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Vitamin D: A Millenium Perspective

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Abstract Vitamin D is one of the oldest hormones that have been made in the earliest life forms for over 750 million years. Phytoplankton, zooplankton, and most plants and animals that are exposed to sunlight have the capacity to make vitamin D. Vitamin D is critically important for the development, growth, and maintenance of a healthy skeleton from birth until death. The major function of vitamin D is to maintain calcium homeostasis. It accomplishes this by increasing the efficiency of the intestine to absorb dietary calcium. When there is inadequate calcium in the diet to satisfy the body's calcium requirement, vitamin D communicates to the osteoblasts that signal osteoclast precursors to mature and dissolve the calcium stored in the bone. Vitamin D is metabolized in the liver and then in the kidney to 1,25-dihydroxyvitamin D [1,25$(OH)_2$D]. 1,25$(OH)_2$D receptors (VDR) are present not only in the intestine and bone, but in a wide variety of other tissues, including the brain, heart, stomach, pancreas, activated T and B lymphocytes, skin, gonads, etc. 1,25$(OH)_2$D is one of the most potent substances to inhibit proliferation of both normal and hyperproliferative cells and induce them to mature. It is also recognized that a wide variety of tissues, including colon, prostate, breast, and skin have the enzymatic machinery to produce 1,25$(OH)_2$D. 1,25$(OH)_2$D and its analogs have been developed for treating the hyperproliferative disease psoriasis. Vitamin D deficiency is a major unrecognized health problem. Not only does it cause rickets in children, osteomalacia and osteoporosis in adults, but may have long lasting effects. Chronic vitamin D deficiency may have serious adverse consequences, including increased risk of hypertension, multiple sclerosis, cancers of the colon, prostate, breast, and ovary, and type 1 diabetes. There needs to be a better appreciation of the importance of vitamin D for overall health and well being. J. Cell. Biochem. 88: 296–307, 2003.

Key words: vitamin D; sunlight; bone; cancer; psoriasis

EVOLUTIONARY PERSPECTIVE ON VITAMIN D

Little is known about when vitamin D was first made and what its function was. Bills [1924], [1927] suggested that the vitamin D content found in oily fish and in fish liver oils was due to the dietary intake of vitamin D from phytoplankton and zooplankton. He demonstrated a seasonal variation in the vitamin D content in oily fish and fish liver oils, with the highest content found in the summer and the lowest in the winter. Saleeby suggested in the early 1920s that vitamin D in cod liver oil was probably made by sunlight falling on the green plankton in the fall waters of the North Atlantic [Steenbock and Black, 1925; Holick, 1989].

Phytoplankton produce over 120 billion tons of organic carbon each year, compared to 20 billion tons produced by terrestrial plants. A single fish consumes ~1.2% of its body weight every 24 h and it has been estimated that 0.5 tons of diatoms make a pound of seal, while a pound of killer whale, a predator of seals, requires 5 tons of diatoms [Prescott, 1968; Holick, 1989]. Thus, it would not be surprising that if phytoplankton were able to photosynthesize even a small amount of vitamin D that fish, through the concentrating power of the food chain, would obtain a large amount of vitamin D in their diet that could ultimately be stored in their body fat [Drummond and Gunther, 1930; Holick, 1989]. In support of this hypothesis, Copping [1934] collected copepods in the North Atlantic and the dried copepods were found to have antirachitic activity.

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To better define whether phytoplankton could photosynthesize vitamin D, Holick et al. [1982a]; Holick [1989] grew 100 L of *Emiliania huxlei* (an organism that has existed unchanged in the Sargasso sea for at least 750 million years) and *Skeletonema menzelii* in pure culture to a cell density of 10^6 cells per ml in glass carboys in the absence of ultraviolet B (UVB) radiation. The cells were harvested by centrifugation, resuspended in synthetic sea water, and 50% of the cells were transferred into a quartz vessel and exposed to simulated solar ultraviolet radiation. ^{3}H-7-dehydrocholesterol was added to the cells before they were extracted with diethylether for recovery determinations and the lipid extracts were chromatographed on a straight phase high performance liquid chromotograph (HPLC). The ultraviolet absorbing peak was analyzed by mass spectroscopy and demonstrated an apparent molecular ion of 396 suggesting that the major component was ergosterol (Fig. 1). The cells that were exposed to UVB radiation demonstrated a marked reduction in ergosterol and the appearance of previtamin D_2 (Fig. 2) that was confirmed by mass spectroscopy. The amount of ergosterol in *E. huxlei* was 1.0 μg of ergosterol/g wet weight. *S. menzelii* was also found to photosynthesize vitamin D_2 from ergosterol [Holick et al., 1982a,b; Holick, 1989].

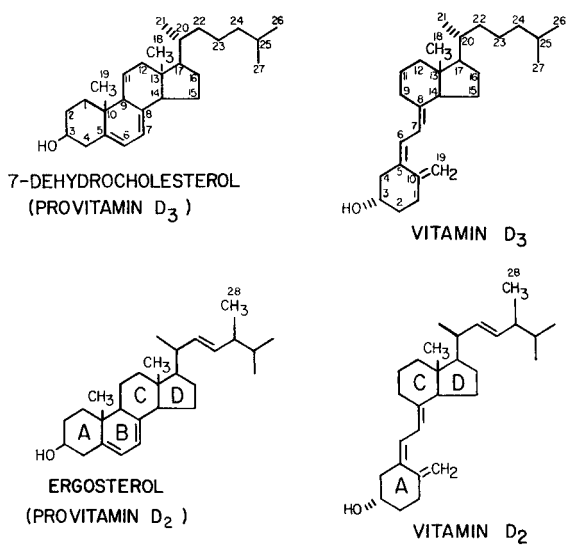

Fig. 1. Structure of vitamin D_3 and D_2 and their respective precursors, 7-dehydrocholesterol, and ergosterol. The only structural difference between vitamin D_2 and D_3 is in their side chains; the side chain for vitamin D_2 contains a double bond between C-22 and C-23 and a C-24 methyl group. (Reproduced with permission) [MacLaughlin and Holick, 1983].

An evaluation of plankton net tows from the Sargasso sea revealed a wide variety of provitamin D's. Brine shrimp and krill were lipid extracted and an HPLC analysis showed several provitamin D's, including 7-dehydrocholesterol, ergosterol, and other unidentified provitamin D's. Upon exposure to simulated sunlight, both krill and brine shrimp converted their provitamin D's to their respective previtamin D's [Holick et al., 1982a; Holick, 1989]. Brine shrimp and krill contained 82 and 74% of their total provitamin D content as 7-dehydrocholesterol.

Although the function of provitamin D is not known in either phytoplankton or zooplankton, there are at least three possible functions based on the provitamin D's ability to absorb UVB radiation. Since the UV absorption spectrum for provitamin D, previtamin D, and vitamin D completely overlap the UV absorption spectra for DNA, RNA, and proteins, it is possible that the provitamin D evolved as a natural sunscreen to protect the UV sensitive macromolecules from solar ultraviolet radiation damage (Fig. 3). Another possible function is that every time provitamin D was converted to a photo product the organism recognized this as a photochemical signal that related to the amount of ultraviolet radiation it was exposed to. Provitamin D, which is structurally rigid and sandwiched in between the fatty acid side chains and polar head group [Tian et al., 1999] in the plasma membrane, has its ring opened upon exposure to ultraviolet radiation; this could have resulted in an alteration in membrane permeability to enhance the entrance of cations such as calcium into the cell.

Bills [1924] evaluated the vitamin D content of over 100 different species of fish and found a wide range of vitamin D activity from as high as 45,000 IU/g of oil in oriental tuna to less than 1 IU/g in gray sole liver oil and sturgeon liver oil. Bills [1927] was unable to demonstrate that when catfish were exposed to high intensity ultraviolet radiation they increased their antirachitic activity in their viseral oils. Sea water absorbs most of the high energy ultraviolet radiation within the first few meters and, therefore, if marine fish were to make vitamin D in the skin they would need to be exposed to sunlight near the surface of the sea. However, the question remained as to whether fish had the capacity to produce vitamin D in their skin. Fish skin was exposed to ultraviolet radiation

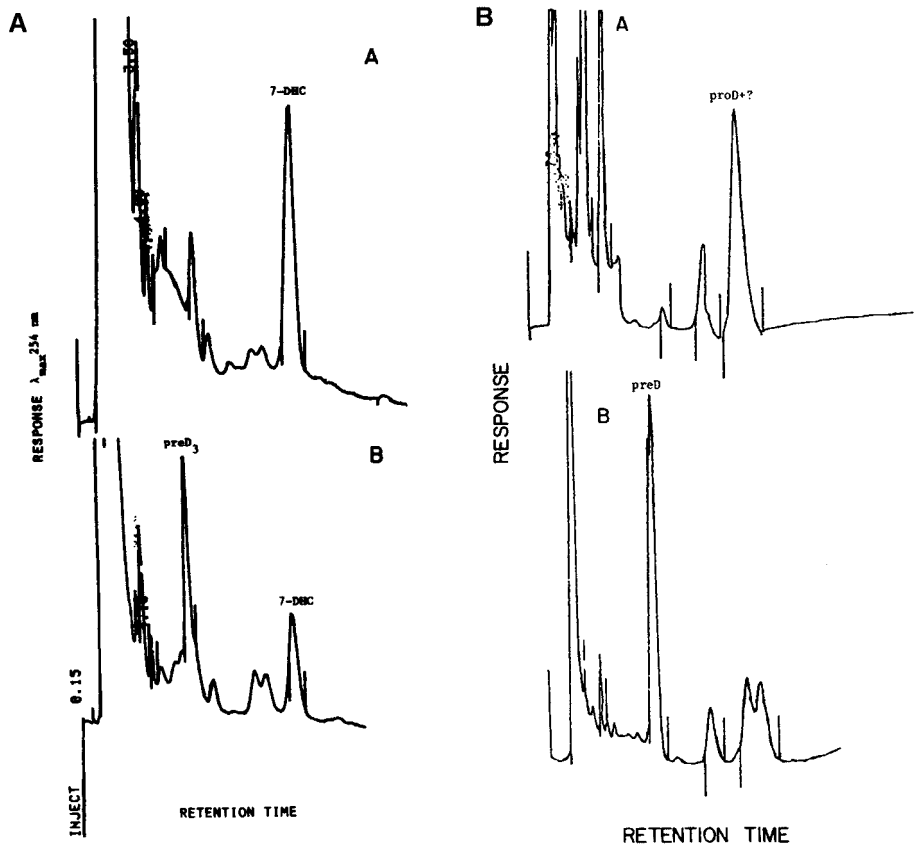

Fig. 2. **A**: High-pressure liquid chromatographic profiles of lipid extracts from *E. huxleyi* (A) shielded from UV radiation or (B) exposed to UV radiation. The chromatography was performed on a Radial-Pak-B column eluted with 8% ethyl acetate in *n*-hexane at 3.0 ml/min. (Reproduced with permission) [Holick, 1989]. **B**: High performance liquid chromatography profiles of lipid extracts from trout skin that was either (A) shielded from UV radiation or (B) exposed to simulated solar radiation. (Reproduced with permission) [Holick, 1989].

and an HPLC analysis revealed a presence of 7-dehydrocholesterol which upon UV irradiation converted to previtamin D_3 (Fig. 2B) [Holick et al., 1982a; Holick, 1989]. Analysis of skin from a variety of marine fish demonstrated the presence of several provitamin D's, including 7-dehydrocholesterol. Thus, like many mammals and humans, fish have provitamin D in their skin. However, what remains a mystery is that whereas fish probably obtain most of their vitamin D through the food chain, which includes phytoplankton that produces mainly vitamin D_2, the only vitamin D that has been identified in fish oils is vitamin D_3 [Bills, 1924, 1927; Holick, 1989]. We performed HPLC analyses on cod fish, flounder, red fish, mackerel, eel, and mullet liver oils and the only vitamin D identified was vitamin D_3. Whether fish only deposit vitamin D_3 in their livers and body fat or whether they have the capacity to convert vitamin D_2 to vitamin D_3 similar to paramecium is unclear [Holick, 1989].

HUMAN PHOTOBIOLOGY

In terms of human history, humans were not confronted with vitamin D deficiency until the Industrial Revolution began. Children who lived in the sunless, narrow alleyways developed severe growth retardation, widening of the ends of the long bones, and bowing and bending of the legs; clinical signs of severe rickets. By the late 1600s, rickets was recognized as a major health problem for young children. By the end of the 19th century it was estimated that upwards of 90% of children who lived in the industrialized cities of both North America and Europe had manifestation of rickets [Holick, 1994]. In addition to the obvious consequences of crippling effects of rickets, young women often had a

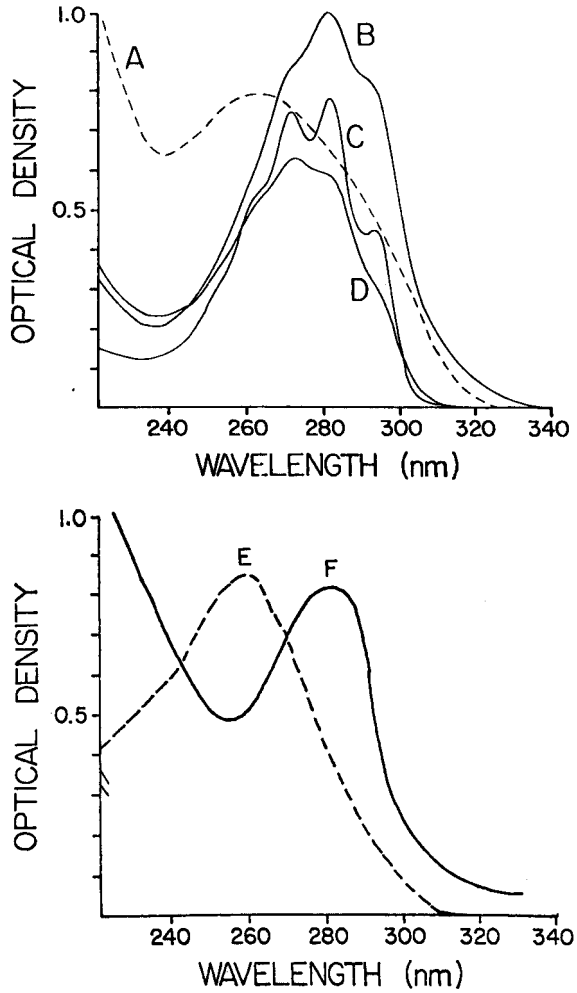

Fig. 3. Ultraviolet absorption spectra for (**A**) prD_3, (**B**) tachysterol, (**C**) provitamin D_3, (**D**) lumistserol, (**E**) DNA, and (**F**) albumin. (Reproduced with permission) [Holick, 1989].

deformed pelvis and had difficulty with birthing. Murdock Cameron was the first to perform Cesarean sectioning to deliver babies in women who could not deliver naturally because of a flattened pelvis. Cesarean birthing became very popular because of rickets.

As early as 1822, Sniadecki [1939] suggested the association with lack of sunlight as the cause of the high incidence rickets in Warsaw, Poland. In 1889, Palm [1890] noted that children living in third world countries had little risk of rickets, while children living in the industrialized cities in Europe were at high risk. He suggested that sun bathing was most important for the treatment and prevention of rickets. Both of these observations would fall on deaf ears until Huldschinsky [1919] reported that exposure to ultraviolet radiation from a mercury arc lamp resulted in the cure of rickets. This prompted Hess and Weinstock [1924] and Steenbock and Black [1924] to demonstrate that irradiation of a variety of substances, including grasses, corn, and olive oil resulted in them becoming antirachitic. This led Steenbock [1924] to recommend the irradiation of milk fortified with provitamin D_2 as a means of preventing and eradicating rickets. This ultimately led to the fortification of milk and other food products with synthetically made vitamin D_2 or vitamin D_3. Vitamin D fortification became so popular in the 1930s and 1940s that Bond bread, hot dogs, Twang soda, and even Schlitz beer was fortified with vitamin D.

During exposure to sunlight, radiation with wavelengths 290–315 nm (UVB) are absorbed by epidermal and dermal stores of 7-dehydrocholesterol [Holick et al., 1981]. This absorption of energy splits the B ring resulting in the formation of previtamin D_3 (Fig. 4) [Holick, 1994; Holick et al., 1995]. Once formed, previtamin D_3 exists in two conformeric forms *cis*, *cis* (czc) and *cis*, *trans* (czt). It is the czt that is the preferred conformer since it is more thermodynamically stable than its czc

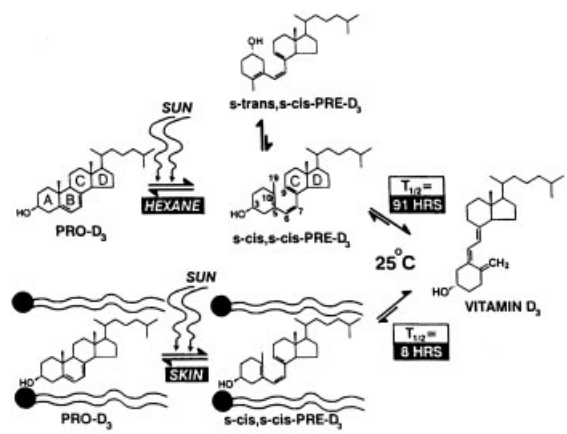

Fig. 4. Photolysis of provitamin D_3 (pro-D_3) into previtamin D_3 (pre-D_3) and its thermal isomerization to vitamin D_3 in hexane and in lizard skin. In hexane, pro-D_3 is photolyzed to s-*cis*, s-*cis*-pre-D_3. Once formed, this energetically unstable conformation undergoes a conformational change to the s-*trans*, s-*cis*-pre-D_3. Only the s-*cis*, s-*cis*-pre-D_3 can undergo thermal isomerization to vitamin D_3. The s-*cis*, s-*cis* conformer of pre-D_3 is stabilized in the phospholipids bilayer by hydrophilic interactions between the 3β-hydroxyl group and the polar head of the lipids, as well as by the van der Waals interactions between the steroid ring and side-chain structure and the hydrophobic tail of the lipids. These interactions significantly decrease the conversion of the s-*cis*, s-*cis* conformer to the s-*trans*, s-*cis* conformer, thereby facilitating the thermal isomerization of s-*cis*, s-*cis*-pre-D_3 to vitamin D_3. (Reproduced with permission) [Holick et al., 1995].

conformer. However, the thermodynamically less stable czc conformer is the only form of previtamin D_3 that can convert to vitamin D_3. Because 7-dehydrocholesterol is incorporated into the lipid bilayer, during exposure to ultraviolet radiation as the 7-dehyrocholesterol is converted to previtamin D_3, it is stabilized to remain only in the czc conformation, which rapidly converts to vitamin D_3 (Fig. 4) [Tian et al., 1994]. Once formed, the more stable vitamin D_3 is sterically unacceptable and is ejected from the plasma membrane into the extracellular space.

Ninety to 100% of most human being's vitamin D requirement comes from exposure to sunlight [Holick, 2002]. It is the UVB portion of the solar spectrum with wavelengths 290–315 nm that is responsible for the production of previtamin D_3 in human skin [Holick et al., 1981; MacLaughlin et al., 1982; Holick, 2002]. Thus, any alteration in the number of UVB photons reaching the epidermis can dramatically affect the cutaneous production of vitamin D_3. Increased skin pigmentation can reduce the production of vitamin D_3 by as much as 50-fold [Clemens et al., 1982]. The application of a sunscreen with a sun protection factor (SPF) of only 8 reduces the UVB penetration into the epidermis by 97.5%, thereby reducing the production of previtamin D_3 by the same amount [Matsuoka et al., 1987]. An increase in the zenith angle of the sun results in more of the UVB photons being absorbed by the stratospheric ozone layer. When the zenith angle of the sun becomes so oblique that very few UVB photons can penetrate to the earth's surface; this results in little, if any, cutaneous production of vitamin D_3. This is the explanation for why during the winter little, if any, vitamin D_3 is produced in the skin at latitudes above and below 35°N and 35°S [Webb et al., 1988]. Time of day, season of the year, latitude, and altitude all markedly affect the cutaneous production of vitamin D_3 [Holick, 2002] (Fig. 5). As the skin ages, there is a decline in the cutaneous levels of 7-dehydrocholesterol. This is why aging can markedly reduce the skin's capacity to produce vitamin D_3 [MacLaughlin and Holick, 1985a]. However, despite the up to fourfold reduction in vitamin D_3 production in a 70-year-old compared to a 20-year-old, the skin has such a high capacity to make vitamin D_3, elders exposed to sunlight will produce an adequate amount of vitamin D to satisfy their vitamin D requirement [Reid et al., 1985; Holick, 1994, 2002; Chel et al., 1998; Chuck et al., 2001].

VITAMIN D DEFICIENCY PAST, PRESENT, AND CONSEQUENCES

Vitamin D deficiency in children was epidemic in most industrialized cities throughout Northern Europe and the United States by the end of the 19th century. The appreciation of the role of vitamin D and sunlight in the prevention and cure of rickets has made a major impact on eradicating rickets as a major health concern for young children. However, rickets is making a resurgence in African American young children who receive their total nutrition from breast feeding [Kreiter et al., 2000]. What is not well appreciated is that vitamin D deficiency is common in otherwise healthy, young [Outila et al., 2001], middle aged, and older adults [Chapuy et al., 1997; Karimi et al., 1998; Malabanan et al., 1998; Kauppinen-Makeline et al., 2001; Lips, 2001]. A recent survey of women of child bearing age in the United States revealed that at the end of the winter 41% of African American women aged at 15–49 years and 4% of Caucasian women at the end of the summer were found to be vitamin D deficient [Nesby-O'Dell et al., 2002]. Indeed, vitamin D deficiency is extremely common even among active, young adults. Tangpricha et al. [2002] reported 36% of medical personnel aged 18–29 years were vitamin D insufficient at the end of the winter in Boston.

Vitamin D deficiency causes a mineralization defect in the adult's skeleton resulting in osteomalacia. The associated secondary hyperparathyroidism causes an increase in the mobilization of the matrix and mineral from the skeleton that can increase risk or precipitate osteoporosis [Holick, 2002]. Osteomalacia is not only associated with mineralization defect of the skeleton, but is also associated with isolated or global bone pain, muscle weakness, and muscle pain which are symptoms that often go undiagnosed or misdiagnosed as some type of collagen vascular disease, such as fibromyalgia [Glerup et al., 2000; Semba et al., 2001; Holick, 2002]. There is mounting evidence that there is a latitudinal association, i.e., living at a higher latitude increases the risk of hypertension [Rostand, 1979; Krause et al., 1998], various common cancers such as colon, breast, prostate, and ovarian [Garland et al., 1989; Garland et al., 1990;

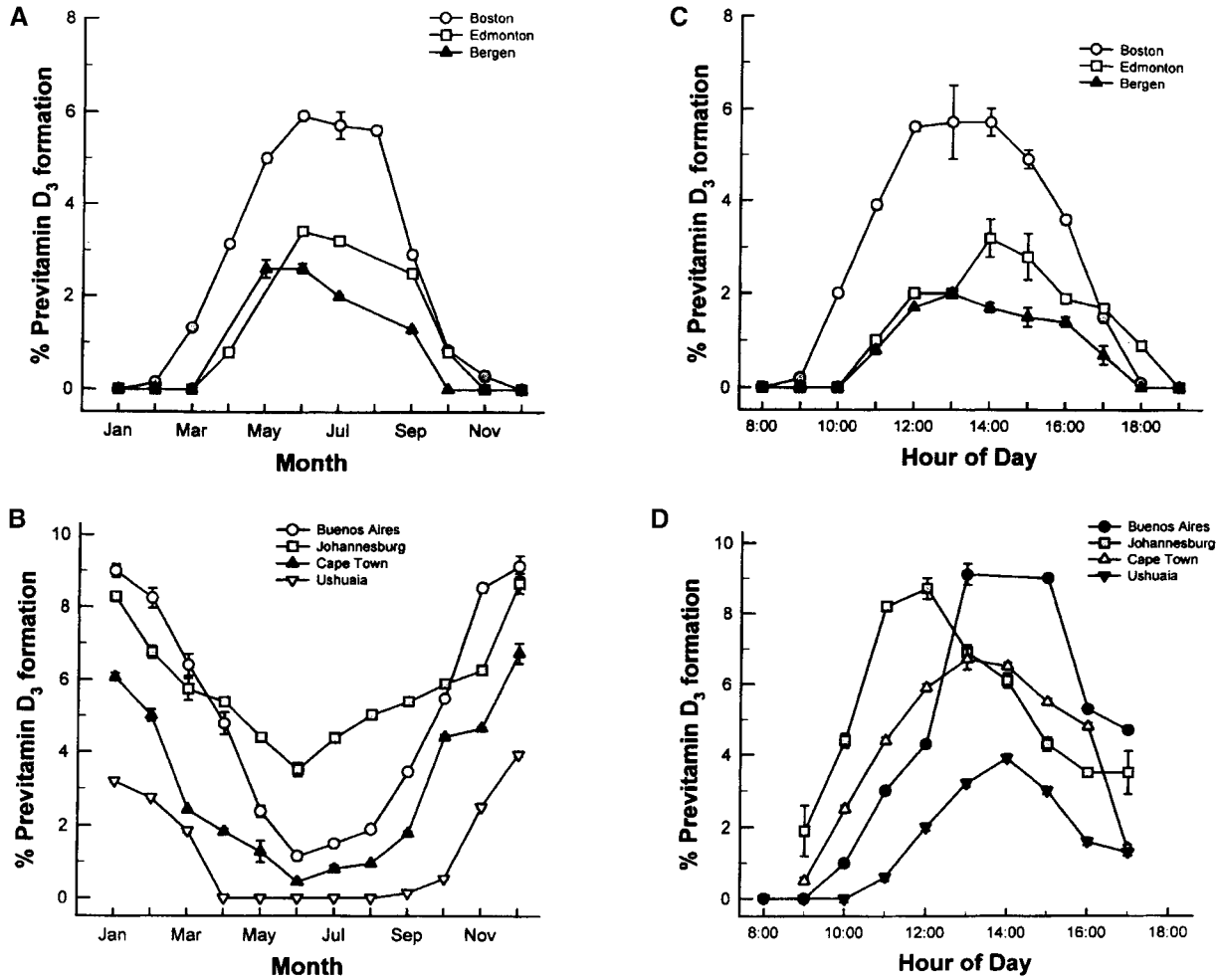

Fig. 5. Influence of season, time of day, and latitude on the synthesis of previtamin D_3 in Northern (**A** and **C**) and Southern hemispheres (**B** and **D**). The hour indicated in C and D is the end of the 1 h exposure time. (Reproduced with permission) [Chen, 1998].

Garland et al., 1992; Hanchette and Schwartz, 1992; Ahonen et al., 2000], and autoimmune disorders including multiple sclerosis [Cantorna et al., 1996] and type 1 diabetes [Mathieu et al., 1999; Hypponen et al., 2001]. It appears that this latitudinal association is due to sunlight mediated vitamin D_3 synthesis [Holick, 2002].

METABOLISM OF VITAMIN D

In the mid 1960s, it was known that when vitamin D was given to vitamin D deficient rats, it took 24 h before maximum intestinal calcium absorption was observed. This led to the belief that vitamin D needed to be activated before it could carry out its physiologic functions on calcium metabolism. In the late 1960s, pigs were given pharmacologic doses of vitamin D_3. The blood was collected and a lipid extraction followed by several chromatographies yielded a vitamin D metabolite that was more polar than vitamin D and it was structurally identified as the 25-hydroxyvitamin D_3 [25(OH)D_3] [Deluca, 1979]. 25(OH)D_3 was about fourfold more potent than vitamin D in stimulating intestinal calcium transport and it only took 12 h for 25(OH)D_3 to maximize intestinal calcium absorption in vitamin D deficient rats.

It was suspected that 25(OH)D was also an intermediate and that it required further metabolism to become active. In 1970, three laboratories (Kodicek, Norman, and DeLuca) independently initiated a quest to identify the biologically active form of vitamin D_3. Originally it was believed that 25(OH)D_3 was metabolized in the small intestine to its activated

form. Using a similar strategy that was used to identify 25(OH)D_3, DeLuca's laboratory fed eight pigs pharmacologic doses of vitamin D_3. They were sacrificed and their small intestines were recovered. An intensive purification of the lipid extract from the pig intestines did not yield any significant amount of identifiable vitamin D metabolite. The reason that the intestine was chosen to identify the active form of vitamin D_3 was based on the fact that the intestine appeared to have the highest concentration of the metabolite, and increasing vitamin D intake increased the amount of apparent activated vitamin D_3 in the small intestine. However, the development of Sephadex LH-20 chromatography revealed that the single more polar peak identified by silicic acid chromatography and labeled as peak V contained at least five different metabolites [Holick and DeLuca, 1971b]. It was realized that only when a physiologic amount of vitamin D_3 was given to vitamin D deficient chickens that the intestine achieved the maximum amount of the desired activated vitamin D metabolite without other interfering metabolites. This led to the idea of obtaining a large number of small intestines from normal chickens. Four hundred pounds of small intestine were collected from a local chicken slaughter house, where 20,000 chickens were slaughtered daily. The intestines were extracted in several hundred gallons of chloroform and methanol and the lipid extract was exhaustively chromatographed. However, the lipid contamination was so extensive and because it was unclear whether any activated vitamin D_3 metabolite was present, the project was abandoned. I had suggested that the only way of knowing how much activated vitamin D_3 there was for isolation purposes was to give vitamin D deficient chickens ^{3}H-vitamin D_3 with a known specific activity. Based on previous observations, it was estimated that it would require 1,500 chicken intestines to obtain between 10 and 15 μg of activated vitamin D_3. Three batches of 500 chickens were made vitamin D deficient at 4 week intervals. Once vitamin D deficient, each chicken received, by Jack Omdahl in wing vein, 10 IU of ^{3}H-vitamin D_3. Twenty four hours later the animals were sacrificed and the small intestine was collected. Based on the specific activity of the ^{3}H-vitamin D_3, it was determined that ~10 μg of activated metabolite was present in the lipid extract that contained about 30 g of yellow viscous oil. The lipid extract was chromatographed on various Sephadex LH 20 columns and Bio beads SX-8 columns for a total of 17 chromatographies yielding 8 μg of active metabolite in over 1 mg of lipid [Holick et al., 1971a,b].

A strategy was developed to purify the metabolite from the overwhelming amount of contaminating lipids. The entire lipid extract was derivatized by trimethylsiliation (TMS). The trimethylsilated material was then treated with diluted hydrochloric acid for a short period of time with the intention of removing the TMS derivatives on primary and secondary hydroxyls leaving the tertiary hydroxyl on carbon 25 derivatized. This would afford the ability to separate it from other contaminating lipids by Sephadex LH20 chromatography. This resulted in isolation of 2 μg of a 25-hydroxy TMS derivative of the metabolite. A mass spectroscopy analysis revealed that the metabolite contained a 1-hydroxyl function and a 25-TMS derivative proving that the structure was 1,25-dihydroxyvitamin D_3 [1,25(OH_2D_3)]. The 25-hydroxy TMS was removed by acid hydrolysis and the parent compound was recovered. Approximately 100–500 ng of the purified metabolite was derivatized by acetylation, TMS, and by hydrogen reduction and the mass spectrum of the metabolite derivatives confirmed the structure as 1,25$(OH)_2D_3$. The 1-hydroxyl was established by demonstrating that this allylic hydroxyl could be eliminated by reduction with hydrogen [Holick et al., 1992].

Once the structure was identified, a major effort was made to chemically synthesize 1,25$(OH)_2D_3$. It took 2 years and a 21 step synthesis, starting with a kilogram of starting material and yielding less than 1 mg of final product [Semmler et al., 1972]. However, the stage was set for the chemistry community to modify the synthesis to a stage where it was commercially developed by Dr. Milan Uskovick at Hoffman LaRoche. 1,25$(OH)_2D_3$ and its synthetic analog 1α-hydroxyvitamin D_3, which was initially made during the synthesis of 1,25$(OH)_2D_3$ [Holick et al., 1973], were developed commercially for the treatment of renal osteodystrophy, hypoparathyroidism, and osteoporosis.

NOVEL FUNCTIONS OF 1,25-DIHYDROXYVITAMIN D_3

Until the late 1970s, it was believed that 1,25$(OH)_2D_3$ had biologic functions only on

calcium and phosphate metabolism. The $1,25(OH)_2D_3$ receptor (vitamin D receptor; VDR) was identified in the nuclei of the small intestine, osteoblasts, and kidney cells. $1,25(OH)_2D_3$ was shown to rapidly increase intestinal calcium absorption, mobilize preosteoclasts to become mature osteoclasts to mobilize calcium stores from the skeleton, and to increase the efficiency of phosphate absorption in the jejunum and ilium [Holick, 2002].

Stumpf et al. [1979] reported that most tissues in the body of vitamin D deficient rats concentrated ^3H-$1,25(OH)_2D_3$ in their nuclei. This suggested that many other tissues in the body had the ability to recognize $1,25(OH)_2D_3$. Tanaka et al. [1982] reported that preleukemic mouse cells that had a VDR showed marked inhibition of growth and enhanced maturation in the presence of $1,25(OH)_2D_3$. This followed with several observations demonstrating that breast cancer cells, osteosarcoma cells, melanoma cells responded to the anitproliferative activity of $1,25(OH)_2D_3$ [Holick, 2002].

Of great curiosity was that the skin was not only the organ responsible for making vitamin D_3, but was also a target tissue for $1,25(OH)_2D_3$ since keratinocytes had a VDR. Smith et al. [1986] reported that human cultured keratinocytes exposed to $1,25(OH)_2D_3$ showed marked inhibition of growth and accelerated maturation. This led to the concept of using $1,25(OH)_2D_3$ as a treatment for the hyperproliferative skin disorder, psoriasis [MacLaughlin et al., 1985b]. Activated vitamin D compounds, including calcipotriene, 1,24-dihydroxyvitamin D_3, and $1,25(OH)_2D_3$ have been developed by pharmaceutical companies and are considered the first line of treatment for psoriasis [Holick et al., 1987; Holick, 1998].

NONRENAL SYNTHESIS OF 1,25-DIHYDROXYVITAMIN D_3 AND ITS IMPLICATIONS FOR HUMAN HEALTH AND DISEASE

The cloning of the 25-hydroxyvitamin D-1α-hydroxylase (1-OHase) [Kitanaka et al., 1998]

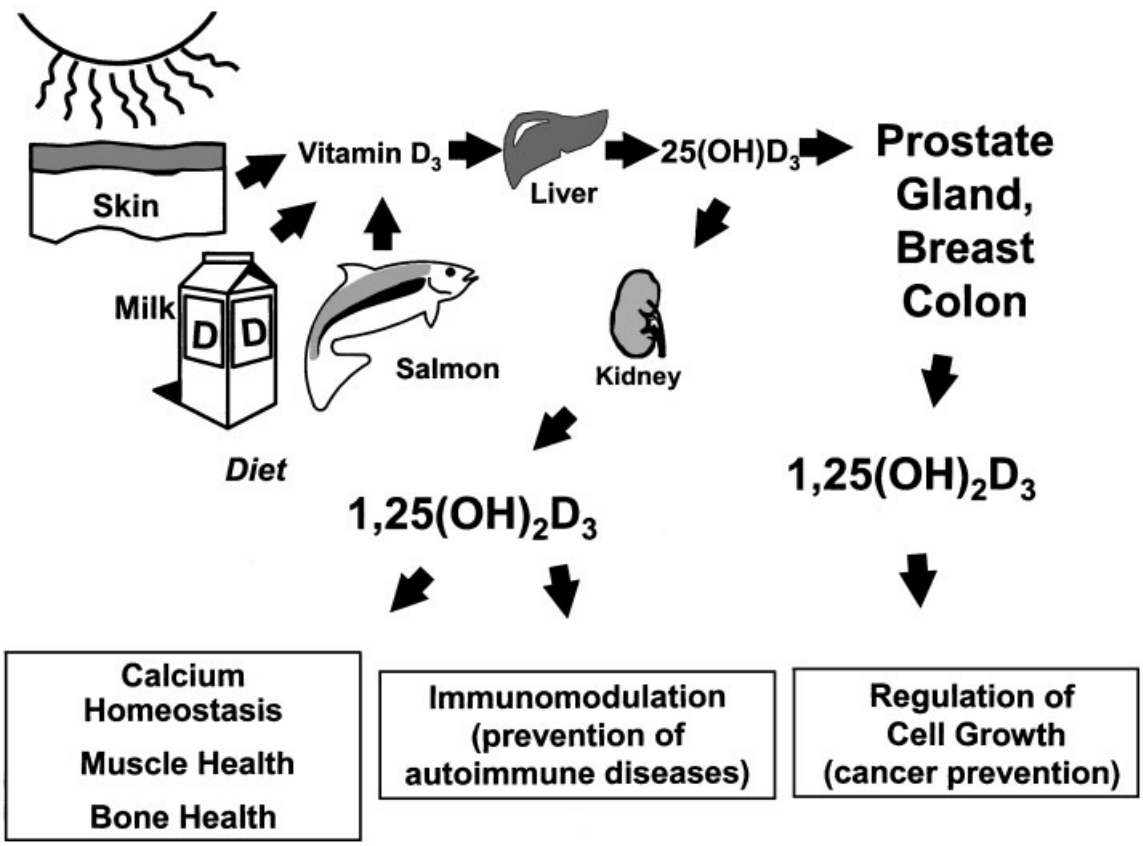

Fig. 6. Production, metabolism and biologic functions of vitamin D_3.

provided the impetus to explore the possibility that tissues other than the kidney could produce $1,25(OH)_2D_3$. It is now recognized that many tissues in the body, including prostate, colon, skin, and osteoblasts have the capacity to express the 1-OHase and thereby synthesize $1,25(OH)_2D_3$ locally [Schwartz et al., 1998; Cross et al., 2001; Tangpricha et al., 2001; Holick, 2002].

Although the physiologic function of the extra renal production of $1,25(OH)_2D_3$ is not well understood, there is mounting evidence that this synthesis may be important for cellular health. Since the 1940s, it has been recognized that people who live at higher latitudes have a higher risk of dying of the most common cancers, including colon, breast, and prostate [Apperly, 1941]. It is also known that normal adults with a 25(OH)D of at least 20 ng/ml have a 50% decreased risk of developing colon cancer [Garland et al., 1989]. It has been speculated that the local production of $1,25(OH)_2D_3$ may be for the purpose of regulating cell growth, which may ultimately decrease risk of developing cancers in these tissues [Holick, 2002] (Fig. 6).

It is recognized that activated T and B lymphocytes as well as monocytes have a VDR and $1,25(OH)_2D_3$ is an effective immune modulator [Manolagas et al., 1985] (Fig. 6). In animal studies, it has been demonstrated that $1,25(OH)_2D_3$ pretreatment can markedly reduce the incidence of type 1 diabetes in NOD mice [Mathieu et al., 1999], inhibit progression of arthritis in mice [Cantoma et al., 1998], and multiple sclerosis-like syndrome in the mice that received myelin [Cantorna et al., 1996]. This may be the explanation for why children who received 2,000 IU of vitamin D daily after the age of 1 year reduced their risk of type 1 diabetes by 80% [Hypponen et al., 2001] and why people who live closer to the equator and presumably make more vitamin D_3 are at decreased risk of developing common autoimmune diseases.

The association of hypertension in people living at higher latitudes has also been associated with vitamin D deficiency [Rostand, 1979]. It is now recognized that $1,25(OH)_2D_3$ is a negative regulator of the renin–angiotension system [Chun et al., 2002]. This could explain, in part, why African Americans who are chronically vitamin D deficient [Harris et al., 2001] also have a higher risk of hypertension and cardiovascular disease [Fahrleitner et al., 2002].

CONCLUSIONS

It is remarkable that humans evolved in a manner, whereby, they depended on sunlight for an essential hormone that was not only responsible for guaranteeing skeletal health, but also played a major role in a wide variety of other organ systems. It is truly amazing that in the 21st century with all of the advances of modern medicine, that vitamin D deficiency has made a resurgence not only in breast fed infants, but also in young, middle aged, and older adults. Vitamin D deficiency and its consequences are extremely subtle, but have enormous implications for human health and disease. It is for this reason that vitamin D deficiency continues to go unrecognized by a majority of health care professionals. There needs to be a program to educate the public at large that not only should they be caring about their blood cholesterol levels, but they should also be aware of their vitamin D status, i.e., 25-hydroxyvitamin D levels.

The lower limit of the normal range for 25(OH)D is totally inadequate. There is strong scientific evidence that demonstrates that 25(OH)D of at least 20 ng/ml is required to maintain calcium homeostasis without developing secondary hyperparathyroidism [Malabanan et al., 1998]. It has also been suggested that a level of 30 ng/ml is not only important for maximum bone health, but may also be important for maintaining cellular health [Holick, 2002].

Sunlight is by far the best and most reliable source of vitamin D for most humans. Exposure of the body, in a bathing suit, to one minimal erythemal dose (MED; i.e., slight redness of the skin) is equivalent to taking between 10,000 and 25,000 IU of vitamin D orally. Therefore, exposure of hands, face, arms, and legs to sunlight to an amount of time equal to about 25% of what it would take to develop a mid sunburn, i.e., one MED, two to three times a week is more than adequate to satisfy the bodies vitamin D requirement and enough to store some vitamin D_3 in the body fat. After this exposure, a sunscreen with an SPF 15 or greater can be applied if the person wishes to remain outdoors. This will afford the individual to take advantage of the beneficial effect of sunlight while preventing the damaging consequences due to excessive exposure to sunlight.

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