitamin D_3 or doses of vitamin D up to 3,000 IU/d without any untoward side effects. Indeed 400–1,000 IU/d to maintain serum 25(OH)D levels between 30–50 ng/ml should be the goal, just as it is in adults. Infants and children have routinely received 400–2,000 IU vitamin D_2 or vitamin D_3/day for the first years of life without any reports of toxicity (23, 80, 105, 107). Typically, doses of more than 50,000 IU/d of vitamin D_2 were found to cause toxicity (12–14).

In Canada, it is recommended that all infants receive 400 IU/d from birth. This recommendation has been successfully implemented and has not resulted in any reported cases of vitamin D intoxication or hypercalcemia. I believe that the 200 IU of vitamin D that is recommended by the American Academy of Pediatrics is suboptimal (124). This dose may prevent overt rickets but will not prevent vitamin D deficiency.

Hopefully, history will not repeat itself. The widespread concern about any direct sun exposure increasing the risk of the relatively benign and nonlethal squamous and basal cell cancers needs to be put into perspective. It is chronic excessive exposure to sunlight and sunburning experiences during childhood that increases risk of nonmelanoma skin cancer (125). Melanoma, one of the most feared cancers because of its ability to rapidly metastasize before it is obvious to either the patient or physician, has been branded as a sun-induced skin cancer. However, most melanomas occur on the least sun-exposed areas, and it has been reported that occupational exposure to sunlight decreases risk of melanoma (125).

The 30-year campaign to recommend abstinence from sun exposure has not stemmed the increase in skin cancer incidence (125). It is curious that in the 1930s and 1940s, when children were encouraged to be exposed to sunlight and artificial UV radiation to treat rickets, the incidence of skin cancer did not increase. Thus, there needs to be a reevaluation of the beneficial effect of sensible exposure to sunlight as noted by the Australian College of Dermatologists and the Cancer Council Australia, which recommend a balance between avoiding an increase risk of skin cancer and achieving enough UV radiation to maintain adequate vitamin D levels.

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