Vitamin D Status in Psoriasis Patients Treated with UVB Therapy

Amra Osmančević

Department of Dermatology and Venereology
Sahlgrenska University Hospital
Institute of Clinical Sciences at
Sahlgrenska Academy
Göteborg, Sweden
2009

UNIVERSITY OF GOTHENBURG

Title: Vitamin D status in psoriasis patients treated with UVB therapy

Author: Amra Osmančević, MD

E-mail:Amra.Osmancevic@vgregion.se

Department of Dermatology and Venereology, Sahlgrenska University Hospital, Institute of Clinical Sciences at Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden

Cover:

The cover picture illustrates the sun, the skin compartments (stratum basale, stratum spinosum, stratum granulosum and stratum corneum) and the chemical structure of vitamin D3. Ultraviolet B radiation stimulates the production of vitamin D3 in stratum basale.

The Digital version can be downloaded from
http://hdl.handle.net/2077/19041

Printed by Geson Hylte Tryck AB, Kungsbacka, Sweden 2009

Mojim roditeljima

To my parents Magbula and Aziz Ahmic

"Nasi daleki preci su vjerovali da zivot treba ispuniti sa tri dobra djela, od djeteta covjeka podici, kucu sagraditi i napisati knjigu zivota..." "Zagubljene slike", Sevko Kadric

"Our old ancestors believed that life could only be fulfilled through the completion of three good deeds: bringing up one's child to adulthood, building a house, and writing a book on life..." "Borttappade bilder", Sevko Kadric

Vitamin D status in psoriasis patients treated with UVB therapy

Amra Osmancevic
Department of Dermatology and Venereology, Institute of Clinical Sciences at Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden

Abstract

The thesis deals with the effect of ultraviolet B (UVB) 280-320 nm on vitamin D production in psoriasis patients during treatment with phototherapy.

Background: Psoriasis is a chronic, inflammatory disease affecting the skin and potentially the joints. Both genetic and environmental factors are important in the aetiology of the disease. Phototherapy (broadband UVB, narrowband UVB (NBUVB) and heliotherapy) is commonly used as treatment of psoriasis.

Vitamin D3, or cholecalciferol, is produced in the basal epidermis by ultraviolet radiation (290-315 nm) of 7-dehydrocholesterol and hydroxylated in the liver to the major circulating metabolite 25-hydroxyvitamin D [25(OH)D]. Hydroxylation to 1,25-dihydroxyvitamin D [$1,25(OH)_2D$] in the kidneys is stimulated by parathyroid hormone (PTH) and suppressed by phosphate. Sun exposure is the strongest factor influencing 25(OH)D.

Aims: 1) To study the effect of UVB on vitamin D synthesis in patients with psoriasis. 2) To examine possible differences between NBUVB and broadband UVB on vitamin D production in psoriatic patients. 3) To investigate the effect of UVB induced vitamin D on bone, lipid and carbohydrate status in psoriasis patients.

Methods: Serum 25(OH)D, $1,25(OH)_2D$, PTH, calcium and creatinine were measured before and after the phototherapy in white, Caucasian patients with active plaque psoriasis. Bone mineral density (BMD) was examined using Dual-Energy X-ray Absorptiometry (DEXA) in postmenopausal women with psoriasis. Lipid and carbohydrate status were assessed in patients treated with heliotherapy.

Results: Psoriasis improved in all patients, with a 75% reduction in PASI (Psoriasis Area and Severity Index) score on all regimes. Serum 25(OH)D increased and PTH decreased after phototherapy. The increase in 25(OH)D was higher in the broadband treated patients compared with NBUVB. There was no correlation between the dose of UVB and the increase of 25(OH)D. Postmenopausal women with psoriasis had higher BMD both at the hip and at the lumbar spine than age-matched controls. The ratio of low-density lipoprotein (LDL) and high-density lipoprotein cholesterol (HDL), and the levels of glycosylated haemoglobin A_1c (HbA_1c) decreased during heliotherapy.

Conclusion: UVB and heliotherapy increased the serum 25(OH)D production, reduced the serum PTH concentrations and improved psoriasis, lipid and carbohydrate status in the patients. Vitamin D production in psoriasis patients increased less with NBUVB than with broadband UVB phototherapy. Postmenopausal women with psoriasis had higher BMD than age-matched controls, a finding that could be related to their higher body weight, physical activity and the UVB exposure.

Key words: Vitamin D, PTH, psoriasis, bone mineral density, ultraviolet UVB
ISBN-978-91-628-7682-1
Göteborg 2009

"Vitamin D under ljusbehandling hos patienter med psoriasis"

Amra Osmancevic, Hudkliniken, Sahlgrenska Universitetssjukhuset

Svensk sammanfattning

Ultraviolett B (UVB) ljus respektive solljus under klimatvård på Gran Canaria förbättrar psoriasis och höjer vitamin D nivåerna i blodet. Vitamin D produktionen hos patienter med psoriasis ökar mer under behandling med bredband UVB (280-320 nm) än med smalspektrum UVB, så kallat TL01 (311 nm).

Undersökning av en grupp postmenopausala kvinnor med psoriasis visade att de hade bättre bentäthet än jämnåriga kvinnor från Göteborg. Detta kan delvis bero på att kvinnorna med psoriasis hade högre kroppsvikt, fysisk aktivitet och positiv effekt av UVB på vitamin D nivåerna.

Behandling med solljus under s.k. klimatvård på Gran Canaria hade gynnsam effekt på psoriasis, ökade vitamin D och förbättrade blodfetterna samt sockeromsättningen hos dessa patienter.

Psoriasispatienter med låga nivåer av vitamin D i blodet (<30 ng/ml) före behandlingsstart, ökade mer i sina D-vitaminvärden efter ljusbehandlingarna än dem som hade högre halter av D-vitamin vid start. Alla patienter gick upp i sina D-vitaminvärden oavsett ålder, hudtyp eller svårighetsgrad av psoriasis. Effekt av solljus på vitamin D under 2 veckors klimatvård på Gran Canaria kan jämföras med effekten av behandling med UVB lampa 2-3 gånger/vecka under 2 till 3 månaders tid.

UVB och solljus ökade vitamin D i blodet, förbättrade psoriasis, var associerat med bättre benmassa, och hade positiv effekt på lipider och sockeromsättning.

Vitamin D status in psoriasis patients treated with UVB therapy

List of papers

This thesis is based on the following papers, which will be referred to in the text by their Roman numerals I-IV:

I Osmancevic A, Landin-Wilhelmsen K, Larkö O, Mellström D, Wennberg AM, Hulthén L, Krogstad AL. UVB therapy increases 25(OH) vitamin D synthesis in postmenopausal women with psoriasis. *Photodermatol Photoimmunol Photomed* 2007; 23(5): 172-8.

II Osmancevic A, Landin-Wilhelmsen K, Larkö O*,* Mellström D, Wennberg AM, Hulthén L, Krogstad AL. Risk factors for osteoporosis and bone status in postmenopausal women with psoriasis treated with UVB therapy. *Acta Derm Venereol.* 2008; 88(3):240-6.

III Osmancevic A, Landin-Wilhelmsen K, Larkö O, Wennberg AM, Krogstad AL.Vitamin D production in psoriasis patients increases less with narrowband than with broadband ultraviolet B phototherapy. *Photodermatol Photoimmunol Photomed*, 2009 (in press).

IV Osmancevic A, Nilsen LT, Landin-Wilhelmsen K, Søyland E, Abusdal Torjesen P, Hagve TA, Nenseter M, Krogstad AL. Effect of climate therapy at Gran Canaria on vitamin D production, blood glucose and lipids in patients with psoriasis. *J Eur Acad Dermatol Venereol*, 2009 (in press).

Contents

Abbreviations

7-DHC	7- dehydrocholesterol
D3	cholecalciferol
25(OH)D	25-hydroxyvitamin D (or calcidiol)
1,25(OH)$_2$D	1,25-dihydroxyvitamin D (or calcitriol)
PTH	parathyroid hormone
VDR	vitamin D receptor
VDP	vitamin D binding protein
UVR	ultraviolet radiation
UVB	ultraviolet radiation B
UVA	ultraviolet radiation A
NBUVB	narrowband ultraviolet radiation B
PASI	Psoriasis Area and Severity Index
HRT	hormonal replacement therapy
DEXA	Dual-Energy X-ray Absorptiometry
BMD	bone mineral density
BMI	body mass index
LDL	low density lipoprotein cholesterol
HDL	high density lipoprotein cholesterol
TNF-α	tumour necrosis factor-α
HbA$_1$c	haemoglobin A$_1$c
PUVA	Psoarlen+UVA
MED	Minimal Erythema Dose
SED	Standard Erythema Dose

Introduction

Vitamins

William Fletcher was the first scientist who in 1905 determined that diseases occurred if special factors (vitamins) were removed from food. He was researching the causes of the disease beriberi when he discovered that eating unpolished rice prevented beriberi and eating polished rice did not. William Fletcher believed that there were special nutrients contained in the husk of the rice.

At the same time biochemist Frederick Gowland Hopkins discovered that certain factors in food were important to health. In 1912, Polish scientist Cashmir Funk named the special nutritional parts of food as a "vitamine" after "vita" meaning life and "amine" from compounds found in the thiamine he isolated from rice husks. Vitamine was later shortened to vitamin.

Vitamins B, C and D were all discovered as a result of research into diseases - beriberi, scurvy and rickets, respectively. Supplementing an imbalanced diet with certain foods was shown to prevent each of these diseases. Subsequently, purification and analysis of disease-preventing compounds in these foods established a new class of nutrients - in addition to proteins, fats and carbohydrates - that are now known to be essential for human health.

Vitamin D

Nomenclature

Cholecalciferol or vitamin D3 is produced in the skin as a result of ultraviolet irradiation of 7-dehydrocholesterol (7-DHC). Ergocalciferol or vitamin D2 is produced by ultraviolet irradiation of the plant sterol ergosterol. “Calciferol” refers to both of these compounds.

Structure

The two forms of vitamin D (D2 and D3) differ chemically in their side chains. These structural differences also alter their metabolism, but in general, the biological activity of their active metabolites is comparable.[1]

The only structural difference between vitamin D2 and D3 is in their side chains. The side chain for vitamin D2 contains a double bond between C-22 and C-23 and a C-24 methyl group *(Figure 1)*.

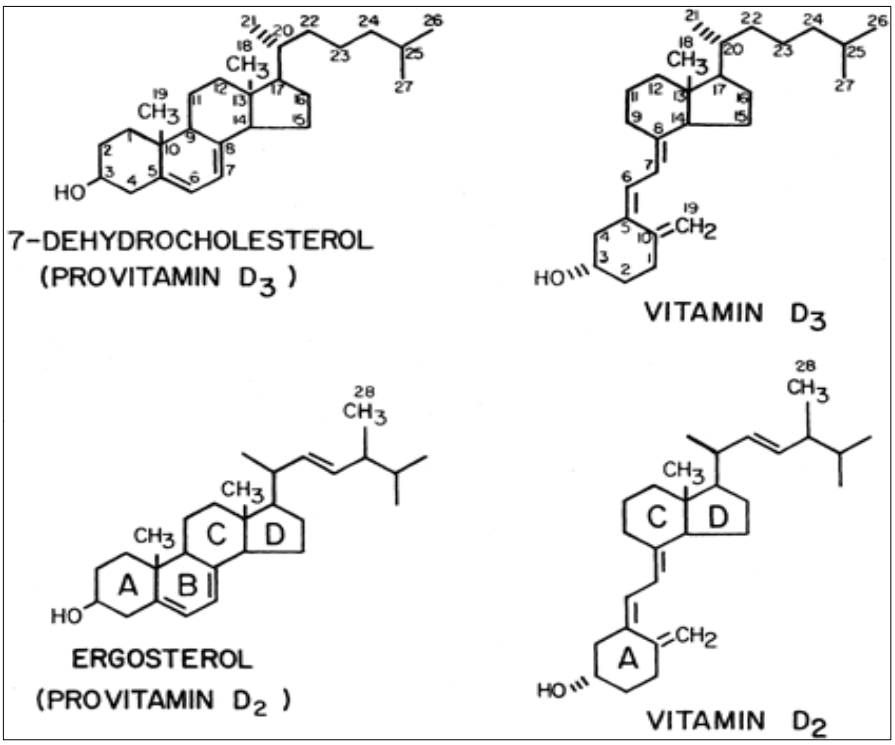

Figure 1: Structure of vitamin D3 and D2 and their respective precursors, 7-dehydrocholesterol, and ergosterol (MacLaughlin and Holick, 1983).

Historical perspective

Sunshine as a means of health

The discovery of vitamin D in food and the realization that the body can produce vitamin D independently are both intimately tied to research into the cause and prevention of childhood rickets.

There has been some evidence since early Greek and Roman times of the bone-deforming disease commonly known as rickets. The first scientific description of a vitamin D deficiency, namely rickets, was provided in the 17th century by both Dr. Daniel Whistler (1645) and Professor Francis Glisson (1650) *(Figure 2).*

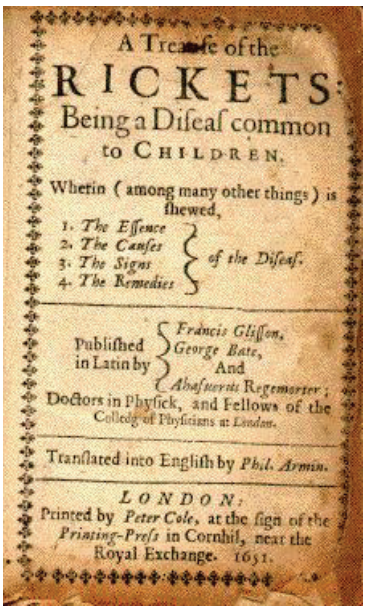
A Treatise of the
RICKETS:
Being a Diseas common
to CHILDREN.
Wherin (among many other things) is
shewed,
1. The Essence
2. The Causes
3. The Signs
4. The Remedies
of the Diseas.
Published in Latin by Francis Glisson, George Bate, And Abasuerus Regemorter; Doctors in Physick, and Fellows of the College of Physitians at London.
Translated into English by Phil. Armin.
LONDON:
Printed by Peter Cole, at the sign of the Printing-Press in Cornhil, near the Royal Exchange. 1651.

Figure 2: D.Whistler; F.Glisson. *A Treatise of the Rickets: Being a Disease Common to Children.* London: P. Cole, 1651

Rickets is a disease of young children with a constellation of physical signs and symptoms including deformities of the skeleton, such as bowed legs, enlargement

of the epiphyses of the long bones and rib cage (rachitic rosary), deformed pelvis, enlarged head, curvature of the spine, poor dentition, and weak flabby legs.[2] During the Industrial Revolution in Europe children developed this disease as a result of a sunless environment in the polluted inner cites. An autopsy study of children who died of various causes in Leyden, The Netherlands, in the end of the 18th century, revealed that 80-90% of these children had evidence of rickets.

In 1822 Sniadecki discovered the importance of exposure to sunlight for the prevention and cure of rickets. He observed that children living in the inner city of Warsaw, Poland, had a higher incidence of rickets than children living in rural areas. He encouraged direct exposure of the skin to sunlight as one most efficient methods for the prevention and cure of rickets.[3]

Little attention, however, was given to the environment as a cause of rickets until 1889, when a British Medical Association investigative committee reported that rickets was infrequently seen in rural districts of the British Isles but was prevalent in large industrial towns.[4] In 1890 Palm collected clinical observations from a number of his colleagues throughout the British Empire and the Orient. He found that rickets was widespread in the industrial centres of Great Britain, whereas in impoverished cities in China, Japan, and India, where people received poor nutrition and lived in squalor, the disease was rare.[5] Based on this epidemiological survey he urged "the systematic use of sunbaths as a preventive and therapeutic measure in rickets and other diseases, and the education of the public to the appreciation of sunshine as a means of health." However, it was difficult at the time for people to believe that such a simple remedy as exposure to sunlight could cure this bone-deforming disease, and little was done to use these astute observations for curing rickets.

The fat-soluble vitamin

In the early 1800s it was a common practice, supported by folklore, to give children cod liver oil to prevent and cure rickets. In the 1890s scientists began to search for specific foods that could prevent rickets. Such reasoning was rooted in the knowledge that two other diseases, scurvy and beriberi, could be prevented by the addition of certain foods (such as citrus fruits that contain vitamin C, and whole grain rice that contains vitamin B_1) to the diet. However, it was not until 1918 when Mellanby reported that he could make beagles rachitic and reverse the bone disease

with cod liver oil that the scientific community began to consider rickets as a nutritional-deficiency disease. In 1921 he wrote, "The action of fats in rickets is due to a vitamin or accessory food factor which they contain, probably identical with the fat-soluble vitamin." Originally it was thought that the antirachitic factor in cod liver oil was vitamin A. However, McColum et al. demonstrated that the antirachitic activity that he called vitamin D was separate from vitamin A. They exposed cod liver oil to heat and oxygen - vitamin A activity was destroyed while when maintaining the antirachitic activity. The vitamin in cod liver oil was designated "D" as vitamins A, B, and C had already been identified.

Light equals vitamin D

At the same time that Mellanby was demonstrating that rickets could be cured by the ingestion of cod liver oil, Huldschinsky was exposing rachitic children to radiation from a mercury arc lamp. He reported dramatic reversal of rickets within four months after ultraviolet radiation therapy from the mercury arc lamp. In 1921 Hess and Unger exposed seven rachitic children in New York City to sunshine and reported, by X-ray examination, that there was marked improvement in each child's rickets. This research had equivocally shown that exposure to sunlight alone could prevent and cure this crippling bone disease. This led Hess and Weinstock to investigate independently the use of ultraviolet irradiation of food (including wheat, lettuce, vegetable oils and animal feed) as a way of imparting antirachitic activity.[6] Steenbock appreciated the use of this technique and introduced the concept of irradiating milk as a means of preventing rickets in children.[7] This led to the fortification of milk with vitamin D, which helped eradicate rickets in the countries that used this practice.

The chemical structures of the D vitamins were determined in the 1930s in Professor A. Windaus's laboratory at the University of Göttingen in Germany. Vitamin D was first isolated from the irradiation of the fungal sterol ergosterol and was designated vitamin D1. However, the product was an impure mixture, and the term was dropped. Vitamin D2, which could be produced by ultraviolet irradiation of ergosterol, was chemically characterized in 1932. Vitamin D3 was not chemically characterized until 1936 when it was shown to result from the ultraviolet irradiation of 7-DHC. Almost simultaneously, the elusive antirachitic component of cod liver oil was shown to be identical to the characterized vitamin D3. These results clearly

established that the antirachitic substance vitamin D was chemically a steroid, more specifically a seco-steroid (*Figure 1*).

Once the structure of vitamin D was characterised and a simple process was developed for its synthesis, vitamin D was directly added to milk, thereby making the ultraviolet irradiation of milk obsolete. Ergocalciferol (D2) is commercially made by irradiating and then purifying the ergosterol extracted from yeast. Cholecalciferol (D3) is produced commercially by extracting 7-DHC from wool fat, followed by UVB irradiation and purification.

Photosynthesis

Vitamin D is produced in the skin from 7-DHC, the last precursor in cholesterol synthesis.[8] In adult human skin, approximately 50% of provitamin D3 (7-DHC) is found in the epidermis and the other half is found in the dermis.[8] During exposure to sunlight, the high-energy UVB photons with energies between 290 and 315 nm, penetrate into the skin where they are absorbed by provitamin D3 -the immediate precursor in cholesterol biosynthetic pathway. UVB photons break the B ring of the cholesterol structure, cleavage between carbons 9 and 10 to form a 9,10 – secosteroid known as previtamin D3. Previtamin D3 also has the capacity to absorb ultraviolet radiation. This results in its isomerisation to two photoproducts known as lumisterol and tachysterol.[9] Both of these photoisomers are inert in calcium metabolism. During prolonged exposure to the sun, the accumulation of previtamin D3 is limited to about 10 to 15% of the original 7-DHC content because the previtamin photoisomerizes to lumisterol 3 and tachysterol 3.[8] Previtamin D3 is biologically inert and thermodynamically unstable. As a result, its double bonds rearrange spontaneously to form vitamin D3 (*Figure 3*). At a physiological temperature of 37° C, this process would take approximately 24 to 48 hours to reach completion. However, it is now recognised that previtamin D3 is rapidly converted to vitamin D3 in human skin. Previtamin D3 exists in two isomeric forms known as the *cis,cis* and *cis,trans* forms. Only the *cis,cis* conformer can be converted to vitamin D3. The *cis,trans* form is thermodynamically more stable but cannot be converted to vitamin D3, thus accounting for the prolonged isomerisation time. It takes time for the *cis,trans* form to isomerise to the *cis,cis* conformer before it can be converted to vitamin D3. However, a unique mechanism operates that allows the efficient conversion of previtamin D3 to vitamin D3 within hours after its formation in the skin. The previtamin D3 is

sandwiched between fatty acids in the bilipid membrane.[10] Vitamin D3 is also able to absorb ultraviolet radiation. Exposure to ultraviolet radiation results in isomerisation of vitamin D3 to form at least three photoproducts, known as 5,6-trans vitamin D3, supersterol I, and supersterol II (*Figure 3*).[11] None of these vitamin D3 photoproducts has any effect on calcium metabolism at physiological concentrations. Once vitamin D3 is made, it undergoes a conformational change that allows it to move from the membrane to the extracellular space and eventually to the dermal capillary bed. Since most of the ultraviolet B radiation is absorbed in the epidermis, more than 70% of previtamin D3 synthesis occurs here.[8] Aging decreases the thickness of both the epidermis and dermis, therefore there is an age-dependent decline in provitamin D3 [8,12] resulting in a decreased capacity of the elderly to produce vitamin D3 in the skin.[13]

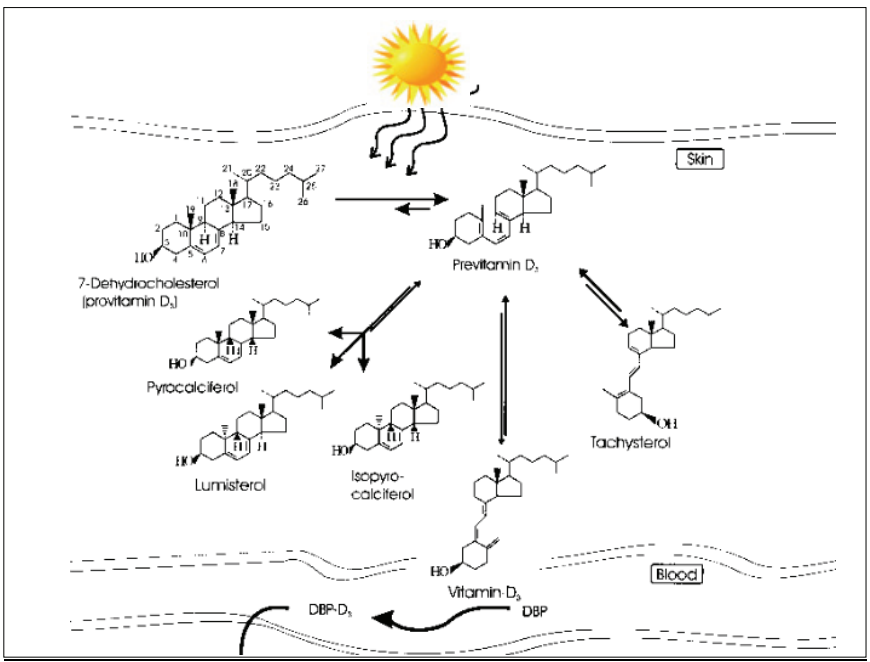

Figure 3*:* Photosynthesis of vitamin D in the skin. DBP= Vitamin D Binding Protein

Factors that influence on vitamin D photosynthesis

Any process that decreases or prevents ultraviolet B photons from reaching the viable epidermis to be absorbed by provitamin D3 results in a diminution in the photosynthesis of previtamin D3. Melanin and sunscreens are effective in absorbing UVB radiation. Thus, an increase in skin pigmentation can markedly diminish the cutaneous production of vitamin D3.[14] Thus heavily pigmented people require at least 5 to 10 times longer exposure than Caucasians to produce adequate vitamin D3 in their skin.[14] The application of a sunscreen with a sun protection factor (SPF) of 8 will reduce, by more than 95%, the cutaneous production of cholecalciferol.[15] Most clothing completely absorbs UVB radiation thus preventing the cutaneous production of vitamin D3 on the areas it covers.[16] It is well-known that sunlight exposure through glass will not result in any significant vitamin D3 production. The reason being that there are substances in glass, including lead, that absorb UVB radiation.[17]

The zenith angle of the sun has a dramatic effect on the total number of UVB photons reaching the earth's surface. During the winter, at latitudes above 40° north and below 40° south of the equator, the UVB photons are efficiently absorbed by the ozone layer, essentially eliminating the ability of the skin to produce vitamin D3.[18] At latitudes above and below 34° south and north respectively, there is cutaneous production of vitamin D3 all year round. The latitude of Sweden is 62° north of the equator and in this geographic area UVB is not transmitted in sunlight from October to March.

How much sunlight is necessary to satisfy the body's requirement?

This is an especially relevant question in view of concern about the damaging effects of excessive exposure to sunlight. An adult Caucasian exposed to sunlight or a (tanning bed) lamp (~32 mJ/cm^2) that emits UVB radiation produces ~1 ng of cholecalciferol/cm^2 skin.[2] A study was conducted where medical students received a whole-body exposure to 1 minimal erythema dose (MED) of solar simulated sunlight together with graded doses of vitamin D. The circulating levels of vitamin D were subsequently measured at various intervals. It was found that a whole-body exposure to 1 MED of UVB radiation resulted in a blood level of vitamin D that was comparable to taking between 10,000 and 25,000 IU of vitamin D orally.[17]

Metabolism

It has been estimated that between 90 and 95% of all people obtain their vitamin D requirement from exposure to sunlight.[17] The vitamin D3 produced in the skin or ingested from the diet can be stored in body fat and released into the circulation at during times when there is an inadequate cutaneous production of vitamin D3. In obese children and adults, the cholecalciferol is sequestered deep in the body fat, making it less bioaviable.[2] Thus, obese individuals are only able to increase their blood levels of vitamin D by approximately 50% compared with normal-weighted individuals.[19] However, vitamin D is biologically inert and must be metabolized in the liver on carbon 25 to form the major circulating form of vitamin D, 25-hydroxycholecalciferol (25(OH)D) or calcidiol, (*Figure 4*).

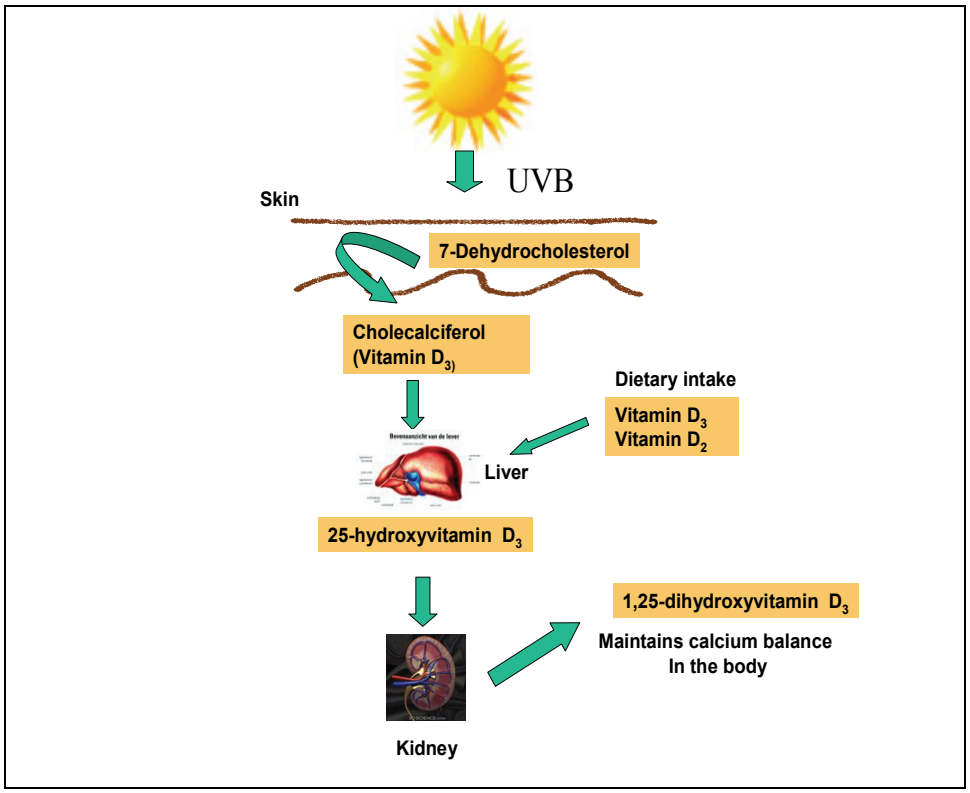

Figure 4: Metabolism of vitamin D

25(OH)D is used clinically to measure vitamin D status for vitamin D deficiency, sufficiency, and intoxication.[17] Because 25(OH)D is biologically inert at physiological concentrations, it must be converted in the kidney to its activated form 1,25 dihydroxycholecalciferol [1,25$(OH)_2$D] or calcitriol, (*Figure 4*).

Vitamin D-binding protein (DBP), a 458-amino acid polymorphic human serum protein, is the major plasma carrier of vitamin D3 and of all its metabolites, which include 25(OH)D and 1,25$(OH)_2$D.[20]

The skin occupies a central position within the vitamin D system. Epidermal keratinocytes also express the vitamin D hydroxylase enzymes 25-hydroxylase (CYP27A1) and 1α-hydroxylase (CYP27B1), enabling them to convert vitamin D3 into 25(OH)D and 1,25$(OH)_2$D, the biologically active form of vitamin D3.[21] In addition, keratinocytes are vitamin D target cells as they contain the vitamin D receptor (VDR) and respond to 1,25$(OH)_2$D with changes in proliferation, differentiation and cytokine production.[22] Taken together, these findings indicate the existence of a unique photoendocrine vitamin D system in keratinocytes - this is corroborated by the demonstration of 1,25$(OH)_2$D synthesis and vitamin D effects in UVB-irradiated skin or keratinocytes.[21,23] In contrast to epidermal keratinocytes, dermal fibroblasts only have the capacity for photoproduction of 25(OH)D but not 1,25$(OH)_2$D.[24] 25(OH)D may then act as a paracrine factor to keratinocytes. Monocytes, like keratinocytes, have the full machinery to photoproduce 1,25$(OH)_2$D.[23]

The physiologic significance of the cutaneous photosynthesis of 1,25$(OH)_2$D is poorly understood. As the amounts of UVB-produced 1,25$(OH)_2$D in keratinocytes are small[23] and circulating 25(OH)D and 1,25$(OH)_2$D are hardly detectable in hepatectomized or nephrectomized animals[25], cutaneous production of active vitamin D does not appear to play a major role outside the skin. However, locally produced 1,25$(OH)_2$D may contribute to UVB effects within the skin, such as its therapeutic action on psoriasis.[26] Furthermore, photoproduced 1,25$(OH)_2$D might serve as an endogenous protection mechanism against UVB-dependent DNA damage, apoptosis and release of proinflammatory cytokines such as IL-6.[27,28]

Levels of 25(OH)D

A 25(OH)D level less than <30 ng/ml (75 nmol/l) is considered to be suboptimal vitamin D status - this is the minimal level of 25(OH)D necessary to suppress

parathyroid hormone secretion.[29-31] A 25(OH)D level of between 21 ng/ml (52 nmol/l) and 29 ng/ml (74 nmol/l) is considered to be vitamin D insufficiency.[31-33] The cut-off level for serum 25(OH)D, which is taken as a diagnostic value for vitamin D deficiency, has varied over the years.[33-35]

Mechanism of action

$1,25(OH)_2D$ interacts with its nuclear VDR, which in turn binds with the retinoic acid-X-receptor. This complex is recognised by specific gene sequences known as the vitamin D responsive elements (VDRE) to unlock genetic information that is responsible for its biologic actions. In the intestine $1,25(OH)_2D$ induces the expression of an epithelial calcium channel, calcium-binding protein (calbindin), and a variety of other proteins to help the transport of calcium from food into the circulation.[36] $1,25(OH)_2D$ also interacts with the VDR in the osteoblast and stimulates the expression of receptor activator of NFκβ ligand (RANKL) similar to PTH.[37] Thus, $1,25(OH)_2D$ maintains calcium homeostasis by increasing the efficiency of intestinal calcium absorption and mobilizing calcium stores from the skeleton.

PTH, hypocalcaemia, and hypophosphatemia are the major stimulators for the renal production of $1,25(OH)_2D$.[37] During pregnancy, lactation, and growth sex steroids, prolactin, growth hormone, and insulin-like growth factor 1 (IGF-1) play a role in enhancing the renal production of $1,25(OH)_2D$ to satisfy increased calcium needs. [37]

Vitamin D deficiency results in a decrease in the efficiency of intestinal absorption of dietary calcium and phosphorus.[37] This causes a transient lowering of the ionized calcium, which is immediately corrected by the increased production and secretion of PTH. PTH sustains the blood-ionized calcium by interacting with its membrane receptor on mature osteoblasts, which induces the expression of RANKL.[37] This plasma membrane receptor protein is recognised by RANK that is present on the plasma membrane of preosteoclasts. The intimate interaction between RANKL and RANK results in increased production and maturation of osteoclasts.[37,38] Osteoclasts release hydrochloric acid and collagenases to destroy bone, resulting in the mobilization of calcium stores from the skeleton. Thus vitamin D deficiency induced secondary hyperparathyroidism results in skeletal wasting that can precipitate and exacerbate osteoporosis.[2] Vitamin D deficiency and attendant

secondary hyperparathyroidism also causes loss of phosphorus into the urine and lowering of serum phosphorus levels. This results in an inadequate calcium x phosphorus product, causing poor or defective mineralization of the bone matrix laid down by osteoblasts.[2] In children, the effect of body weight and gravity on a poorly mineralized skeleton results in the classic bony rachitic deformities in the lower limbs (bowed legs and knocked knees). Adults have enough mineral in their skeleton to prevent skeletal deformities. However, in vitamin D deficient state, the newly laid-down osteid cannot be properly mineralised, leading to osteomalacia. Unlike osteoporosis, which is a silent disease until fracture occurs, osteomalacia is associated with either widespread or localised throbbing bone pain. The likely cause is that the unmineralised osteoid becomes hydrated and provides little support for the sensory fibres in periosteal covering.[39] Osteomalacia cannot be distinguished from osteoporosis/osteopenia by neither X-ray analyses nor by bone densitometry - they appear identical.

Besides the small intestine and the osteoblast, VDR has been identified in almost every tissue and cell in the body, including brain, heart, skin, pancreas, breast, colon and immune cells.[37,40] $1,25(OH)_2D$ helps regulate cell growth and maturation, stimulates insulin secretion, inhibits renin production, and modulates the functions of activated T and B lymphocytes and macrophages.[37,41]

It is documented that risk of morbidity or mortality from colon, prostate, breast, ovarian, oesophageal, non-Hodgkin's lymphoma, and a variety of other aggressive cancers is related to living at higher latitudes and being at higher risk of vitamin D deficiency.[42-45] Initially the explanation for why increased sun exposure decreased the risk of fatality from common cancers was due to the increased production of vitamin D in the skin, leading to the increased production of $1,25(OH)_2D$ in the kidneys.[42] Because it was known that the VDR existed in most tissues in the body and that $1,25(OH)_2D$ was a potent inhibitor of both normal and cancer cell growth[10,37] it was assumed that the increased renal production of $1,25(OH)_2D$ could downregulate cancer cell growth and therefore mitigate the cancer's activity and decrease mortality. However, it was also known that the production of $1,25(OH)_2D$ in the kidneys was tightly controlled and that increased intake of vitamin D or exposure to sunlight did not result in an increase in circulating concentrations of $1,25(OH)_2D$.[37] However, this still did not explain the anti-cancer effect of sunlight - vitamin D concentration. The skin not only makes cholecalciferol, but it also has the enzymatic machinery to convert 25(OH)D to $1,25(OH)_2D$, similar to activated

macrophages.[46,47] In 1998 Schwartz et al.[48] reported normal and malignant prostate cancer cells also had the enzymatic machinery to make $1,25(OH)_2D$. Increased exposure to sunlight or vitamin D intake leads to increased production of 25(OH)D. Higher concentrations of 25(OH)D are used by prostate cells to make $1,25(OH)_2D$, which help keep prostate cell proliferation in check and therefore decreases the risk of malignancy.[37] It has since been observed that breast, colon, lung, brain and a wide variety of other cells in the body are able to produce $1,25(OH)_2D$.[37,49-53] Thus it has been suggested that raising blood levels of 25(OH)D provides most of the body's tissues with enough substrate to make $1,25(OH)_2D$ locally to act as a sentinel to help control cellular growth and maturation and decrease the risk of malignancy.[37] Both prospective and retrospective studies revealed that, if the 25(OH)D level is at least 20 ng/ml, than there is an approximate 30-50% decreased risk of developing and dying of colon, prostate, and breast cancers.[37,42,45,54]

There are several studies suggesting that increased exposure to limited amounts of sunlight decreases the risk of developing and dying of the most deadly form of skin cancer, melanoma.[55,56]

Effects on autoimmune diseases

Activated T and B lymphocytes, monocytes, and macrophages have VDR.[41,53,57,58] $1,25(OH)_2D$ interacts with its VDR in immune cells and has a variety of effects on regulating lymphocyte function, cytokine production, macrophage activity, and monocyte maturation.[37,41,53,58,59] Thus, $1,25(OH)_2D$ is a potent immunomodulator. Insights into the important role of vitamin D in the prevention of autoimmune diseases have come from a variety of animal studies. Nonobese diabetic mice that typically develop type I diabetes by 200 days reduced their risk of developing this disease by 80% when they received a physiological dose of $1,25(OH)_2D$ daily.[60] Mice that were pretreated with $1,25(OH)_2D$ before they were injected with myelin to induce a multiple-sclerosis-like disease were immune from it.[61] Similar observations were made in a mouse model that develops Crohn's disease.[62] These animal model studies have given important insights into the role of $1,25(OH)_2D$ in reducing the risk of developing common autoimmune diseases such as multiple sclerosis[63] and rheumatoid arthritis.[64] Most compelling is the observation that children in Finland who received 2000 IU of vitamin D daily from one year and were followed up for the next 25 years had an 80% decreased risk of developing

type I diabetes, whereas children who were vitamin D deficient had a four-fold increased risk of developing this disease later in life.[65]

Effects on hypertension

In 1997 Rostand [66] reported that people living at higher latitudes throughout the world were at higher risk of developing hypertension. He suggested that this may be related to being more prone to developing vitamin D deficiency. To determine the possible link between sun exposure and the protective effect in preventing hypertension, Krause et al. [67] exposed a group of hypertensive adults to a tanning bed that emitted light and UVB and UVA radiation similar to summer sunlight. A similar group of hypertensive adults was exposed to a similar tanning bed that emitted light and UVA radiation similar to winter sunlight, i.e., no UVB radiation. All subjects were exposed 3 times a week for 3 months. After 3 months, it was observed that hypertensive patients who were exposed to the tanning bed that emitted UVB radiation had a 180% increase in their circulating concentrations of 25(OH)D. There was no change in blood levels of 25(OH)D in the comparable group that was exposed to the tanning bed that did not emit UVB radiation. The patients who had increased 25(OH)D levels also had a decrease of systolic and diastolic blood pressures by 6 mm Hg, bringing them into the normal range. The placebo group (exposed to light that did not emit UVB radiation) did not change their 25(OH)D levels and no effect was observed on their blood pressure.

It has also been observed that patients with cardiovascular heart disease are more likely to develop heart failure if they are vitamin D deficient.[68] The exact mechanism responsible for vitamin D sufficiency protecting against cardiovascular heart disease is not fully understood. It is known that $1,25(OH)_2D$ is one of the most potent hormones for downregulation of the blood pressure hormone renin in the kidneys.[69] Furthermore vascular smooth muscle cells have a VDR that in the presence of $1,25(OH)_2D$ induces relaxation and thereby vasodilatation.[70,71]

Antimicrobial effects

In addition to phagocytes and cytokines, a major component of the innate immune system is a diverse combination of cationic peptides that include the α- and β-defensins and cathelicidins, which have potent microbicidal activities at low concentrations.[72]

The antimicrobial peptide cathelicidin is a vitamin D target gene,[73] and induces upregulation of CYP27B1 and VDR in monocytes. Cathelicidin expression was also shown to be increased in human skin *in vivo* by topical application of active vitamin D compounds such as calcipotriol and $1,25(OH)_2D3$.[74]

Cathelicidin plays also the role in wound healing and inflammatory skin disease.[75] Cathelicidin is not only an effector molecule of innate immunity by its antimicrobial activity but it also exhibits biological activities on adaptive immunity, angiogenesis and cell proliferation and migration.[72] Therefore, its function is pivotal for wound repair.[76] In addition to its upregulation in the skin following cutaneous injury, high expression of cathelicidin and of antimicrobial peptides has also been noted in psoriasis, accounting for the rare occurrence of skin infections in this condition.[72]

Substitution

Today, companies such as Hoffmann-La Roche and BASF produce large quantities of the primary form of vitamin D, also known as vitamin D3 or cholecalciferol. Cholesterol from animal products (such as lanolin from sheep wool) is purified and used as starting material for purification of the precursor 7-DHC, which is then converted into vitamin D3 by irradiation. This synthetic vitamin D3 is added to many foods, particularly milk products; it is also a key ingredient in the multivitamin supplements that many people take regularly.

Doses of calciferols are often expressed in international units (IU), there being 40 IU/μg of cholecalciferol and 38.8 IU/μg of ergocalciferol.[77]

Conclusions

It has been estimated that the body requires daily 3000-5000 IU of vitamin D.[2,78] The most likely reason for this is that essentially every tissue and cell in the body has a VDR and thus a requirement for vitamin D.[2] Vitamin D is critically important for the maintenance of calcium metabolism and good skeletal health throughout life.[79,80] The revelations that vitamin D regulates the immune system, controls cancer cell growth and regulates the blood pressure hormone renin provides an explanation for why vitamin D sufficiency has been observed to be so beneficial in the prevention of many chronic illnesses that plague both children and adults. People of darker skin colour are more prone to vitamin D deficiency at Northern

latitudes. Vitamin D deficiency has been recognised even in some of the sunniest climates, including Saudi Arabia and India.[81,82]

Thus, vitamin D status has such important health implications that a measurement of 25(OH)D should be part of a routine physical examination for children and adults of all ages.

In the absence of sun exposure, 1000 IU of cholecalciferol a day is necessary to maintain a healthy blood level of 25(OH)D of between 32 and 40 ng/ml (80 -100 nmol/l).[2] Increasing the intake of vitamin D fortified foods and increasing fatty fish consumption will help satisfy the requirement for vitamin D in the absence of exposure to sunlight. Multivitamins typically contain 400 IU of vitamin D. Thus, diet plus supplementary vitamin D can result in attaining the recommended 1000 IU of cholecalciferol.

It has been estimated that exposure to sunlight for usually no more than 5-15 minutes per day (between 10 am and 3 pm) on the limbs or hands, face and arms during the spring, summer, and autumn (not during the winter unless located below 35° north) provides the required 1000 IU of cholecalciferol.[2] Limited exposure should be followed by the application of a broad spectrum sunscreen with an SPF of at least 15 to prevent damaging effects due to excessive exposure to sunlight and to prevent sun burning.[2]

Psoriasis

Psoriasis is a chronic, non-contagious skin disease characterized by red, inflamed cutaneous lesions covered with silvery-white scale. The term *psoro* comes from the Greek word for itch; *psoriasis* corresponds with the term *itchy*. In 1841 Ferdinand Hebra, a Viennese dermatologist, was the first to ascribe the name 'psoriasis'. He described the clinical picture of psoriasis that is used today. The hereditary factor of psoriasis had already been established by that time.

Psoriasis affects both genders equally and can occur at any age, although it most commonly appears for the first time between the ages of 15 and 25. The prevalence of psoriasis in Western populations is estimated to be around 2-3%. The disease affects the skin and potentially the joints. The prevalence of psoriatic arthritis in psoriasis patients is estimated to be between 25-31%.[83]

Both genetic and environmental factors are important in the aetiology of the disease. Psoriasis can be triggered by certain external stimuli, for example, bacterial infections or injuries.[84] Genetic susceptibility factors that contribute to predisposition to psoriasis are being identified[85], but so far, the cause of disease is not fully understood.

The disease is an immune-mediated disorder in which excessive reproduction of skin cells is secondary to factors produced by the immune system. T-cells, dendritic antigen-presenting cells (APC) and cytokine networks are recognized as playing a major role in the pathogenesis of psoriasis.[85] Dysregulation of T-cell APC interactions and over expression of proinflammatory cytokines lead to the characterised hyperproliferation and decreased differentiation of keratinocytes expressed by the increased cell turnover in epidermis.[85] It is not known what initiates the activation of these cells. Such triggers activate the otherwise dormant innate immunity in the skin.

In patients with psoriasis, innate immunity then becomes hyperactivated. Pathomorphologically, this is represented by the activity of natural killer (NK) cells, increased antigen presentation by Langerhans cells, an influx of CD4+ T cells, neutrophils, an activation of tumour necrosis factor *a* (TNF-*a)*–releasing macrophages[86-88] and hyperprolifreation of keratinocytes. This activation leads finally to the recruitment of effector T-helper cell type 1 (TH-1) cells. The epidermis of patients with psoriasis responds in a typical pattern to T-cell activation and cytokine production. TH-1 cells produce large amounts of interferon gamma (IFN-γ), TNF-*a*, and interleukin (IL) 2 and activate macrophages to secrete even more TNF-*a*.[88,89] TNF-*a* and IFN-γ, have been shown to play a key role in psoriasis.[90] IFN-γ is responsible for the epidermal hyperplasia.[91] IFN-γ can trigger psoriasis at injection sites and T cells in psoriatic plaques produce it in high amounts. High TNF-*a* levels have been found in psoriatic skin as well as serum and are considered to derive mainly from activated macrophages.[86,92] Furthermore, TNF-*a* and IFN-γ levels in serum correlate with disease activity of psoriatic patients.[93,94] Why the T cells become active and remain in that state is as yet unclear. Chronic inflammation of psoriatic lesions suggests that there is an underlying inborn "error" in the regulatory T-cell population and the persistence of a yet unknown trigger that induces an exaggerated innate immune response.[84] Recently it has been shown that regulatory T cells are functionally deficient in patients with psoriasis and not present in sufficient numbers in lesions to achieve a

downregulation of the hyperresponse.[95] Furthermore, the basement membrane zone of patients with psoriasis seems to be altered structurally in a way that basal layers of keratinocytes can be more easily promoted to proliferate than usual, particularly in the presence of the inflammatory T-cell cytokines.[95,96]

Disease management is dependent on severity, psychosocial effects and the patient's lifestyle. The treatment consists of various local and systemic treatments. Local treatments include creams and ointments containing tar, dithranol, salicylic acid or vitamin D-related compounds (calcipotriol (Daivonex)®, calcitriol (Silkis)® or tacalcitol (Curatoderm)®). Occasionally, corticosteroid-containing ointments are used for a short time. Vitamin D3 analogues inhibit proliferation, induce terminal differentiation of human keratinocytes and exhibit immunomodulating properties.[97] Several studies have shown that calcipotriol as well as calcitriol and tacalcitol are efficacious, safe and can be used on a long-term basis for psoriasis.[98-101] Vitamin D3 analogues can be used in combination with phototherapy.[102]

Effects of vitamin D3 analogues in psoriasis

Psoriasis is characterized by keratinocyte hyperproliferation, abnormal keratinocyte differentiation, and immune-cell infiltration into the epidermis and dermis. The most common form of psoriasis is plaque psoriasis or psoriasis vulgaris. At the molecular level, psoriasis lesions show a prominent loss of loricrin and filaggrin in the suprabasal layers of the epidermis and abnormal overexpression of other differentiation markers such as involucrin, transglutaminase I (TGase I), psoriasin, migration inhibitory factor related protein-8, and skin-derived antileukoproteinase. The expression of normal suprabasal keratins K1 and K10 is inhibited and replaced by the expression of the hyperproliferative keratins K6 and K16.

The observations that keratinocytes and T cells express VDR and that 1,25-$(OH)_2$D is a potent stimulator of keratinocyte differentiation provided a reasonable basis for the clinical use of VDR ligands for the treatment of psoriasis.[103,104] The first clinical evidence to support the use of vitamin D analogues was obtained fortuitously when a patient treated orally with 1α-hydroxyvitamin D3 for osteoporosis showed remarkable remission of psoriatic lesions.[105] Subsequently, promising clinical results were obtained in studies using oral 1α-hydroxyvitamin D3, oral and topical 1,25-$(OH)_2$D (calcitriol), and topical 1,24-$(OH)_2$D. In these clinical trials, approximately

70–80% of the patients showed improvement, and complete clearance of the target lesions was observed in 20–25% of patients.[106]

The antipsoriatic activity of VDR ligands could be attributed to their differentiation, antiproliferative, and immunomodulatory properties. VDR ligands exhibit multipronged effects in psoriatic lesions and affect the function of keratinocytes, T cells, and APC. VDR ligands promoted differentiation and inhibited the proliferation of keratinocytes.[106,107] Differentiation of keratinocytes results in the formation of a cornified envelope (CE) that provides the barrier function of the skin. The expression of involucrin, a component of the CE, and TGase I, the enzyme that cross-links the components of CE, was increased by $1,25(OH)_2D$ and other VDR ligands.[108] Treatment of keratinocytes with the medium containing high calcium also stimulated keratinocyte differentiation by increasing the expression of involucrin and TGase I. $1,25(OH)_2D$ promoted keratinocyte differentiation, increased the expression of calcium receptor in keratinocytes[109] and indirectly induced the expression of keratin 1, involucrin, TGase I, loricrin, and filaggrin, which are required for CE formation. VDR ligands decreased the expression/level of proinflammatory cytokines IL-2, IFN-γ, IL-6, and IL-8[110-113] in T cells, all of which play a role in cutaneous inflammation, and proliferation of T lymphocytes and keratinocytes. Furthermore, topical calcipotriol increased antinflammatory cytokine IL-10 and decreased IL-8 in psoriatic lesions [114], and $1,25\text{-}(OH)_2D$ also increased the expression of IL-10 receptor in keratinocytes.[115]

APCs or dendritic cells (DCs) also play an important role in psoriasis and autoimmune diseases because they are involved in autoantigen presentation. It appears that APCs are one of the major targets of $1,25\text{-}(OH)_2D$-mediated immunosuppressive action and VDR ligands prevent the differentiation, maturation, activation, and survival of DCs, leading to T cell hyporesponsiveness.[116] $1,25\text{-}(OH)_2D$ also increased the expression of IL-10 and decreased the expression of IL-12, two major cytokines that are involved in Th1-Th2 balance.[117]

Psoriasis and comorbidities

Psoriasis is considered a chronic and debilitating inflammatory disease associated with serious comorbiditites.[118,119] The chronic inflammation in psoriasis can predispose patients to other inflammatory conditions. For example, individuals

with psoriasis are at increased risk for insulin resistance, obesity, dyslipidemia, and hypertension - components that characterize the metabolic syndrome. The metabolic syndrome is an important driver of adverse cardiovascular outcomes. It is likely that proinflammatory cytokines, such as TNF-*a*, and other factors that are overproduced in patients with psoriasis likely contribute to the increased risk for development of metabolic syndrome.[120,121]

The high prevalence of atherosclerosis is also reported in psoriasis patients. In the pathogenesis of this phenomenon high serum lipid levels have been suggested. High serum lipid levels are common in psoriasis and may be responsible for an elevated prevalence of cardiovascular events in these patients.[122] It may be useful to perform early screening and treatment of hyperlipidaemia in psoriasis to prevent atherosclerosis and its complications. Inflammation plays a key role in the pathogenesis of psoriasis and a number of chronic inflammatory systemic diseases listed above. Activated inflammatory cells and pro-inflammatory cytokines contribute to the development of psoriatic lesions and play an important role in the breakdown of atherosclerotic plaques. Psoriasis and atherosclerosis also have similar histological characteristics involving T cells, macrophages and monocytes. In particular, extravasation of T cells through the epithelium is characteristic of both psoriatic and atherosclerotic plaques.

Inflammatory factors have also been associated with insulin resistance and β-cell failure, both are key features of type 2 diabetes mellitus.[123] There is evidence that vitamin D may stimulate pancreatic insulin secretion directly. Vitamin D exerts its effect through nuclear receptors that are found in a wide variety of tissues, including T and B lymphocytes, skeletal muscle, and the pancreatic islet β-cells.[124] The stimulatory effects of vitamin D on insulin secretion may be manifest only when calcium levels are adequate.[124] There is some evidence that increased PTH activity is associated with, and possibly causes, reduced insulin sensitivity.[124] The prevalence of impaired glucose tolerance and diabetes mellitus is increased in patients with primary hyperparathyroidism.[125,126]

Vitamin D has a wide range of effects on the immune system: it promotes the differentiation of monocytes into macrophages thus increasing their cytotoxic activity; reduces the antigen-presenting activity of macrophages to lymphocytes; prevents dendritic cell maturation; inhibits T lymphocyte-mediated immunoglobulin synthesis in B cells and inhibits delayed-type hypersensitivity

reactions.[34,127,128] In contrast, vitamin D exerts an antiproliferative effect on activated lymphocytes while suppressing the generation and activity of new NK cells.[129,130] Furthermore vitamin D has been reported to downregulate the production of several cytokines: IL-2, IL-6 and IL-12, IFN-γ, TNF-α, and TNF-β.[128,131] Alternations in vitamin D status and/or action may affect insulin sensitivity, β-cell function or both. Furthermore, several vitamin D-relatid genes are associated with different pathogenetic traits of the disease. Therefore, vitamin D and its related metabolic and immune pathways may be involved in the pathogenesis of type 2 diabetes mellitus at both environmental and genetic levels.[123]

Osteoporosis

Information about prevalence of osteoporosis among patients with psoriasis and epidemiology of risk factors for osteoporosis in this group is rare. Previous studies did not detect any differences in bone mineral density (BMD) between patients with psoriasis and healthy controls.[132] It is not known if psoriasis disease itself has any impact on bone metabolism.

However, a previous study on psoriasis patients showed no evidence that patients with chronic plaque psoriasis, despite risk factors, had low BMD, although the subgroup with joint involvement appeared to be at higher risk of osteoporosis and therefore required prevention.[132] Reduced BMD has been linked to palmoplantar pustular psoriasis.[133] The existence of less severe periarticular osteoporosis is considered possible but there is no data concerning the existence of systemic osteoporosis in patients with psoriatic arthritis.[134]

Vitamin D is important for bone metabolism.[135] Vitamin D deficiency thus contributes to the pathogenesis of osteoporosis and hip fractures.[136] The supplementation strategy with calcium and vitamin D supplements is cost saving for osteoporotic fracture.[137]

Phototherapy

It has been known for more than 2000 years that several skin diseases improve upon exposure to the sun (heliotherapy), but the systematic investigation of phototherapeutic modalities did not start until the beginning of the 20th century. UV radiation that reaches the skin is either reflected or absorbed by structures of the skin. UVC (<280 nm) is mostly absorbed in the stratum corneum, and UVA (320-400 nm) shows deeper penetration than UVB (280-320 nm).[139-143] Thus, UVB

is mainly absorbed by epidermal components including keratinocytes, melanin and Langerhans' cells.[144] Biological effects of UV radiation are generated through interaction with absorbing molecules called chromophores. In the case of UVB, the most important chromophores are proteins such as keratin, melanin, collagen and elastin, urocanic acid, DNA and previtamin D.[8,145-147]

In 1903, Niels Finsen received the Nobel Prize for developing phototherapy as a treatment for tuberculosis of the skin. Around the turn of the 19th century, Sardemann used for the first time ever a carbon arc lamp to treat psoriasis in a patient.[148] In 1923, Alderson then described "heliotherapy in psoriasis". He used a quartz-jacketed mercury discharge lamp to treat his first patients.[149] Three years later Goeckerman[150] demonstrated the beneficial effect of natural sunlight in combination with tar for psoriasis vulgaris "Goekermann regimen".[151] In 1953, Ingram initiated the combination of UVB radiation, dithranol and tar-bathing for psoriasis.[152] In 1974, the combination of oral psoralen intake and subsequent UVA irradiation was reported by Parrish[153] and Wolff[154], publications that marked the initiation of modern psoralen+UVA (PUVA) photochemotherapy for psoriasis. Phototherapy is thus an old and established treatment modality for this disease. According to Feldman et al.[155], with regard to efficacy, safety and cost-effectiveness, UVB phototherapy appears to be the best first-line treatment for the control of generalized psoriasis. Data from Fischer & Alsins[156] and Parrish & Jaenicke[157] subsequently showed that wavelengths around 311 nm provoke least erythema while being most effective for clearing psoriasis. According to these results a fluorescent bulb was developed (TL-01), emitting a major peak at 311 (± 2nm) and a minor peak at 305 nm. This treatment was later called narrowband UVB (NBUVB) and following its introduction several studies were published on its superior efficacy in phototherapy of psoriasis.[158-161] There is a large body of evidence indicating that NBUVB is more effective than broadband UVB as a monotherapeutic agent in the treatment of psoriasis even in children.[160,162-164]. Whereas broadband UVB is considered to be most effective close to the minimal erythema dose (MED), NBUVB has also been shown to be effective in suberythemogenic doses.[165] However, Diffey[166] could show in a mathematical model that clearance of psoriasis plaques is achieved faster with higher MED rates. For treatment of psoriasis with NBUVB, three rather than two or five radiations a week are effective[167,168] and low incremental regimens are sufficient according to

Wainwright et al.[169], who showed this regimen to be as effective as high incremental regimens but less erythemogenic.

Ultraviolet radiation clears psoriasis with varying efficacy depending on the wavelength. Wavelength dependency was determined with monochromatic UV radiation and resulted in the action spectrum for psoriasis as established by Parrish.[149,156,157] Wavelengths of less than 300 nm also achieve significant clearing of psoriasis, but cause more side effects in terms of erythema and burning and are thus less efficient.[170] Further conclusions from this spectral analysis were that throughout the UVB and UVA ranges, the daily dose to clear psoriasis is equivalent to the MED.[148] In other words, if a UV source emits the psoriasis action spectrum, one MED per day would be the best dosage regimen to clear the patients psoriasis.[161] On the other hand the production of vitamin D is mediated by UVB dose equivalent to one MED.

Mechanism of action

Phototherapy (broadband UVB, NBUVB and heliotherapy) are commonly used treatment modalities for widespread psoriasis.

Ultraviolet radiation influences the pathologic immune response in psoriasis in multiple ways. Firstly, it alters the deviated antigen presentation, which is a major trigger in psoriasis lesions. Langerhans cell numbers decrease by 90% after 7 UV treatment sessions.[171] Furthermore the remaining dendritic cells acquire cytoskeleton damage by photo-oxidative stress, and this reduces subsequently their ability to express high numbers of costimulatory surface markers to efficiently stimulate T cells. UV radiation also alters the cytokine secretion pattern of macrophages at the site of inflammation and can diminish their numbers by induction of apoptosis.[172] Under the influence of UV radiation, they produce IL-10, an important immunosuppressive cytokine that shifts the TH-1 environment back toward a TH-2 setting. Furthermore, macrophages produce IL-15 under the influence of UV radiation, a cytokine that recruits new T cells into the plaque. These new inactivated T-cells replace the originally present, hyperactivated set of T cells that become apoptotic under UV irradiation.[173] T cells are already susceptible to very low doses of UVB and are thus more or less a selective target of photons.[174]

The downregulation of T-cell activity goes along with a decrease in IL-2, a general cytokine of T-cell activation in skin after UV irradiation.[175] This can partly be attributed to another effect of UV radiation: among the new T-cells recruited into the lesion are regulatory T-cells, and UVB and PUVA have been shown to induce regulatory T-cell production and thus promote intrinsic immunologic control of pathologic overreaction.[95,176,177] Ultraviolet radiation, however, also acts on the innate defence because it has been shown to switch off the activity of neutrophil and causes a dose-dependent inhibition of NK cell function.[178,179] Doses that easily reach the dermis in clinical practice have been shown to diminish NK cell activity by at least 50%.[171] This effect is exerted through reactive oxygen species induced by UV, in particular the superoxide anion (O_2^-) plays a key role here.[171]

UV radiation increases the IL-1 receptor antagonist in the epidermis more than inducing IL-1, a fact that strongly promotes keratinocyte differentiation and thus contributes to restoring the disturbed epidermal microarchitecture of psoriasis. Interestingly the effects on TNF-*a* are controversial. Ultraviolet B increases TNF-*a*, whereas UVA radiation reduces it. Ultraviolet radiation therefore does not inhibit this major cytokine of psoriasis pathology.[180]

The effects of UV radiation on psoriasis can be divided into 2 mechanisms; immediate effects and delayed effects. Immediate effects are, for example, cell membrane damage by lipid peroxidation, DNA damage, induction of cytoplasmatic transcription factors, and isomerization of chromophores such as urocanic acid.[170,181-184] These effects are largely cytopathic and induce growth arrest or even apoptosis. Delayed effects result from cells surviving the immediate effects of photon bombardment and lead to a modulation of the psoriasis microarchitecture, particular the TH-1–dominated immune response.[166,170]

Heliotherapy, natural sunlight, has been used as a UV source throughout history long before modern phototherapy was developed. Heliotherapy has been shown to be generally effective in clearing psoriasis.[185] Climatotherapy comprises alternative treatment methods employing the healing capacities of natural resources, including sunlight, temperature, humidity, barometric pressure and air. No other skin disease responds to heliotherapy as dramatically as psoriasis vulgaris.

Aims of the investigation

The overall aims of the 4 studies were to 1) increase the knowledge about the effects of UVB on vitamin D production during treatment with phototherapy and heliotherapy in patients with psoriasis; 2) identify any differences between the effect of NBUVB and broadband UVB on vitamin D synthesis in this group of patients; and 3) investigate the effects of UVB-induced vitamin D synthesis on bone, lipid and carbohydrate status in psoriasis patients.

Paper **I**. The aim was to examine whether broadband UVB therapy was capable of inducing vitamin D synthesis, thereby supplying psoriasis patients with vitamin D.

Paper **II**. The aim was to investigate bone mineral density (BMD) and factors contributing to osteoporosis in a group of postmenopausal women with psoriasis who had been exposed to UVB therapy versus an age-matched control population.

Paper **III**. The aim was to study whether the effect of NBUVB on vitamin D synthesis differed from the effect of broadband UVB in this respect.

Paper **IV**. The aim was to investigate the effect of climate therapy on vitamin D synthesis, blood glucose and lipids, vitamin B12, C reactive protein and haemoglobin in patients with psoriasis.

Material and methods

Subjects

White, Caucasian patients with active plaque psoriasis were included in the studies (Paper I–IV). Clinical characteristics of the subjects included in the different studies are presented in Table 1.

Study	Number of patients	Sex M/F	Age (year) (mean±SD)	PASI score before	PASI score after	Age at onset of psoriasis (mean±SD)
Paper I	24	0/24	69 ± 5.9	6-12 (range)	1-4 (range)	34.0 ± 23
Paper II	35	0/35	69.3 ±6.3			37.0 ±23.5
Paper III	68	51/17	54.1±16.0	9.0±4.7	2.6±1.6	26.3 ± 14.2
Paper IV	20	14/6	47.2±10.7	9.8±4.5	2.4±1.7	

Table 1: Clinical characteristics of the psoriatic subjects included in study I–IV of this Thesis. PASI (Psoriasis Area and Severity Index)

Anthropometry and bone measurement (paper I and II)

Body weight and height were measured in underwear and without shoes to the nearest 0.5 kg and cm, respectively. Body mass index (BMI) was calculated as body weight divided by height squared (kg/m^2), measured at the time of the Dual-Energy X-ray Absorptiometry (DEXA) examination.

Bone mineral density (BMD) was examined by DEXA with Hologic Delphi A at the hip and the lumbar spine after UVB therapy. BMD results were recorded as the T-score (SD from the mean peak value in young sex-matched adults) and the Z-

score (difference in SD from the mean of a healthy, age- and sex-matched sample) for each subject. The Z-score was adjusted in the BMD measurements from population data and inlaid as a control value in Hologic Delphi A.

Vitamin D analyses

Paper I and III

Serum samples for 25(OH)D and $1,25(OH)_2D$ were obtained at baseline and after the last dose of radiation. Serum 25(OH)D and $1,25(OH)_2D$ were measured using the ^{125}I (radioimmunoassay) RIA method (DiaSorin, Stillwater, MN, USA). A serum 25(OH)D concentration of ≤30 ng/ml was considered as vitamin D insufficiency (Paper I).

Paper IV

Similar vitamin D methods were used but analysed at the Hormone Laboratory, Aker University Hospital, Oslo, Norway. The serum concentrations of 25(OH)D and $1,25(OH)_2D$ were measured by RIA and competitive RIA, respectively (DiaSorin, Stillwater, MN, USA).

Other biochemistry

Paper I and III

Blood samples for creatinine, calcium and intact parathyroid hormone (PTH) were obtained at baseline and after the last dose of radiation. Serum PTH was measured using the immunochemical luminescence method (mass concentration), serum calcium by photometry, 600 nm and serum creatinine by using an enzymatic method (μmol/l). The serum concentrations of thyroid hormones, and osteocalcin, calcium and creatinine were measured before the first and after the last dose of radiation (Paper I). Thyroxine hormone (free T4) and thyroid-stimulating hormone (TSH) were examined by electrochemical luminescence ECLIA (nmol/l) and (mU/l) respectively. Serum osteocalcin was examined using an immunochemical radio immunoassay (μg/l).

Paper IV

The serum concentrations of calcium, ionised calcium (Ca^{++}), PTH, plasma folate, homocysteine, vitamin B12, erythrocyte haemoglobin (EHb), erythrocyte folate (EFo), erythrocyte hematocrit (EHct), C reactive protein (CRP) and micro CRP (mCRP) were obtained before the sun exposure, after one day and after 15 days of exposure. The serum concentrations of creatinine, glucose, Apolipoprotein A1 (APO-A1), Apolipoprotein B (APO-B), Lipoprotein A (Lp(a)), total cholesterol, high-density lipoprotein cholesterol (HDL), low-density lipoprotein cholesterol (LDL), triglyceride and glycosylated haemoglobin A_1c (HbA_1c) were analysed using routine laboratory methods, (Department of Medical Biochemistry, Rikshospitalet, Oslo, Norway) before and after 15 days of sun exposure.

Procedures of UV exposure and UV measurement

Paper I

Patients were treated with broadband UVB (280-320 nm, PhilipsTL12 and Corona 4, ESSHÅ, Elagentur, AB, Värnamo, Sweden) 2 to 3 times/week for 8 to 12 weeks (mean number of treatments 23.3±5.5) The radiation source was a fluorescent cabinet that enclosed subjects, consisting of fluorescent tubes mounted in a round assembly, placed at a distance of 30 cm apart from the body surface of the standing subject *(Figure 5)*. The irradiance was measured by PUVA Combi Light (ESSHÅ, Elagentur, AB, Värnamo, Sweden).

The subjects were exposed (whole body) to individually adjusted doses of UVB depending on skin phototype and erythemal response to therapy. The median cumulative UVB dose during the series of exposures was 0.9 J/cm^2 (range 0.4 to 5.8 J/cm^2).

Paper III

The patients were previously treated with UV therapy. The selection of UV lamp was based on patients' previous experience of the respective broadband or narrowband therapy. They were treated with the same lamp (NBUVB or TL12) throughout the study. Twenty-six patients were treated with broadband UVB (280-320 nm, Philips TL12 and Corona 4, ESSHÅ, Elagentur, AB, Värnamo, Sweden)

and 42 patients were treated with NBUVB (311-312 nm, Corona 4, ESSHÅ, Elagentur, AB, Värnamo, Sweden).

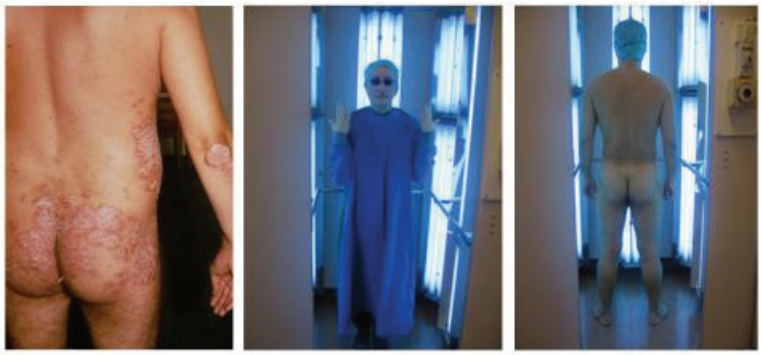

Figure 5: Psoriasis. Treatments with UVB lamp.

All patients were treated according to standardised schedule adjusted to each patient 2 to 3 times/week for 8 to 12 weeks. The patients reached approximately the same outcome with similar total number of treatments (mean number of treatments 27.2±5.3 in the broadband UVB treated group and 28.1±5.6 in the NBUVB treated group).

The radiation source consisted of fluorescent tubes mounted in a round assembly and placed at a distance of 30 cm apart from the body surface of the standing subject. Irradiation was measured by PUVA Combi Light (ESSHÅ, Elagentur, AB, Värnamo, Sweden).

Subjects were exposed (whole body) to individually adjusted doses of UVB depending on skin phototype and erythemal response to therapy. The median cumulative UVB dose during the series of exposures in the group treated with broadband UVB was 12 J/cm^2 and in the group treated with NBUVB 37 J/cm^2.

Paper IV

The study was carried out during 15 days in March 2006 at Valle Marina Treatment Centre, Gran Canaria (27ºN, 15ºW) *(Figure 6)*. The patients followed a strict exposure schedule the first day of the study, exposing first the front side of the body for 30 minutes, the back side for 30 minutes, followed by 15 minutes exposure of each side during the period between 11 am to 1 pm local time. They were allowed to stay outside after lunch, when an adequate amount (2 mg/cm^2) of

sunscreen with sun protection factor (SPF) of 25 (Pediatrics Photoprotector ISDN, 25B-10A-IR) was used for the whole body.[186] For remaining days patients were advised to gradually increase the hours of exposure per day, and to limit the use of sunscreen to parts of the body that are susceptible to burning.

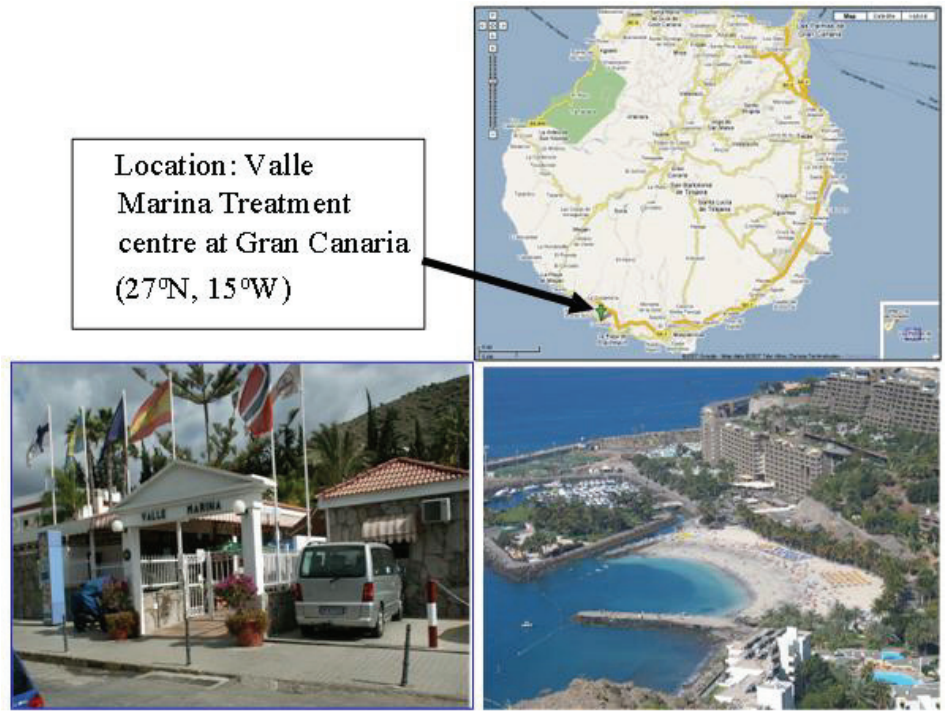

Figure 6: Valle Marina

The patients registered time spent in the sun every day per 20 minutes from 9 am to 5 pm local time, as well as the use/amount of sunscreen and SPF factor. Spectral UVB (280–315 nm), UVA (315–400 nm) and CIE-weighted UV irradiances were measured every hour using 2 broadband instruments (Solar Light Co PMA 2100 with UVB sensor PMA 2101 and UVA sensor PMA 2110, and Gigahertz-Optik GmbH X1 1 Optometer with UVB and UVA sensors XD-501).The CIE-action spectrum is a reference action spectrum for UV induced erythema in Caucasian human skin, valid for the UV region from 250–400 nm.[187] Sensors were calibrated and intercompared against a spectroradiometer (Brewer#185, measurement range 286.5–365 nm, extended for UVA 365–400 nm) at Izaña, Tenerife, prior to the study (by Mr. Alberto Redondas, Instituto Nacional de Meteorologia (INM), Spain) according to internationally accepted procedures *(Figure 7).*[188,189] The overall

measurement uncertainty can be estimated within ±25%, and is due to the uncertainty in different instruments, temperature variations, azimuth variations and non-ideal cosine response in broadband sensors. Spectral UVB and UVA irradiances, in addition to CIE-weighted UVB and UVA irradiances, were calculated for the whole period using a radiation transfer model, libRadtran for irradiance calculations.[190]

Figure 7: Calibrated broadband CIE-weighted UVB and spectral UVA-probes against spectroradiometer at Izaña, Tenerife, at the INM (Instituto Nacional de Meteorologia España), by Alberto Redondas

The model was run for the following conditions: cloudless sky, albedo of 0.05, sea level and ozone values from the TOMS satellite.[191] The UV irradiances were adjusted according to measurements taken at the treatment centre to account for the real weather situation and possible discrepancies from model parameters, such as different albedo and aerosol amount. UV doses were estimated for each patient after 1 day and after 15 days of sun exposure by combining calculated UV irradiances with sun exposure time from patients' diaries.

Results are presented as spectral UVB and UVA doses, as well as CIE-weighted UV doses in Standard Erythema Dose (SED) (1 SED = 100 J/m^2 = 0.01 J/cm^2). UV doses for each patient were set equal to the ambient UV doses divided by two, since only half the body can be exposed at any time. Doses on the first day

excluded exposure time when sunscreen is used, since patients reported using approximately the proper amount of sunscreen after lunch (30 ml, SPF 25).

All exposure time is included for remaining days since patients reported using small amounts of sunscreen only on easily burned locations.

Concomitant medication

The patients were without any psoriasis medication for 4 weeks prior to the interventions in study I, III and IV.

Questionnaires

Medical, family and gynaecological history, nutrient intake, physical activity, history of psoriasis, previous psoriasis treatment and sun exposure in Paper II were obtained with questionnaires. Previous (at the age of 25 years) body height and weight were asked for.

Exposure to sun during the summer was graded from 1 to 4, grade **1** was sun exposure <½ h/day, grade **2** was sun exposure ½–1 h/day, grade **3** was sun exposure 1–2 h, and grade **4** was defined as sun exposure >2 h/sunny summer day.

Time spent outdoors per day (independent of sun exposure) was registered and graded from 1 to 4 in a similar way.

Travel to sunny countries in the last two and ten years respectively was analysed and graded from 0 to 4. Grade **0** was no travel to sunny countries, grade **1** was 1-4 travels, grade **2** was 5-10 travels, grade **3** was 10-20 travels and grade **4** was >20 travels in the last 10 years.

Ethical considerations

Each patient was given written information about the aim of the study. The study was approved by the Ethics Committee at the University of Gothenburg and the Swedish National Data Inspection Board (Paper I, II and III) and Regional Committee of Medical Ethics and the Norwegian National Data Inspection Board (Paper IV). Declaration of Helsinki protocols was followed and the written informed consent of patients was obtained.

Statistics

Data are given as mean ± SD or median (min-max) unless otherwise stated. Simple descriptive statistics and univariate correlations were performed using the statistics routines of software (Excel, Microsoft Inc, SPSS, Version 15).

Student's paired *t*-test was used for comparisons of blood test results before and after sun exposure. Associations between variables were tested by Pearson and Spearman correlation analysis. Probability values (2-sided) were considered significant at values of <0.05.

Results

Comparison between the studies

Phototherapy induced vitamin D production in patients with psoriasis.

Serum levels of 25(OH)D increased during the treatment with artificial UV (broadband UVB ($p<0.00001$), NBUVB ($p<0.0001$)) and during the heliotherapy ($p<0.0001$) (Table 2, *Figure 8 and 9).*

	Postmenopausal women treated with broadband UVB (Paper I)	Broadband (Paper III)	Narrowband (Paper III)	Heliotherapy (Paper IV)
25(OH)D(ng/ml)				
before	36.8 ±17.0	37.9±16.9	34.8±11.9	22.9±6.0
after	59.6±18.7	69.4±19.7	55.3±17.6	41.8±6.3
$1,25(OH)_2D$ (pg/ml)				
before	53.2±17.0	59.4±16.8	62.1±25.6	58.6±16.8
after	61.7±11.0	66.5±19.3	62.0±19.1	73.1±23.7
PTH (ng/l)				
before	63.3±26.2	31.6±17.2	33.7±17.7	41.8±17.3
after	48.4±17.3	23.3±12.8	29.2±14.1	38.6±12.9

Table 2: Changes in serum concentrations of 25(OH)D, $1,25(OH)_2D$ and PTH in psoriasis patients treated with broadband UVB, NBUVB and heliotherapy. (Mean ±SD)

The increase in 25(OH)D was higher in the broadband treated patients compared to NBUVB treated group ($p=0.008$) and compared to patients treated with heliotherapy ($p<0.01$). The increase in 25(OH)D during two weeks of climatotherapy was similar to the increase in 25 (OH)D during the treatment with NBUVB.

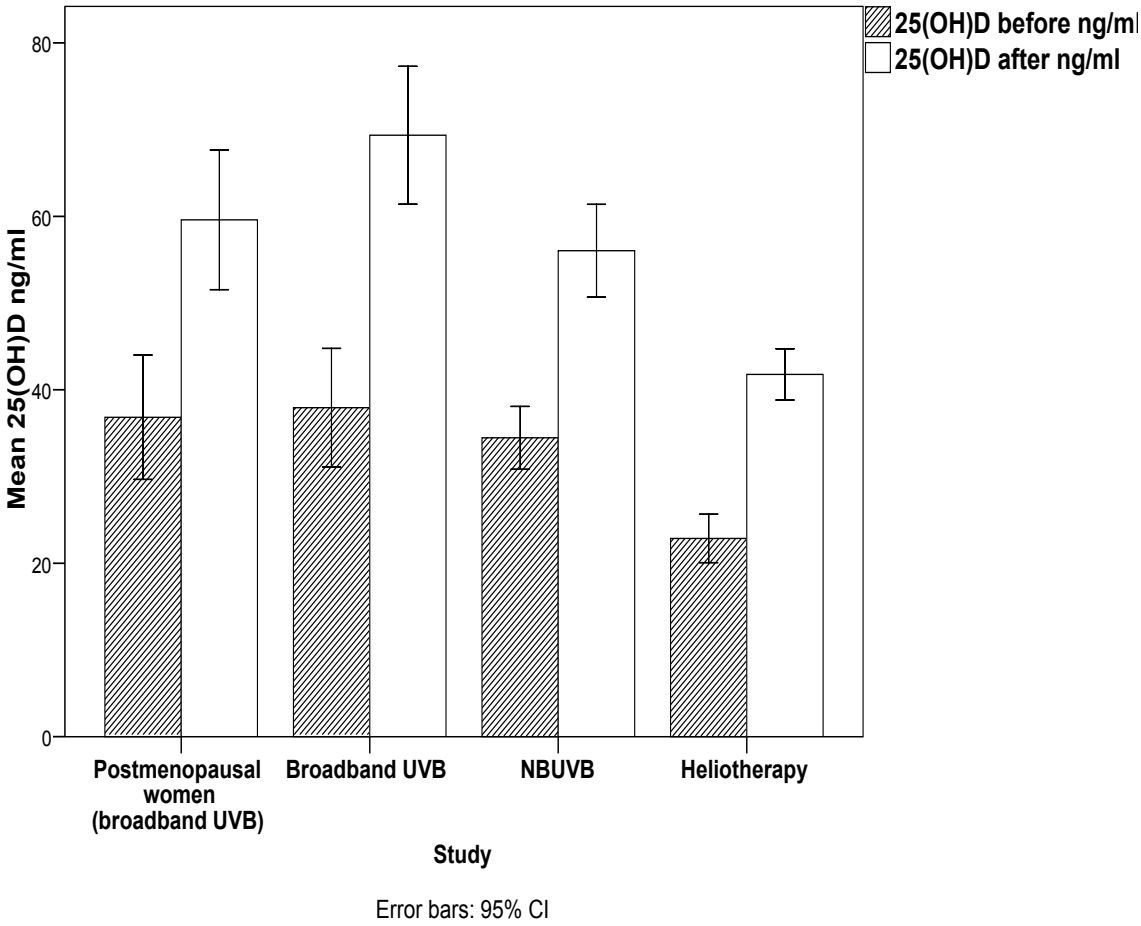

Figure 8: The mean 25(OH)D serum concentrations before and after the treatment with broadband UVB lamp (Paper I and III), NBUVB lamp (Paper III) and heliotherapy (Paper IV), respectively. Error bars: 95% CI.

The serum concentrations of 25(OH)D increased in postmenopausal women with psoriasis treated with broadband UVB. There was no difference in the increase of 25(OH)D between the postmenopausal women treated with broadband UVB, younger patients treated with broadband, NBUVB, or heliotherapy, respectively.

Patients treated with broadband UVB during winter months increased their serum levels of 25(OH)D from 28.5±8.8 to 64.6±24.1 ng/ml ($p<0.001$) and patients treated with NBUVB increased their 25(OH)D from 28.3±6.8 to 47.2±15.5 ng/ml

(p<0.001); (p=0.012 between lamps). Patients treated with 2 weeks heliotherapy increased 25(OH)D from 22.9±6.0 to 41.8±6.3 ng/ml (p<0.001), respectively, (p<0.015 between broadband UVB treated patients during winter and patients treated with heliotherapy).

Two weeks of heliotherapy were as effective as treatment with NBUVB 2-3 times per week for 2 to 3 months. Serum concentration of 25(OH)D at the start in Paper I and III was however, higher than in patients treated with heliotherapy (p=0.0001). The mean serum concentration of 25(OH)D after UV therapy was also higher in patients treated with artificial lamps (p<0.0001) compared with heliotherapy.

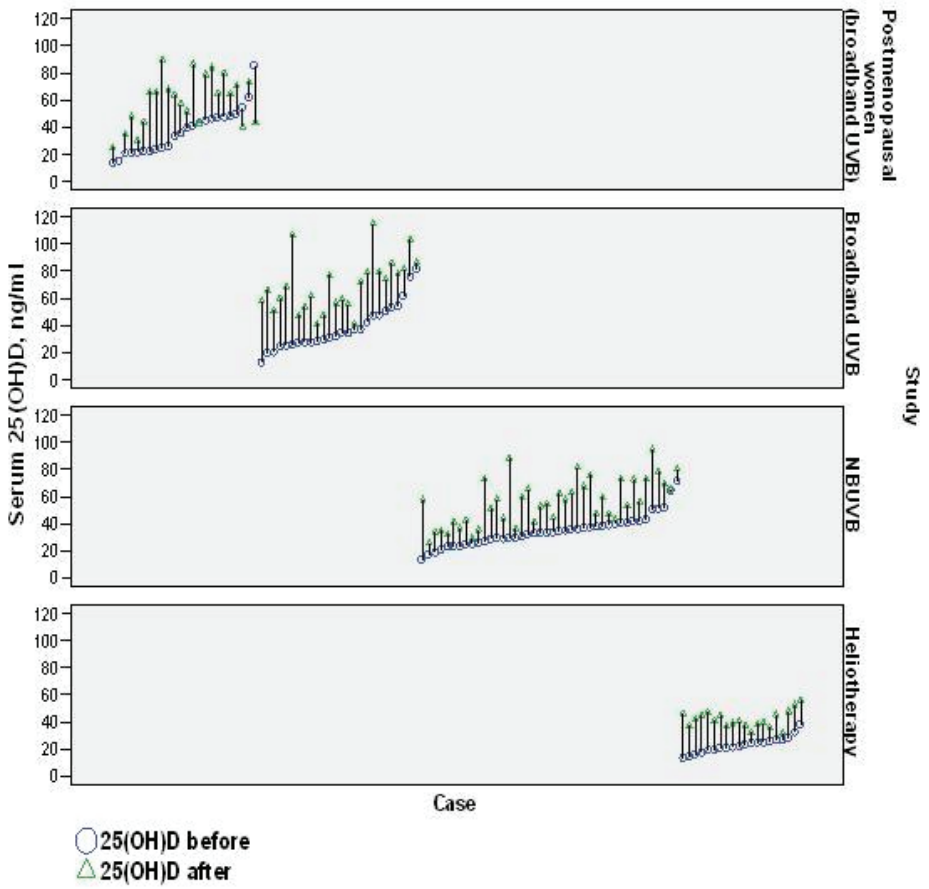

Figure 9: Changes in serum concentrations of 25(OH)D induced by broadband UVB, NBUVB and heliotherapy in each psoriatic patient.

PTH decreased after the treatment with broadband UVB and after heliotherapy (Table 2). The decrease in PTH was most prominent in the group of postmenopausal women treated with broadband UVB, Paper I.

The suppression of PTH in postmenopausal women treated with broadband UVB was higher than in patients treated with NBUVB (p=0.005), Paper III, and in patients treated with heliotherapy (p=0.026), Paper IV, respectively (Table 2, *Figure 10 and 11).*

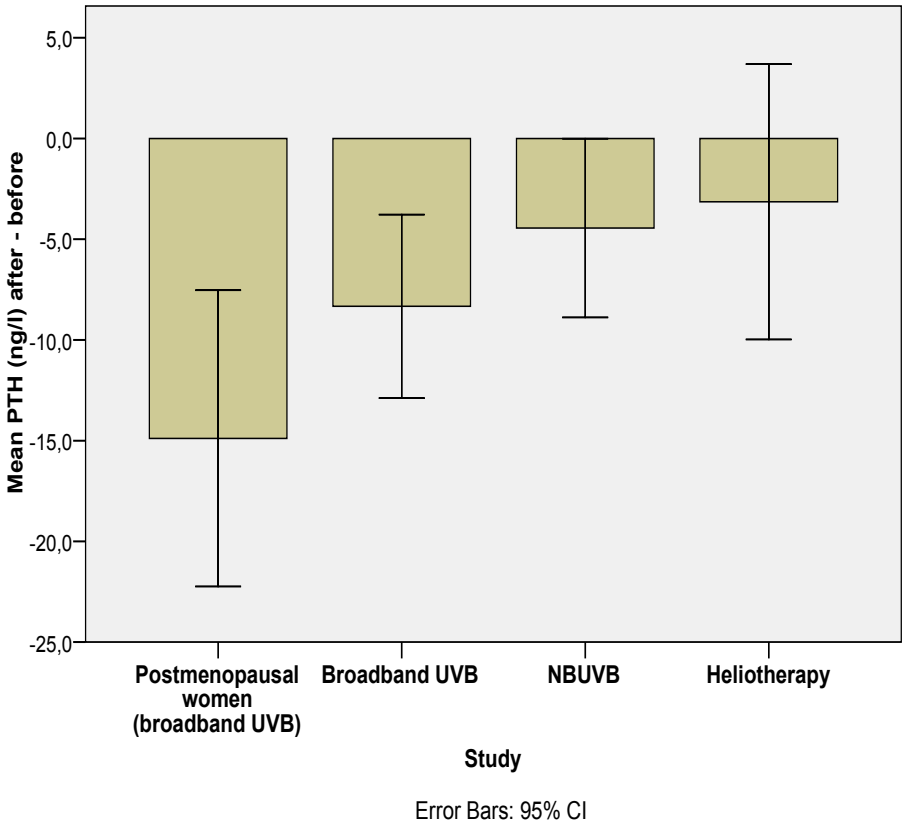

Figure 10: Changes in serum PTH after different phototherapies in psoriatic patients.

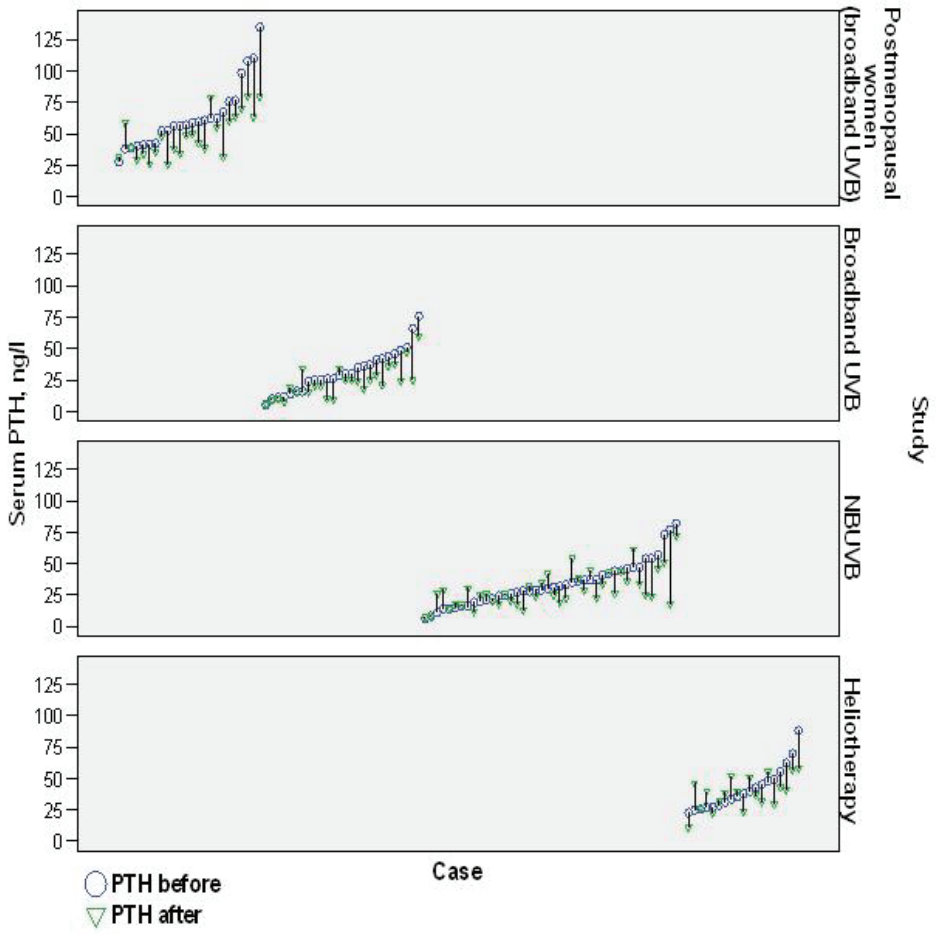

Figure 11: Changes in serum concentrations of PTH induced by broadband UVB, NBUVB and heliotherapy in each psoriatic patient.

1,25(OH)2D increased more during the heliotherapy than with NBUVB (p=0.02). There was no correlation between the dose of UVB and the increase of 25(OH)D, or $1,25(OH)_2D$ respectively.

Psoriasis improved in all patients, with a reduction in PASI score of about 75% on all regimens. PASI at start was similar in all groups and improved similarly on all regimens.

Paper I

Twenty-four women aged 60–81 years with a mean BMI of 27.7±4 kg/m^2 were treated with broadband UVB, 2–3 times/week during 8–12 weeks. Psoriasis improved in all patients, with a reduction in PASI from 6–12 before to 1–4 after UV therapy.

Vitamin 25(OH)D

Serum levels of 25(OH)D increased from 36.8 ±17.0 ng/ml before the UVB treatment to 59.6 ± 18.7 ng/ml after ($p<0.001$) (Table 2, *Figure 8*).

Two patients showed a reduction in their circulating vitamin D levels following phototherapy. Both subjects had high level of 25(OH)D at the start of the treatment – one subject due to travel to a sunny country one month before the UVB therapy, and one subject commenced her course of UVB at the start of the autumn following summer, so their vitamin D levels were falling after the summer peak.

Before UVB therapy, a serum concentration of 25(OH)D below 30 ng/ml was found in 10 patients (42%) and serum concentrations of 25(OH)D of >30 ng/ml were present in 14 patients. All increased 25(OH)D after UVB ($p=0.001$), while PTH decreased ($p=0.001$). The increase in 25(OH)D was enhanced in patients with low vitamin D values.

The serum concentration of osteocalcin was similar in patients with low and normal vitamin D before and after UVB therapy.

Six of the patients had taken vitamin and mineral supplements (vitamin E, Omega 3 and multivitamins including vitamin D3 (200 IU/day)). Despite this intake of supplements, two of these patients had vitamin D insufficiency (25(OH)D <30 ng/ml). All these women experienced an increase in serum 25(OH)D similar to that in patients without supplements.

Serum calcium, osteocalcin, thyroid hormones and creatinine were unaltered after the UVB therapy.

Serum PTH

Serum PTH decreased after UVB exposure from 63.3±26 ng/l to 48.4±17.3 ng/l ($p<0.001$) in all women (Table 2, *Figure 10*).

Serum PTH decreased from 69.6±30 ng/l to 49.8±17 (=29%) ($p<0.001$) in patients ≥70 years of age and from 55.9±19 ng/l to 46.6±19 (=17%) ($p=0.039$) in patients <70 years of age.

Secondary hyperparathyroidism (PTH >65 ng/l) was found in 7 patients (29%) before treatment. In these patients, the median PTH value decreased from 98.2 ng/l before the UVB therapy to 63.7 ng/l (54%) after ($p< 0.001$). None of the patients had hypercalcaemia. The median 25(OH)D in these patients was 22.6 ng/ml before UVB treatment and 64.7 ng/ml (186%) after.

Bone status

The mean Z-score value (SD from the mean peak value in age-matched adults) was +0.54 SD (±1.1) at the hip and +0.50 SD (±1.4) at the lumbar spine.

The mean T-score value (SD from mean peak value in young adults) was -0.90 SD (±1.15) at the hip and -1.49 SD (±1.4) at the lumbar spine.

Normal BMD (T-score >-1 SD) was found in 10 of the youngest patients, osteopenia (T-score between –1 to -2.5 SD) in 5 patients and osteoporosis (T-score <-2.5 SD) in 9 patients.

Mean serum levels of 25(OH)D before the UVB treatment were 38.4±21 ng/ml in the group with normal BMD, 36.0±13 ng/ml in the group with osteopenia and 35.5±16 ng/ml in the women with osteoporosis (ns. between groups).

After the UVB treatment, serum 25(OH)D increased in all groups ($p<0.01$), respectively. The mean serum level of $1,25(OH)_2D$ did not differ between the different BMD groups before or after the UVB treatment. There was no relationship between psoriasis onset and bone status. Any changes in BMD were not expected to occur in 8-12 weeks of UV therapy.

Paper II

Anthropometry and bone status

Thirty-five women with psoriasis were compared with 2448 age-matched control women from the Geriatric out-patient clinic, Göteborg, regarding risk factors for osteoporosis. Both the Z score and T score at the hip and lumbar spine were higher in women with psoriasis than in controls.

A positive family history for kyphosis was less common in patients than in controls (p=0.0086). The duration of HRT was shorter in patients than in controls (p=0.045).Prevalence of myocardial infarction and angina pectoris was more common in the patient group. Patients had higher consumption of cheese and coffee and were more physically active than controls. The degree of physical activity in patients was 3.7±2.2 of 1–4 (low to high) per week vs. 1.8±1 in controls (p=0.0001).

Subjects with normal BMD (n=14) were younger (67±6 years) than the patients with osteopenia and osteoporosis (72±6 and 70±6 years, respectively). BMI and the number of UVB treatments did not differ between groups. The age at onset of psoriasis correlated positively with BMD in all patients.

All fractures occurred after falling during patients' leisure time and 87% involved a wrist fracture.

The patients with normal BMD had highest childbirth, latest menopause and longest duration of HRT.

Milk consumption varied from 0 to 5 glasses per day but did not differ between groups. Coffee consumption varied between 1 to 8 cups/day and was highest in the patients with osteoporosis. Vitamin and mineral supplementations (vitamin E, omega 3, calcium or multivitamin) did not differ between groups. Current smoking was more common in patients with osteopenia and osteoporosis.

Physical leisure time activities were highest in patients with normal BMD. The activities were mainly described as Grade **3 (**regular exercise within different activities depending on the season such as swimming, skiing, walking and gardening).

Sun exposure of two hours or more during summer was highest in patients with normal BMD. The patients with normal BMD had the highest number of trips to sunny countries in the last 2 years. Spending time outdoors (independent on sun exposure) was similar in all groups and varied between 1–2 h/day.

None of the patients had systemic treatment with corticosteroids but 27 (77%) had topical treatment with cortisone ointments alone or in combination with vitamin D analogue (calcipotriol). Patients with normal BMD used more calcipotriol while osteoporosis patients used more topical corticosteroid.

Paper III

Twenty-six psoriatic patients (7 women and 19 men) with broadband UVB treatment were compared with 42 (10 women and 32 men) psoriatic patients with NBUVB treatment for 8 to 12 weeks.

Serum 25(OH)D increased after both broadband and NBUVB (Table 2, *Figures 8 and 9*). The increase in 25(OH)D was higher in the broadband treated patients ($p=0.008$) compared with NBUVB. Serum PTH decreased on broadband UVB ($p<0.05$).

Secondary hyperparathyroidism (PTH >65ng/ml) was present in 5 men and their PTH values normalised after UVB therapy. 25(OH)D increased similarly in patients with 25(OH)D <30ng/ml as in patients with 25(OH)D ≥30 ng/ml at baseline. There were no changes in 1,25 $(OH)_2D$, creatinine or serum calcium in the groups after treatment.

There was no correlation between the dose of UVB and the increase of 25(OH)D. The treatment time was four times longer for the NBUVB treated patients than for patients treated with the broadband UVB.

Patients with skin type II (n=23), obtained as expected, a lower median dose of UVB (broadband 6.9 J/cm^2 and narrowband 31.4 J/cm^2) than patients with skin type III (n=44, dose of broadband UVB 16.4 J/cm^2 and 51.8 J/cm^2 dose of NBUVB) and IV (n=1, treated with broadband UVB, dose 15.7 J/cm^2). Patients with skin type II (n=23) increased in 25(OH)D from 33.8±16.8 to 56.5±20.7 ng/ml ($p<0.001$) and patients with skin type III (n=44) increased from 36.9±12.3 to 64.0±18.0 ng/ml ($p<0.001$), respectively (ns between skin type II and III).

Psoriasis improved in all patients, with a similar reduction in PASI score in both groups. Improvement in psoriasis correlated positively with increase in 25(OH)D ($p=0.047$).

The baseline 25(OH)D levels were lower in those who started in winter compared with those who started during spring ($p=0.0001$).

The age difference was explained by the later introduction of the NBUVB lamps (younger patients).

Paper IV

Sun exposure for 15 days led to reduction in the PASI score in patients with psoriasis. Furthermore, the sun exposure increased serum concentrations of both 25(OH)D and $1,25(OH)_2D$.

Patients with 25(OH)D ≤ 20 ng/ml (50 nmol/l) at baseline (1 woman and 5 men) increased more in 25(OH)D than patients with 25(OH)D >50 nmol/l (p=0.03).

The serum concentrations of $1,25(OH)_2D$ increased after 15 days of sun exposure (p=0.01 day 1–15 and p=0.004 day 2–15).

The serum concentrations of PTH decreased (p=0.04 day 2–15).

BMI was unaltered during the study period. The sun exposure improved the lipid and carbohydrate status of the patients. The LDL/HDL cholesterol ratio decreased from 2.4 to 1.9 (p<0.001). The serum concentrations of APO-A1, EHb, folic acid, HDL, homocysteine and uric acid increased during 15 days of climate therapy. The serum concentrations of APO-B, vitamin B12 and HbA_1c decreased during the sun exposure period. The serum concentrations of calcium, creatinine, EHct, glucose, CRP, mCRP, total cholesterol, LDL, Lp(a) and triglyceride were not influenced by climate therapy.

Diastolic blood pressure increased from 84.5±9.7 to 90.9±10.6 mmHg (p=0.007) while systolic blood pressure and pulse were unaltered after 2 weeks of sun exposures.

Daily sun exposure time per patient increased throughout the treatment period. The patients received on average 5.1±2.3 SED (median=4.0 SED), range 2.6–10.3 SED on the first day of exposure. The mean dose was similar for the patients with skin type II and III. The mean dose after 15 days sun exposure was 166±25 SED (range 104–210), 135 and 169 SED for the patients with skin type II and III, respectively. The variation between minimum and maximum patient doses each day was large. The patients´ sunbathing with sunscreen during the first day reported a use of approximately 30 ml of cream each on body sites easily burned. Although 14 out of the 20 patients reported erythema after the first day of sun exposure, but none reported blistering erythema.

The reduction in PASI score was 73%, but there was no correlation between the improvement in PASI score and vitamin D or the UV dose, respectively.

Furthermore, we did not find any correlation between changes in the single component of PASI score, including area, erythema, infiltration and desquamation, and the dose of received UVB.

Serum concentration of $1,25(OH)_2D$ at baseline correlated positively to serum concentration of 25(OH)D at baseline ($p<0.0005$) and negatively to increase of 25(OH)D ($p=0.023$). The increase in 25(OH)D correlated to the increase in $1,25(OH)_2D$ after 15 days of climate therapy ($p<0.0005$). Serum concentrations of 25(OH)D at baseline correlated positively to serum HDL at baseline ($r=0.46$, $p=0.040$).

Discussion

Serum 25(OH)D in psoriasis patients during treatment with phototherapy (Papers I, III and IV)

Serum 25(OH)D levels increased in psoriasis patients following treatment with heliotherapy, broadband UVB and NBUVB phototherapy. Psoriasis improved in all patients, with a reduction in PASI score of about 75% on all regimens. UVB and sun exposure are the strongest factors influencing 25(OH)D.[8,18,33,138,192] The same wavelength of the UVB spectrum (290-315 nm) that is responsible for D3 vitamin synthesis in the skin also improves psoriasis lesions, and has therefore been used in psoriasis therapy.

Vitamin D3 production in patients with psoriasis increased less with NBUVB than with broadband UVB phototherapy. One explanation might be that the optimal wavelength for initiation of the vitamin D3 pathway was 300±5 nm in vitro and in vivo[193] which is in the broadband UVB range (280-320 nm). The synthesis of vitamin D3 was stimulated by wavelengths between 290-315 nm, but not longer than 315 nm. The present results from our study (Paper III) showed that a wavelength of 311 nm was effective for inducing vitamin D3 synthesis, but not to the same extent as wavelengths in the broadband UVB range. UVB treatment of psoriasis was a sufficiently time-consuming procedure to increase vitamin D3 also with NBUVB. The time required for NBUVB to have an effect can reduce the difference in the potential for vitamin D3 production between the two lamps. The treatment time correlated strongly with the type of lamp (patients treated with NBUVB required 4 times longer exposure times than patients treated with broadband UVB). This is consistent with other studies demonstrating that the dose response of the erythemal spectra of NBUVB should be about 4.2 times that of broadband UVB [194]. The dose of UVB also correlated with the type of lamp, but we could not find any correlation between the dose of UVB and the increase of 25(OH)D levels. This might be explained by autoregulation of skin synthesis, storage, and slow, steady release of vitamin D3 from the skin into the circulation.[8]

The serum concentrations of 25(OH)D almost doubled during 15 days of climate therapy (Paper IV). Patients with lower 25(OH)D levels at baseline responded better to sunlight and phototherapy (Paper I and IV) which is consistent with other

studies.[195] All patients reached serum levels of 30 ng/ml (75 nmol/l) after two weeks of sun exposure. A circulating level of 25(OH)D of >30 ng/ml, or >75 nmol/l, appears to be necessary to maximize the health benefits of vitamin D.[195] Sun exposure is the major source of vitamin D3 for most humans.[195] Skin pigment, sunscreen use, aging, time of day, season, and latitude all affect previtamin D3 synthesis.[196]

The ability of the skin to produce vitamin D3 declines with age [12] due to insufficient sunlight exposure[29,197] and a reduction in the functional production capacity of the skin.[12,13,198] All the patients in our study (Paper I) were post-menopausal women and we were unable to see any clear negative correlation between age and vitamin D3 synthesis in line with a previous population study carried out in the same city.[138] Age did not correlate with the increase in 25(OH)D in our intervention studies (Paper I, III, and IV).

The increase in 25(OH)D during 2 weeks of heliotherapy was very similar to the increase in 25(OH)D during treatment with broadband UVB and NBUVB for 2 to 3 months (Paper I and III). During prolonged exposure to the sun, the accumulation of previtamin D3 is limited to about 10 to 15% of the original 7-DHC content, because the previtamin also undergoes photoisomerization into two biologically inert photoproducts, lumisterol-3 and tachysterol-3.[8]

The vitamin D3 production is a unique, autoregulated mechanism which occurs at two levels. Excessive sun exposure does not lead to overdosing of vitamin D3 due to conversion of previtamin D3 to inactive photoproducts (lumisterol 3 and tachisterol 3) as well as conversion of vitamin D3 to its isomers in the skin (5,6-trans vitamin D3, supersterol I, supersterol II) which are thought to have low calcemic effect at physiological concentrations. Vitamin D3 is synthesized in the skin and released steadily and slowly from the skin into the circulation.[8]

There was no difference in the increase of 25(OH)D between the different skin types in the present studies. The reason could be that the subjects were exposed to individually adjusted doses of UVB depending on skin phototype and erythemal response to therapy. All patients had previous experience of UVB therapy for their psoriasis disease. As expected, fair-skinned patients required lower doses of UVB (broadband and narrowband) than patients with skin type III and IV. This finding is consistent with other studies examining the effect of skin pigmentation on

vitamin D3 synthesis.[199] Melanin pigment in human skin competes for, and absorbs the UVB photons responsible for the vitamin D3 synthesis.[199]

During the winter months vitamin D3 production is insufficient to meet the optimal requirements in both younger and older adults at Northern latitudes.[200] As seen in a previous study[201], lower baseline 25(OH)D levels were found in those who initiated treatment in winter. Psoriasis lesions usually deteriorate during winter, and many patients are therefore given repeated UVB treatment during this season. In addition to healing psoriasis lesions, UVB therapy also provides these patients with vitamin D3 during the winter months, when levels of 25(OH)D in Northern countries are generally low. Broadband UVB induced vitamin D3 production and suppressed PTH (Paper I, III and IV). The increase in 25(OH)D was similar for the broadband and NBUVB lamps during the spring period. The influence of ambient UVB on vitamin D status during the spring months (March-June) might explain this result.

We found no correlation between the increase of the dose of UVB and the increase of serum 25(OH)D levels within the groups. This might be due to the fact that serum concentrations of 25(OH)D were measured at different time points and a plateau level was reached after three weeks, which had also been seen in a previous study.[202] A recent in vitro study has demonstrated that the dose-response relationship of UV exposure and cholecalciferol synthesis was nonlinear. It was hypothesized that exposure to additional UV may not result in a proportional increase in vitamin D levels.[203]

The correlation between sunlight measures and serum 25(OH)D has been shown to be weak.[204] Patients (Paper IV) reached their plateau of daily sun exposure after the first week. It might be that the vitamin D3 production was most prominent during the first week, when the patients had experienced redness and some of them even got sunburned.

The increase of 25OHD during 15 days of climate therapy was significant even though the patients used sunscreens on body sites susceptible to sunburn, and even though the skin was affected by psoriasis lesions. An SPF-8 sunscreen reduces the skin's production of vitamin D3 by 95%. Clothing completely blocked all solar UVB radiation and thereby prevented vitamin D3 production.[15]

We found no correlation between the reduction in PASI score and serum concentrations of 25(OH)D during climate therapy, consistent with another study

(Paper I). Furthermore, there was no dose-dependent correlation between vitamin D metabolite levels, PASI score and the received UV dose.

Improvement in psoriasis correlated positively with an increase in 25(OH)D3 levels in Paper III ($p=0.047$; the group of patients treated with broadband UVB and NBUVB) but not in the other studies (Paper I and IV). We do not have a good explanation for this. Instead we propose that a larger population of psoriasis patients should be examined to clarify this finding.

It has been alleged that NBUVB is more effective than broadband UVB for reducing PASI scores, as well as safer and better tolerated by patients than PUVA when taken at suberythemogenic doses.[205] The NBUVB lamp seemed to be easier to handle and better tolerated, giving it some advantages over the standard broadband UVB[206] which has led to a reduction in the usage of broadband UVB. One of the drawbacks with the new lamp is that the radiation times are almost doubled.[207] The reduction of PASI scores in our study (Paper III) was similar in all patients, irrespective of the choice of lamp. The patients were not allowed to apply calcipotriol to psoriasis lesions during the study and several patients noticed a somewhat delayed effect of the UVB therapy on the healing process. Several studies have shown more rapid healing of psoriasis with the combination of calcipotriol and UVB radiation than with monotherapy with either treatment.[102,208]

Our method for measuring and estimating UV doses (Paper IV) is simpler compared with using personal UV dosimeters, but it is associated with more uncertainties. These are discussed by Snellman et al[209], but the predominant uncertainty is probably due to the fact that the skin dose equals half the ambient dose. This is appropriate as a first approximation, in particular for the abdomen and back during sun bathing if the patients turn to expose these sites to an equal extent. Measurements using personal dosimeters in other studies support this assumption.[209-211] For the extremities, doses are reported to be higher or lower depending on activities, and all vary more than during sun bathing.[209,211,212] The upper extremities probably often receive more than 50% of the ambient UV[209-212] and are therefore more susceptible to sunburn. These sites are often covered with sunscreen through the treatment period.

The correlation between personal report of sun exposure and ambient UV light measured by dosimeter (assessment of radiation dose) is weak.[204]

The positive correlation between serum 25(OH)D and serum $1,25(OH)_2D$ (Paper IV) suggests that the production of the latter is substrate-dependent in vitamin D-deficient patients.[136,213] Serum concentration of 25(OH)D at baseline was lower in patients treated with heliotherapy ($p=0.0001$) than in patients treated with broadband UVB and NBUVB, and in postmenopausal women with psoriasis treated with broadband UVB.

Serum 25 OH vitamin D levels in the postmenopausal psoriatic women before UVB therapy (Paper I) were similar to those in age- and location-matched control women.[138] The serum concentration of 25(OH)D at baseline was similar in the patients treated with broadband UVB and NBUVB, and in postmenopausal women with psoriasis treated with broadband UVB. After broadband UVB treatment, serum 25(OH)D increased whilst serum PTH decreased. In addition to the positive calciotropic effect of vitamin D on bone metabolism and the suppression of PTH[214,10], vitamin D also plays an important role in healing psoriasis.[215-217] This indicates that the positive effect of UVB on psoriasis might be due to vitamin D3 production in the skin.

The increase in serum 25(OH)D found after UVB exposure was consistent with the results from sun exposure studies, where sunlight was the strongest factor influencing serum vitamin D in women in the general population.[79,138]

The regular use of sunbeds that emits vitamin D-producing ultraviolet radiation is associated with higher 25(OH)D concentrations in healthy adults, and may thus be of benefit to the skeleton in vitamin D insufficiency.[79] Others have found that vitamin D3 is more efficacious than vitamin D2 (the major vitamin in supplements) for increasing serum 25(OH)D levels.[218] UVB therapy increased serum 25(OH)D levels even in patients taking vitamin D supplements. This is in line with previous studies which reported that UV-induced vitamin D3 synthesis had a greater influence on the serum levels of circulating calcidiol than the peroral intake of supplements.[10,218]

The controlled use of UVB is a safe and effective method for the treatment of psoriasis[151] and could be used as treatment for vitamin D deficiency.[136]

The cut-off level for serum 25(OH)D, which is used as a diagnostic marker for vitamin D deficiency, has varied over the years.[33-35] The early biochemical changes in vitamin D insufficiency include a rise in serum PTH, which begins to increase as serum 25(OH)D levels fall below 30 ng/ml or 75 nmol/l.[35] This level of 25(OH)D

has become the suggested cut-off point for vitamin D deficiency or inadequacy.[29,30,35,219]

In those studies, it is not possible to draw conclusions about the ability of psoriatic skin to produce vitamin D3. A larger study is needed to examine the correlation between PASI and the capacity of psoriatic skin to produce vitamin D3 during UVB exposure.

Serum $1,25(OH)_2D$ in psoriasis patients during treatment with phototherapy (Papers I, III and IV)

Vitamin D undergoes metabolism in the liver to 25(OH)D, and then in the kidney to a number of metabolites, the most important of which is $1,25(OH)_2D$. This is the classical pathway and quantitatively the most important for producing $1,25(OH)_2D$. However, the keratinocyte is fully capable of producing its own $1,25(OH)_2D$.[46] The skin is the only tissue yet known in which the complete UVB-induced pathway from 7-DHC via intermediates (previtamin D3, vitamin D3, 25(OH)D) to the final product $1,25(OH)_2D$, takes place under physiological conditions.[26] Levels of $1,25(OH)_2D$ tended to increase during phototherapy, but statistically significant increases were noticed only during heliotherapy, and only in postmenopausal women with 25(OH)D3 below 30 ng/ml, and in women aged ≥ 70 years. One explanation might be that these patients had lower serum concentrations of 25(OH)D at the start of the treatment.

It has been postulated that the synthesis of $1,25(OH)_2D$ is tightly regulated, and that increases in 25(OH)D concentrations due to exposure to sunlight have no effect on serum $1,25(OH)_2D$ levels.[195,220] Some other studies[221,222] have shown a positive correlation between serum 25(OH)D and serum $1,25(OH)_2D$ in vitamin D-deficient patients, indicating substrate-dependent synthesis of $1,25(OH)_2D$. The similar observation that both 25(OH)D and $1,25(OH)_2D$ increased in vitamin D deficient persons following UVB exposure[223] or after vitamin D supplementation[221] has been reported previously. Parathyroid hormone is a well-known stimulator of $1,25(OH)_2D$ synthesis. The positive correlation between $1,25(OH)_2D$ and intact PTH reflects the physiological effects of PTH on the $1,25(OH)_2D$ response to low calcium. The positive relationship between PTH and $1,25(OH)_2D$, and the inverse relationship between PTH and 25(OH)D has been shown in a previous study from

the same city.[224] The persistent elevation of $1,25(OH)_2D$ concentrations in patients with low levels of 25(OH)D might be explained by PTH-induced chronic stimulation of the renal 25(OH)D-1α-hydroxylase.[223]

The increase of $1,25(OH)_2D$ levels between patients treated with heliotherapy and patients treated with NBUVB differed ($p=0.02$). This might be also explained by lower values of 25(OH)D at baseline in patients treated with heliotherapy.

Keratinocytes are capable of producing a variety of vitamin D metabolites, including $1,25(OH)_2D$, $24,25(OH)_2D$, $1,24,25(OH)_3D$[46] from exogenous and endogenous sources of 25(OH)D. The process by which $1,25(OH)_2D$ is produced and catabolised is tightly regulated and coupled to the differentiation of these cells. The epidermis is likely to contribute to circulating levels of $1,25(OH)_2D$, as human keratinocytes rapidly and extensively convert 25(OH)D to $1,25(OH)_2D$.[46,225] Peak intracellular levels of $1,25(OH)_2D$ are reached within 1 hour of adding 25(OH)D.[46] However, when renal production of $1,25(OH)_2D$ is normal, circulating levels of $1,25(OH)_2D$ are sufficient to limit the contribution from epidermal production.[225] It is now clear that epidermal 1α-hydroxylase and renal 1α-hydroxylase are identical proteins. The 1α-hydroxylase molecule is a 56-kDa protein with 506 amino acids.

Production of $1,25(OH)_2D$ regulates itself in the cell through a negative feedback loop which is similar to that observed in the kidney, but differs from that seen in the macrophage where this feedback is missing.[46] An important difference between keratinocytes and renal cells in the regulation of 25(OH)D metabolism by $1,25(OH)_2D$ is that the concentration of $1,25(OH)_2D$ required to inhibit the 1α-hydroxylase and induce 24-hydroxylase in renal cells appears to be several times greater than that required to achieve comparable effects in keratinocytes.[46,225] Thus, $1,25(OH)_2D$ production by keratinocytes is extremely sensitive to exogenous calcitriol. Acute nephrectomy leads to a very low extrarenal production of $1,25(OH)_2D$[226], and pig skin perfused with 25(OH)D showed low amounts of calcitriol which increased after 4-8 hours of perfusion.[227] In contrast to the renal 1α-hydroxylase which is tightly regulated by feedback mechanisms, the extrarenal 1α-hydroxylase is only constitutively expressed.[21] As a consequence, there should be no relevant feedback mechanisms between the systemic and epidermal vitamin D pathways that ultimately depend on UVB radiation.[21] Thus, the local UVB-triggered production of calcitriol may primarily regulate epidermal cellular functions in an auto- and paracrine manner, but this should not be crucial for systemic

vitamin D effects[21] and systemic vitamin D deficiency does not stimulate epidermal synthesis of $1,25(OH)_2D$.[228]

Cutaneous production of $1,25(OH)_2D3$ may regulate growth, differentiation, apoptosis and other biological processes in the skin.[229] The NBUVB has been shown to have less capacity to induce a local skin production of $1,25(OH)_2D3$ at 44% of the monochromatic irradiation at 300 ±2.5 nm.[193]

There is no information in the literature whether psoriatic skin is capable of synthesizing vitamin D.

It is not clear if the serum 25(OH)D level is linked to the level of the active form of vitamin D3 present in the skin. It has been suggested that cutaneous conversion of 25(OH)D to $1,25(OH)_2D$ does not play a role because the amount of free 25(OH)D3 that penetrates the cell membrane of epidermal keratinocytes is too small to produce sufficient amounts of $1,25(OH)_2D$.[229] The main form of circulating 25(OH)D is presented in a complex with vitamin D-binding protein (DBP) with only a very small amount (0.03%) available as the free form. Furthermore, the deeper layers of the epidermis are not vascularised, which further impairs the passage of the 25(OH)D3-DBP complex from blood to epidermal keratinocytes.[229]

We found no correlation between reduction in PASI score and serum concentrations $1,25(OH)_2D$. Nevertheless, the known therapeutic effect of UVB light therapy for the treatment of psoriasis may be mediated via UVB-induced production of $1,25(OH)_2D$.[26] The calcitriol concentration in UVB-treated skin measured by microdialysis was dependent on the UVB dose.[230] In vitro studies have shown that the substrate concentration of cholecalciferol in keratinocytes mainly determines the synthesis rate of $1,25(OH)_2D$ in these cells.[231] Thus, higher synthesis rates of cholecalciferol should result in a faster and more pronounced release of $1,25(OH)_2D$ into the extracellular fluid. UVB-induced membrane damage to epidermal keratinocytes may also increase the outflow of newly synthesized calcitriol.[230]

The $1,25(OH)_2D$ molecule and its analogues, as well as UVB phototherapy, exert antiproliferative, prodifferentiative, and immune-modulatory effects on keratinocytes that are of particular importance for the therapy of hyperproliferative skin diseases such as psoriasis vulgaris.[21,232] However, the full range of UVB and vitamin D3 effects is not completely understood.

The observation that $1,25(OH)_2D$ induces keratinocyte differentiation was first made by Hosomi et al.[233] and provided an explanation for the earlier and unexpected finding of $1,25(OH)_2D$ receptors in the skin. $1,25(OH)_2D$ is likely to be an autocrine or paracrine factor for epidermal differentiation, since it is produced by the keratinocyte. The receptor for, and the production of $1,25(OH)_2D$ vary with the differentiation in a manner suggesting feedback regulation;[234] both are reduced in the later stages of differentiation. $1,25(OH)_2D$ increases involucrin, transglutaminase activity, and cornified envelope formation in keratinocytes.[233]

Serum PTH in psoriasis patients during treatment with phototherapy (Papers I, III and IV)

Plasma concentrations of 1,25(OH)2D3 (calcitriol) depend mainly on renal function, plasma concentrations of intact PTH, and dietary intake of calcium and phosphate. High PTH and low dietary intake of calcium and phosphate are the main stimulators of the production of calcitriol. PTH increases with increasing age, possibly due to less sunlight exposure and/or reduced calcium/vitamin D intake.[224] The clear concomitant decrease in serum PTH after UVB exposure in our studies indicates that the skin has a good ability to synthesise vitamin D3 even at high age and with part of the skin covered by psoriasis lesions. Our results indicate that in women aged >70 years, UVB therapy radically increased vitamin D levels and suppressed PTH levels in serum. However, after adjustment for age, sun exposure was the only external factor that influenced serum vitamin D.[138] This is in line with the positive effect of UVB on the vitamin D3 synthesis in the women in our study, irrespective of age.

Serum PTH levels in postmenopausal women with psoriasis was higher both before and after UVB exposure compared with the population sample from the same region.[224] We found 7 patients (Paper I) and 5 patients (Paper III), respectively, with secondary hyperparathyroidism due to vitamin D deficiency. In these patients, PTH was suppressed by the UVB-induced increase in serum vitamin D3. Irradiation with UVB was as effective as oral vitamin D3 for increasing serum 25(OH)D and suppressing secondary hyperparathyroidism.[136] 25(OH)D concentrations below 30 ng/ml (75 nmol/l) resulted in secondary hyperparathyroidism and a decrease in BMD.[235]

Decreased renal function, oestrogen deficiency and low calcium intake may also induce secondary hyperparathyroidism.[30] Broadband UVB induced vitamin D3 synthesis and suppressed PTH (Papers I, III and IV).

Bone status in postmenopausal women with psoriasis treated with UVB phototherapy (Paper I and II)

Postmenopausal women with psoriasis (Paper I and II) had higher BMD both of the hip and the lumbar spine compared with age-matched controls.

Multiple risk factors that contribute to low serum 25(OH)D and osteoporosis have been identified. They include inadequate sun exposure[18], insufficient intake of fortified foods or vitamin D supplements[236], low body mass index, white ethnicity, lack of exercise, use of medications that accelerate vitamin D metabolism, diseases that alter vitamin D metabolism such as malabsorption syndromes, and chronic liver disease.[30,35,237]

In our study (Paper I), patients with 25(OH)D levels below 30 ng/ml and secondary hyperparathyroidism had lower BMD in terms of both T and Z scores of the hip and the lumbar spine compared with those with higher vitamin D levels, consistent with another study.[79]

The second study (Paper II) showed that women with psoriasis had higher BMD both of the hip and the lumbar spine compared with age-matched controls. Higher body weight and BMI are factors which may have contributed to the higher BMD in patients compared with controls. Information about the prevalence of osteoporosis among psoriasis patients and the epidemiology of risk factors for osteoporosis in this group are sparse. However, a previous study showed no evidence that patients with chronic plaque psoriasis had low BMD despite risk factors, although the subgroup with joint involvement appeared to be at higher risk of osteoporosis and therefore required prevention therapy.[132] Reduced BMD has been linked to palmoplantar pustular psoriasis.[133] The existence of less severe periarticular osteoporosis is considered possible, but there are no data concerning the prevalence of systemic osteoporosis in patients with psoriatic arthritis.[134] In general, bone loss increases with increasing age. BMD has been shown to be a predictive indicator for bone fractures in healthy subjects and in patients with osteoporosis.[238]

Vitamin D is important for bone metabolism.[135] Vitamin D deficiency thus contributes to the pathogenesis of osteoporosis and hip fractures.[136] Supplementation strategies involving calcium and vitamin D supplements are cost-saving for preventing osteoporotic fractures.[137]

Treatment with UVB in patients with psoriasis is most common during the darker period of the year when UVB is lacking and levels of vitamin D are low in Northern countries. Furthermore, UVB therapy heals psoriasis and supplies these patients with vitamin D at levels similar to those in the general population,[138] which might have positive effects also on bone status.

A family history of fractures, physical activity, smoking and oestrogen substitution are important factors influencing bone mass.[239-241] Low body weight is related to low skeletal muscle mass and increased risk of fractures.[241,242] Muscle tissue and strength are important for body balance and to prevent of falls.[243] Previous studies have confirmed a protective effect of weight gain against fractures.[237] Moreover, the relative risk of fractures (with the exception of spinal fractures) correlated positively with current height, height at the age of 25, and height loss since the age of 25.[244] The decrease in height since the age of 25 in patients with psoriasis with osteoporosis compared with patients with normal BMD was consistent with findings in other studies.[237]

A positive family history of hip fractures and especially kyphosis was more common in controls than in patients with psoriasis. That could be a possible explanation for the lower BMD of the lumbar spine in controls compared with patients with psoriasis. Heredity for hip fractures was mainly found in psoriatic patients with osteoporosis. A maternal history of hip fractures doubled the risk of having a hip fracture.[240,242] A history of fractures (particularly a family history of hip fractures) confers an increased risk of fractures that is independent of BMD.[245] Furthermore, the effect of family history is not general, but implies a site-specific predisposition to fractures.[246]

Menopause in patients with psoriasis occurred on average at a higher age in the group with normal BMD compared with the osteoporosis group. Early menopause is widely regarded as a risk factor for osteoporosis.[247] However, bone mass declines and the risk of fractures increases with increasing age, especially after the menopause along with diminishing endogenous secretion of oestradiol.[237,239,241]

Osteoporosis in postmenopausal women is more related to hormonal aberrations than to lifestyle factors.[237] Patients who used HRT had higher BMD.[237,248] The duration of HRT was longer in controls than in psoriasis patients. Within the psoriasis group, the duration of HRT was longer in patients with normal BMD. The duration of contraceptive usage was considerably longer in psoriasis patients with normal BMD than in the other two groups. Prior use of, and the duration use of oral contraceptive agents is associated with higher BMD.[249]

A history of postmenopausal symptoms in psoriasis patients did not influence BMD, consistent with a previous study.[250]

Physical activity correlated positively with BMD in psoriasis patients. Physical activity has been claimed to be beneficial to bone mass and protective against fractures.[251] Regular walking in middle-aged and elderly women is associated with a reduced risk of vertebral deformity.[252] Subjects who took a daily walk of at least 30 min had a significantly better climbing capacity, higher BMD and lower concentration of serum triglycerides than subjects who walked less.[253] Lifetime exercise was also positively associated with BMD of the hip.[254]

Current smoking was positively correlated with osteoporosis in our study, consistent with previous studies.[255,256] Smoking is widely considered to be a risk factor for future fracture.[257,258]

None of the patients had undergone systemic treatment with corticosteroids, but 77% had used topical treatment with corticosteroid ointments alone or in combination with vitamin D analogues (calcipotriol). The usage of topical corticosteroids was sparse, and the dosages were too low to have any systemic effects or impair on BMD. Prior and current exposure to corticosteroids confers an increased risk of fractures.[259]

Many of the patients, 63%, had joint pain (half of these patients had normal BMD) and reported limited mobility or function in at least one joint, but none of them had psoriasis arthritis. Higher body weight or BMI were positively associated with joint pain, consistent with a previous study.[260]

The limitation of this study (Paper II) was the small number of patients with psoriasis, and larger studies are needed before any firm conclusions regarding bone status and possible risk factors in psoriatic patients can be drawn.

Lipid status and blood glucose in psoriasis patients during treatment with heliotherapy (Paper IV)

Dyslipidemia

The ratio of low-density lipoprotein cholesterol (LDL) and high-density lipoprotein cholesterol (HDL) decreased, and the levels of haemoglobin A_1c (HbA_1c) also decreased in psoriasis patients during heliotherapy.

A strong link has been suggested between psoriasis and abnormalities in fatty acid metabolism.[118] Patients with psoriasis exhibit a dyslipidemic profile, including increased levels of plasma cholesterol, triglycerides (TG), LDL, very low-density lipoprotein (VLDL) cholesterol, and decreased levels of HDL cholesterol and antioxidant capacity.

Consistent with these findings, a potentially atherogenic lipid profile has also been observed in psoriasis patients compared with matched controls.[261] A recent study investigating the role of altered lipid metabolism in psoriasis suggests that a dyslipidemic profile may precede the onset of psoriasis.[262] Compared with healthy controls, patients with recent onset of psoriasis showed significantly elevated levels of HDL cholesterol and apolipoprotein (Apo) A-1 and had altered cholesterol/TG ratios in VLDL particles.[262] Adjustment for age, sex, smoking, physical exercise, alcohol use, BMI, and systolic blood pressure did not influence the results, leading to the conclusion that lipid abnormalities in psoriasis patients may be genetically determined.[262] Furthermore, HDL-associated Apo A-1 can inhibit the activation of monocytes/macrophages by interfering with their interaction with stimulated T-cells, leading to decreased TNF-α and IL-1 production.[263] Therefore, it is possible that lower levels of HDL may contribute to a state of chronic inflammation.[263] Moreover, VLDL and LDL have been shown to enhance the proliferation and LDL receptor expression of human epidermal keratinocytes in vitro, and increased VLDL or LDL could thus contribute to the pathogenesis of psoriasis.[264]

Metabolic syndrome

Large epidemiological studies have confirmed that psoriasis and psoriatic arthritis are associated with metabolic diseases including obesity, dyslipidemia and diabetes.[265] The metabolic syndrome comprises a cluster of risk factors including

central obesity, atherogenic dyslipidemia, hypertension and glucose intolerance, and is a strong predictor of diabetes, cardiovascular diseases, and stroke.[266,267] Patients with psoriasis also show an increased prevalence of the metabolic syndrome compared with nonpsoriatic dermatological patients.[268] The importance of the metabolic syndrome is that it may confer a higher cardiovascular risk than its individual components.[265]

Psoriasis patients with metabolic syndrome had disease onset at an earlier age, and longer disease duration compared with psoriasis patients without metabolic syndrome[268], suggesting that psoriasis duration is a risk factor for the metabolic syndrome. On the other hand, abdominal obesity is a proinflammatory state with the visceral adipose tissue providing a rich source of inflammatory mediators known as adipocytokines. These include adiponectin, leptin, resistin and visfatin, and may provide an important link between obesity, insulin resistance and related inflammatory disorders. Other products of adipose tissue that have been characterized include TNF-α and IL-6.[269] These products all play well-defined roles in the pathogenesis of psoriasis and at the interface between the immune and metabolic systems. In line with this hypothesis is the evidence that obesity is a risk factor for other inflammatory diseases such as atherosclerosis and asthma.

Hypertension

Diastolic blood pressure increased in patients with psoriasis following 15 days of climate therapy. Other studies have described that sun-like UV irradiation decreased resting pulse rate, decreased recovery pulse rate, and decreased systolic blood pressure in healthy persons.[270] We have no explanation for the present contradictory finding, but it could be due to chance or possibly changes in physical activity or alcohol or nutrient intake.

A higher prevalence of hypertension in psoriasis patients compared with controls has been reported.[271] The possibility exists that hypertension and psoriasis share common risk factors.[118] For example, angiotensin II, a product of angiotensin converting enzyme (ACE), is known to regulate vascular tone and to stimulate the release of proinflammatory cytokines.[272] Increased levels of serum ACE in psoriasis patients have been reported[273], as well as an increase in plasma renin activity. The prevalence of hypertension in psoriasis and the basis of this association require further study.

Insulin resistance/diabetes

An association between psoriasis and increased serum fasting glucose levels, hyperinsulinemia, insulin resistance, and type 2 diabetes has been demonstrated.[274] A cross-sectional study found that psoriasis patients were more likely to be insulin resistant and to have impaired glucose tolerance, higher fasting insulin levels, and impaired β-cell function than non-psoriatic subjects.[274] However, there were no correlations between these measures and psoriasis disease severity and duration.[274] Although the literature supports an association between psoriasis and insulin resistance/type 2 diabetes, the confounding factor of obesity should be taken into consideration. Obesity, especially abdominal obesity, is strongly associated with the development of the metabolic syndrome and type 2 diabetes.[275]

Neimann et al. demonstrated that obesity and diabetes were more strongly associated with severe psoriasis than were hypertension, hyperlipidemia, or smoking.[271] Therefore, lifestyle factors (e.g. lack of exercise, poor diet) could contribute to the development of obesity and accompanying insulin resistance in psoriasis patients.

Proinflammatory cytokines involved in psoriasis, such as TNF-α and IL-1β, play a central role in the development of atherosclerosis.[276] Atherosclerosis is considered to be an inflammatory disease and involves the production of proinflammatory cytokines by immune cells (macrophages, monocytes, T cells) at the sites of atherosclerotic lesions.[276]

Another link between psoriasis and atherosclerosis involves hyperhomocysteinemia.[277] Hyperhomocysteinemia is considered to be a risk factor for atherosclerotic vascular disease[278], and increased levels of homocysteine are implicated in atherosclerosis and vascular disease, in part through promotion of endothelial injury and thrombotic pathways.[279] In a case-control study homocysteine levels were found to be higher in patients with psoriasis than in healthy controls, and correlated with increasing psoriasis severity.[280, 281] Although folate deficiency may have a role in this condition, the exact mechanisms involved in hyperhomocysteinemia remain unclear.[280] Vitamin B12 (cyanocobalamin) can be photolysed by UV light.[282] UVB readily destroys tryptophan (Trp) residues of LDL and HDL. Moreover, LDL and HDL cholesterol are natural carriers of vitamin E and carotenoids. These two antioxidants are also rapidly bleached by UVB.[283] The

decrease in serum vitamin B12 during climate therapy can be explained by its photodegradation.

Role of vitamin D in the pathogenesis of type 2 diabetes mellitus

Increase in HDL-cholesterol and decrease in HbA_1c during climate therapy could be explained by several factors. One possible mechanism could be a direct effect of vitamin D on insulin sensitivity.[124] Another is that sun exposure usually implies greater outdoor physical activity, leading to beneficial effects on lipids and insulin sensitivity, unrelated to serum 25(OH)D concentrations.[124] The diet might also influence glucose and lipid metabolism. The observed associations between vitamin D, insulin, and glucose metabolism in humans have not yet been confirmed by intervention studies and, hence, a causal association has not been established.[124] Although climate therapy did not change the basal glucose levels of the patients, the HbA_1c levels decreased about 10 %, indicating improved insulin sensitivity.

There are seasonal variations in lipid levels, with levels of total cholesterol, LDL, triglyceride and lipoprotein A being highest in the winter when UVB induced production of vitamin D3 is at its minimum.[284] Serum concentrations of 25(OH)D at baseline correlated positively with serum HDL at baseline, consistent with a previously published study.[285] We found no other correlation between vitamin D and blood lipids.

Maintained body weight speaks against any caloric reduction as an explanation for the improved glucose and lipid metabolism.

Heliotherapy enhanced vitamin D3 production in patients with psoriasis. This might be one reason why positive effects were also seen on glucose and lipid metabolism.

Conclusions

- UVB therapy improved psoriasis scores, increased serum 25(OH)D synthesis and reduced serum PTH concentrations in postmenopausal women (paper I).

- Postmenopausal women with psoriasis had higher BMD than age-matched controls. Higher body weight, physical activity and UVB exposure could explain this finding (paper II).

- Serum concentrations of 25(OH)D in psoriasis patients increased less with narrowband UVB than with broadband UVB phototherapy. Psoriasis improved on both regimens (paper III).

- Climate therapy administered in Gran Canaria enhanced vitamin D3 production and improved PASI scores in patients with psoriasis. Positive effects were also seen on glucose and lipid metabolism (paper IV).

Future prospects

Extensive research has been carried out on the synthesis of vitamin D and the importance of this vitamin for human health in general. This thesis contributes to the research including vitamin D status in psoriasis patients during treatment with phototherapy.

The studies showed that the production of 25(OH)D did not correlate to the dose of UVB indicating a complexity in the regulation of the synthesis and metabolism of vitamin D.

It is not known whether skin affected by psoriasis or eczema differ in vitamin D production compared to normal skin. Our hypothesis is that different skin regions vary in their production of vitamin D.

More research is needed to develop safe intentional recommendations for sun exposure to obtain appropriate vitamin D production especially in the Scandinavian population. It is also necessary to establish new recommendations for daily vitamin D supplements in different patient groups.

Possible future research includes studies on:

- The capacity of psoriatic skin to produce and metabolise vitamin D during UVB exposure using the microdialysis technique.
- The ability of different skin areas to synthesize vitamin D for optimizing the recommendations for safe sun exposure.
- Vitamin D status in different population groups at increased risk of vitamin D deficiency including dark-skinned people and immigrants.
- To clarify the effect of climate therapy and life style factors such as physical activity and diet, on psoriasis, vitamin D, insulin sensitivity and lipid status.
- To clarify the relationship between psoriasis, type 2 diabetes and cardiovascular disease.

Acknowledgements

I wish to express my deep appreciation and sincere gratitude to:

Anne Lene Krogstad, my supervisor, for scientific guidance, generous support, stimulating supervision, fruitful discussions, and for your kindness, elegance, generosity and belief in me.

Kerstin Landin-Wilhelmsen, my assisting supervisor, my role model, for teaching me so much about scientific writing and proof-reading, for your diligence and your constructive help and support.

Olle Larkö, my co-supervisor, for your great interest in science, support and encouragement.

Ann-Marie Wennberg, my co-supervisor, for your friendly support, guidance and encouraging conversations.

I would like to thank my co-authors **Dan Mellström, Lena Hulthen, Lill Tove Nilsen, Elisabeth Søyland, Peter Abusdal Torjesen, Marit Nenseter and Tor Arne Hagve** for good and stimulating collaboration.

Martin Gillstedt, for statistical advice, and for taking the time and kindly helping me with all the graphs.

Valter Sundh, for statistical advice and assistance.

Morgan Carlsson, for skilful preparation of posters and help with the illustrations.

Lena Mattsson, for assistance with patients and friendly support over the years.

Inger Forsell, for diligent help with administrative procedures.

Agneta Fälted, for assistance with patients, literature explorations, and friendly support.

Staff at the outpatient clinic for psoriasis in Majorna for their help and support, taking care of both patients and samples.

Staff and colleagues at the Department of Dermatology Sahlgrenska University Hospital, Göteborg for their help and support.

Louise Hellner, **Lisbet Selvén** and **staff at the Department for Clinical Chemistry** for their help with all the analyses.

Angelica Jarlert and **staff at the Osteoporosis laboratory** for the performance of DEXA measurements.

The Section for Climate therapy, Dept of Rheumatology (Behandlingsreiser) for their support in carrying out the study on psoriasis patients during heliotherapy at Gran Canaria.

The staff at the Valle Marina Treatment Centre, at the laboratory of Immunology and at the Department of Dermatology, Dr Negrin University Hospital, Las Palmas for careful follow-up of the patients during the study, and assistance with collecting and storing the samples.

My family:

My parents **Magbula** and **Aziz Ahmic** for your endless love and unfailing support that always gave me confidence, peace and faith.

My husband **Edin** and our children **Amar, Adin** and **Selma** for all your patience, understanding, support and for your love.

My brother **Amer Ahmic** and his family for gracious support.

All my relatives and friends in **Bosnia and Herzegovina** and in **Sweden**.

This work was supported by grants from the federal government under the ALF agreement, the Göteborg Medical Society, the Regional Health Authority of West Sweden, the Edward Welander and Finsen's Foundation, the LUA Foundation at Sahlgrenska University Hospital and grants from the Section for Climate therapy, Dept of Rheumatology, Rikshospitalet University Hospital Oslo.

References

1 Bikle DD. Vitamin D: Production, metabolism, and mechanisms of action. In: *Diseases of Bone and Calcium Metabolism, Hyperparathyroidism*, Vol. Chapter 3: Endotex.com. 2004; 1-27.

2 Holick MF. The vitamin D epidemic and its health consequences. *J Nutr* 2005; 135: 2739S-48S.

3 Sniadecki J. Cited by W. Mozolowski: "Jedrzej Sniadecki (1768-1883) on the cure of rickets". *Nature* 1939; 143:121.

4 Owen I. Geografical distribution of rickets, acute and subacute rheumatism, chorea, cancer and urinary calculus in the British Islands. *Br Med J* 1889; 1: 113-6.

5 Palm T. The geographic distribution and etiology of rickets. *Practitioner* 1890; 45: 321-42.

6 Hess AF. On the Induction of Antirachitic Properties in Rations by Exposure to Light. *Science* 1924; 60: 269.

7 Steenbock H. The Induction of Growth Promoting and Calcifying Properties in a Ration by Exposure to Light. *Science* 1924; 60: 224-5.

8 Holick MF. The cutaneous photosynthesis of previtamin D3: a unique photoendocrine system. *J Invest Dermatol* 1981; 77: 51-8.

9 Holick MF, MacLaughlin JA, Doppelt SH. Regulation of cutaneous previtamin D3 photosynthesis in man: skin pigment is not an essential regulator. *Science* 1981; 211: 590-3.

10 Holick MF. Vitamin D: A millenium perspective. *J Cell Biochem* 2003; 88: 296-307.

11 Webb AR, DeCosta BR, Holick MF. Sunlight regulates the cutaneous production of vitamin D3 by causing its photodegradation. *J Clin Endocrinol Metab* 1989; 68: 882-7.

12 MacLaughlin J, Holick MF. Aging decreases the capacity of human skin to produce vitamin D3. *J Clin Invest* 1985; 76: 1536-8.

13 Need AG, Morris HA, Horowitz M *et al.* Effects of skin thickness, age, body fat, and sunlight on serum 25-hydroxyvitamin D. *Am J Clin Nutr* 1993; 58: 882-5.

14 Clemens TL, Adams JS, Henderson SL *et al.* Increased skin pigment reduces the capacity of skin to synthesise vitamin D3. *Lancet* 1982; 1: 74-6.

15 Matsuoka LY, Wortsman J, Hollis BW. Use of topical sunscreen for the evaluation of regional synthesis of vitamin D3. *J Am Acad Dermatol* 1990; 22: 772-5.

16 Matsuoka LY, Wortsman J, Dannenberg MJ *et al.* Clothing prevents ultraviolet-B radiation-dependent photosynthesis of vitamin D3. *J Clin Endocrinol Metab* 1992; 75: 1099-103.

17 Holick MF. McCollum Award Lecture, 1994: vitamin D--new horizons for the 21st century. *Am J Clin Nutr* 1994; 60: 619-30.

18 Webb AR, Kline L, Holick MF. Influence of season and latitude on the cutaneous synthesis of vitamin D3: exposure to winter sunlight in Boston and Edmonton will not promote vitamin D3 synthesis in human skin. *J Clin Endocrinol Metab* 1988; 67: 373-8.

19 Wortsman J, Matsuoka LY, Chen TC *et al.* Decreased bioavailability of vitamin D in obesity. *Am J Clin Nutr* 2000; 72: 690-3.

20 Verboven C, Rabijns A, De Maeyer M *et al.* A structural basis for the unique binding features of the human vitamin D-binding protein. *Nat Struct Biol* 2002; 9: 131-6.

21 Bar M, Domaschke D, Meye A *et al.* Wavelength-dependent induction of CYP24A1-mRNA after UVB-triggered calcitriol synthesis in cultured human keratinocytes. *J Invest Dermatol* 2007; 127: 206-13.

22 Segaert S, Garmyn M, Degreef H *et al.* Retinoic acid modulates the anti-proliferative effect of 1,25-dihydroxyvitamin D3 in cultured human epidermal keratinocytes. *J Invest Dermatol* 1997; 109: 46-54.

23 Vantieghem K, Kissmeyer AM, De Haes P *et al.* UVB-induced production of 1,25-dihydroxyvitamin D3 and vitamin D activity in human keratinocytes pretreated with a sterol Delta7-reductase inhibitor. *J Cell Biochem* 2006; 98: 81-92.

24 Vantieghem K, De Haes P, Bouillon R *et al.* Dermal fibroblasts pretreated with a sterol Delta7-reductase inhibitor produce 25-hydroxyvitamin D3 upon UVB irradiation. *J Photochem Photobiol B* 2006; 85: 72-8.

25 Jones G, Strugnell SA, DeLuca HF. Current understanding of the molecular actions of vitamin D. *Physiol Rev* 1998; 78: 1193-231.

26 Lehmann B, Querings K, Reichrath J. Vitamin D and skin: new aspects for dermatology. *Exp Dermatol* 2004; 13 Suppl 4: 11-5.

27 De Haes P, Garmyn M, Degreef H *et al.* 1,25-Dihydroxyvitamin D3 inhibits ultraviolet B-induced apoptosis, Jun kinase activation, and interleukin-6 production in primary human keratinocytes. *J Cell Biochem* 2003; 89: 663-73.

28 De Haes P, Garmyn M, Verstuyf A *et al.* 1,25-Dihydroxyvitamin D3 and analogues protect primary human keratinocytes against UVB-induced DNA damage. *J Photochem Photobiol B* 2005; 78: 141-8.

29 Chapuy MC, Preziosi P, Maamer M *et al.* Prevalence of vitamin D insufficiency in an adult normal population. *Osteoporos Int* 1997; 7: 439-43.

30 Lips P. Vitamin D deficiency and secondary hyperparathyroidism in the elderly: consequences for bone loss and fractures and therapeutic implications. *Endocr Rev* 2001; 22: 477-501.

31 Malabanan A, Veronikis IE, Holick MF. Redefining vitamin D insufficiency. *Lancet* 1998; 351: 805-6.

32 Dawson-Hughes B, Heaney RP, Holick MF *et al.* Estimates of optimal vitamin D status. *Osteoporos Int* 2005; 16: 713-6.

33 Grant WB, Holick MF. Benefits and requirements of vitamin D for optimal health: a review. *Altern Med Rev* 2005; 10: 94-111.

34 Zittermann A. Vitamin D in preventive medicine: are we ignoring the evidence? *Br J Nutr* 2003; 89: 552-72.

35 Favus MJ. Postmenopausal osteoporosis and the detection of so-called secondary causes of low bone density. *J Clin Endocrinol Metab* 2005; 90: 3800-1.

36 Van Cromphaut SJ, Dewerchin M, Hoenderop JG *et al.* Duodenal calcium absorption in vitamin D receptor-knockout mice: functional and molecular aspects. *Proc Natl Acad Sci U S A* 2001; 98: 13324-9.

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 Huang JC, Sakata T, Pfleger LL *et al.* PTH differentially regulates expression of RANKL and OPG. *J Bone Miner Res* 2004; 19: 235-44.

39 Holick MF. Sunlight and vitamin D: both good for cardiovascular health. *J Gen Intern Med* 2002; 17: 733-5.

40 Stumpf WE, Sar M, Reid FA *et al.* Target cells for 1,25-dihydroxyvitamin D3 in intestinal tract, stomach, kidney, skin, pituitary, and parathyroid. *Science* 1979; 206: 1188-90.

41 Adorini L. 1,25-Dihydroxyvitamin D3 analogs as potential therapies in transplantation. *Curr Opin Investig Drugs* 2002; 3: 1458-63.

42 Garland C, Shekelle RB, Barrett-Connor E *et al.* Dietary vitamin D and calcium and risk of colorectal cancer: a 19-year prospective study in men. *Lancet* 1985; 1: 307-9.

43 Garland CF, Garland FC. Do sunlight and vitamin D reduce the likelihood of colon cancer? *Int J Epidemiol* 1980; 9: 227-31.

44 Grant WB. An estimate of premature cancer mortality in the U.S. due to inadequate doses of solar ultraviolet-B radiation. *Cancer* 2002; 94: 1867-75.

45 Ahonen MH, Tenkanen L, Teppo L *et al.* Prostate cancer risk and prediagnostic serum 25-hydroxyvitamin D levels (Finland). *Cancer Causes Control* 2000; 11: 847-52.

46 Bikle DD, Nemanic MK, Gee E *et al.* 1,25-Dihydroxyvitamin D3 production by human keratinocytes. Kinetics and regulation. *J Clin Invest* 1986; 78: 557-66.

47 Adams JS, Gacad MA, Anders A *et al.* Biochemical indicators of disordered vitamin D and calcium homeostasis in sarcoidosis. *Sarcoidosis* 1986; 3: 1-6.

48 Schwartz GG, Whitlatch LW, Chen TC *et al.* Human prostate cells synthesize 1,25-dihydroxyvitamin D3 from 25-hydroxyvitamin D3. *Cancer Epidemiol Biomarkers Prev* 1998; 7: 391-5.

49 Tangpricha V, Flanagan JN, Whitlatch LW *et al.* 25-hydroxyvitamin D-1alpha-hydroxylase in normal and malignant colon tissue. *Lancet* 2001; 357: 1673-4.

50 Mawer EB, Hayes ME, Heys SE *et al.* Constitutive synthesis of 1,25-dihydroxyvitamin D3 by a human small cell lung cancer cell line. *J Clin Endocrinol Metab* 1994; 79: 554-60.

51 Cross HS, Bareis P, Hofer H *et al.* 25-Hydroxyvitamin D(3)-1alpha-hydroxylase and vitamin D receptor gene expression in human colonic mucosa is elevated during early cancerogenesis. *Steroids* 2001; 66: 287-92.

52 Maas RM, Reus K, Diesel B *et al.* Amplification and expression of splice variants of the gene encoding the P450 cytochrome 25-hydroxyvitamin D(3) 1,alpha-hydroxylase (CYP 27B1) in human malignant glioma. *Clin Cancer Res* 2001; 7: 868-75.

53 Tsoukas CD, Provvedini DM, Manolagas SC. 1,25-dihydroxyvitamin D3: a novel immunoregulatory hormone. *Science* 1984; 224: 1438-40.

54 Garland FC, Garland CF, Gorham ED *et al.* Geographic variation in breast cancer mortality in the United States: a hypothesis involving exposure to solar radiation. *Prev Med* 1990; 19: 614-22.

55 Berwick M, Armstrong BK, Ben-Porat L *et al.* Sun exposure and mortality from melanoma. *J Natl Cancer Inst* 2005; 97: 195-9.

56 Kennedy C, Bajdik CD, Willemze R *et al.* The influence of painful sunburns and lifetime sun exposure on the risk of actinic keratoses, seborrheic warts,

melanocytic nevi, atypical nevi, and skin cancer. *J Invest Dermatol* 2003; 120: 1087-93.

57 Bhalla AK, Amento EP, Clemens TL *et al.* Specific high-affinity receptors for 1,25-dihydroxyvitamin D3 in human peripheral blood mononuclear cells: presence in monocytes and induction in T lymphocytes following activation. *J Clin Endocrinol Metab* 1983; 57: 1308-10.

58 Mathieu C, Adorini L. The coming of age of 1,25-dihydroxyvitamin D(3) analogs as immunomodulatory agents. *Trends Mol Med* 2002; 8: 174-9.

59 Deluca HF, Cantorna MT. Vitamin D: its role and uses in immunology. *FASEB J* 2001; 15: 2579-85.

60 Mathieu C, Waer M, Laureys J *et al.* Prevention of autoimmune diabetes in NOD mice by 1,25 dihydroxyvitamin D3. *Diabetologia* 1994; 37: 552-8.

61 Cantorna MT, Hayes CE, DeLuca HF. 1,25-Dihydroxyvitamin D3 reversibly blocks the progression of relapsing encephalomyelitis, a model of multiple sclerosis. *Proc Natl Acad Sci U S A* 1996; 93: 7861-4.

62 Cantorna MT, Munsick C, Bemiss C *et al.* 1,25-Dihydroxycholecalciferol prevents and ameliorates symptoms of experimental murine inflammatory bowel disease. *J Nutr* 2000; 130: 2648-52.

63 Embry AF, Snowdon LR, Vieth R. Vitamin D and seasonal fluctuations of gadolinium-enhancing magnetic resonance imaging lesions in multiple sclerosis. *Ann Neurol* 2000; 48: 271-2.

64 Merlino LA, Curtis J, Mikuls TR *et al.* Vitamin D intake is inversely associated with rheumatoid arthritis: results from the Iowa Women's Health Study. *Arthritis Rheum* 2004; 50: 72-7.

65 Hypponen E, Laara E, Reunanen A *et al.* Intake of vitamin D and risk of type 1 diabetes: a birth-cohort study. *Lancet* 2001; 358: 1500-3.

66 Rostand SG. Ultraviolet light may contribute to geographic and racial blood pressure differences. *Hypertension* 1997; 30: 150-6.

67 Krause R, Buhring M, Hopfenmuller W *et al.* Ultraviolet B and blood pressure. *Lancet* 1998; 352: 709-10.

68 Zittermann A, Schleithoff SS, Tenderich G *et al.* Low vitamin D status: a contributing factor in the pathogenesis of congestive heart failure? *J Am Coll Cardiol* 2003; 41: 105-12.

69 Li YC, Kong J, Wei M *et al.* 1,25-Dihydroxyvitamin D(3) is a negative endocrine regulator of the renin-angiotensin system. *J Clin Invest* 2002; 110: 229-38.

70 Weishaar RE, Simpson RU. Involvement of vitamin D3 with cardiovascular function. II. Direct and indirect effects. *Am J Physiol* 1987; 253: E675-83.

71 O'Connell TD, Weishaar RE, Simpson RU. Regulation of myosin isozyme expression by vitamin D3 deficiency and 1,25-dihydroxyvitamin D3 in the rat heart. *Endocrinology* 1994; 134: 899-905.

72 Zasloff M. Sunlight, vitamin D, and the innate immune defenses of the human skin. *J Invest Dermatol* 2005; 125: xvi-xvii.

73 Wang TT, Nestel FP, Bourdeau V *et al.* Cutting edge: 1,25-dihydroxyvitamin D3 is a direct inducer of antimicrobial peptide gene expression. *J Immunol* 2004; 173: 2909-12.

74 Weber G, Heilborn JD, Chamorro Jimenez CI *et al.* Vitamin D induces the antimicrobial protein hCAP18 in human skin. *J Invest Dermatol* 2005; 124: 1080-2.

75 Segaert S, Simonart T. The epidermal vitamin D system and innate immunity: some more light shed on this unique photoendocrine system? *Dermatology* 2008; 217: 7-11.

76 Schauber J, Dorschner RA, Coda AB *et al.* Injury enhances TLR2 function and antimicrobial peptide expression through a vitamin D-dependent mechanism. *J Clin Invest* 2007; 117: 803-11.

77 Norman AW. Problems relating to the definition of an international unit for vitamin D and its metabolites. *J Nutr* 1972; 102: 1243-6.

78 Barger-Lux MJ, Heaney RP, Dowell S *et al.* Vitamin D and its major metabolites: serum levels after graded oral dosing in healthy men. *Osteoporos Int* 1998; 8: 222-30.

79 Tangpricha V, Turner A, Spina C *et al.* Tanning is associated with optimal vitamin D status (serum 25-hydroxyvitamin D concentration) and higher bone mineral density. *Am J Clin Nutr* 2004; 80: 1645-9.

80 Bischoff-Ferrari HA, Dietrich T, Orav EJ *et al.* 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.

81 Sedrani SH. Vitamin D status of Saudi men. *Trop Geogr Med* 1984; 36: 181-7.

82 Sedrani SH, Elidrissy AW, El Arabi KM. Sunlight and vitamin D status in normal Saudi subjects. *Am J Clin Nutr* 1983; 38: 129-32.

83 Gladman DD. Psoriatic arthritis. *Dermatol Ther* 2004; 17: 350-63.

84 Bos JD, de Rie MA, Teunissen MB *et al.* Psoriasis: dysregulation of innate immunity. *Br J Dermatol* 2005; 152: 1098-107.

85 Bowcock AM, Krueger JG. Getting under the skin: the immunogenetics of psoriasis. *Nat Rev Immunol* 2005; 5: 699-711.

86 Wang H, Peters T, Kess D *et al.* Activated macrophages are essential in a murine model for T cell-mediated chronic psoriasiform skin inflammation. *J Clin Invest* 2006; 116: 2105-14.

87 Stratis A, Pasparakis M, Rupec RA *et al.* Pathogenic role for skin macrophages in a mouse model of keratinocyte-induced psoriasis-like skin inflammation. *J Clin Invest* 2006; 116: 2094-104.

88 Clark RA, Kupper TS. Misbehaving macrophages in the pathogenesis of psoriasis. *J Clin Invest* 2006; 116: 2084-7.

89 Austin LM, Ozawa M, Kikuchi T *et al.* The majority of epidermal T cells in Psoriasis vulgaris lesions can produce type 1 cytokines, interferon-gamma, interleukin-2, and tumor necrosis factor-alpha, defining TC1 (cytotoxic T lymphocyte) and TH1 effector populations: a type 1 differentiation bias is also measured in circulating blood T cells in psoriatic patients. *J Invest Dermatol* 1999; 113: 752-9.

90 Ozawa M, Aiba S. Immunopathogenesis of psoriasis. *Curr Drug Targets Inflamm Allergy* 2004; 3: 137-44.

91 Fierlbeck G, Rassner G, Muller C. Psoriasis induced at the injection site of recombinant interferon gamma. Results of immunohistologic investigations. *Arch Dermatol* 1990; 126: 351-5.

92 Ettehadi P, Greaves MW, Wallach D *et al.* Elevated tumour necrosis factor-alpha (TNF-alpha) biological activity in psoriatic skin lesions. *Clin Exp Immunol* 1994; 96: 146-51.

93 Bonifati C, Carducci M, Cordiali Fei P *et al.* Correlated increases of tumour necrosis factor-alpha, interleukin-6 and granulocyte monocyte-colony stimulating factor levels in suction blister fluids and sera of psoriatic patients--relationships with disease severity. *Clin Exp Dermatol* 1994; 19: 383-7.

94 Jacob SE, Nassiri M, Kerdel FA *et al.* Simultaneous measurement of multiple Th1 and Th2 serum cytokines in psoriasis and correlation with disease severity. *Mediators Inflamm* 2003; 12: 309-13.

95 Kagen MH, McCormick TS, Cooper KD. Regulatory T cells in psoriasis. *Ernst Schering Res Found Workshop* 2006: 193-209.

96 Bata-Csorgo Z, Hammerberg C, Voorhees JJ *et al.* Kinetics and regulation of human keratinocyte stem cell growth in short-term primary ex vivo culture. Cooperative growth factors from psoriatic lesional T lymphocytes stimulate

proliferation among psoriatic uninvolved, but not normal, stem keratinocytes. *J Clin Invest* 1995; 95: 317-27.

97 van der Vleuten CJ, Gerritsen MJ, Steijlen PM *et al.* A therapeutic approach to erythrodermic psoriasis: report of a case and a discussion of therapeutic options. *Acta Derm Venereol* 1996; 76: 65-7.

98 Bourke JF, Iqbal SJ, Hutchinson PE. The effects of UVB plus calcipotriol on systemic calcium homeostasis in patients with chronic plaque psoriasis. *Clin Exp Dermatol* 1997; 22: 259-61.

99 Fogh K, Kragballe K. Recent developments in vitamin D analogs. *Curr Pharm Des* 2000; 6: 961-72.

100 Langner A, Ashton P, Van De Kerkhof PC *et al.* A long-term multicentre assessment of the safety and tolerability of calcitriol ointment in the treatment of chronic plaque psoriasis. *Br J Dermatol* 1996; 135: 385-9.

101 van de Kerkhof PC, Berth-Jones J, Griffiths CE *et al.* Long-term efficacy and safety of tacalcitol ointment in patients with chronic plaque psoriasis. *Br J Dermatol* 2002; 146: 414-22.

102 Kragballe K. Vitamin D and UVB radiation therapy. *Cutis* 2002; 70: 9-12.

103 Feldman D, Chen T, Hirst M *et al.* Demonstration of 1,25-dihydroxyvitamin D3 receptors in human skin biopsies. *J Clin Endocrinol Metab* 1980; 51: 1463-5.

104 Provvedini DM, Tsoukas CD, Deftos LJ *et al.* 1,25-dihydroxyvitamin D3 receptors in human leukocytes. *Science* 1983; 221: 1181-3.

105 Morimoto S, Kumahara Y. A patient with psoriasis cured by 1 alpha-hydroxyvitamin D3. *Med J Osaka Univ* 1985; 35: 51-4.

106 Nagpal S, Lu J, Boehm MF. Vitamin D analogs: mechanism of action and therapeutic applications. *Curr Med Chem* 2001; 8: 1661-79.

107 Takahashi H, Ibe M, Kinouchi M *et al.* Similarly potent action of 1,25-dihydroxyvitamin D3 and its analogues, tacalcitol, calcipotriol, and maxacalcitol on normal human keratinocyte proliferation and differentiation. *J Dermatol Sci* 2003; 31: 21-8.

108 Pillai S, Bikle DD. Role of intracellular-free calcium in the cornified envelope formation of keratinocytes: differences in the mode of action of extracellular calcium and 1,25 dihydroxyvitamin D3. *J Cell Physiol* 1991; 146: 94-100.

109 Ratnam AV, Bikle DD, Cho JK. 1,25 dihydroxyvitamin D3 enhances the calcium response of keratinocytes. *J Cell Physiol* 1999; 178: 188-96.

110 Manolagas SC, Provvedini DM, Tsoukas CD. Interactions of 1,25-dihydroxyvitamin D3 and the immune system. *Mol Cell Endocrinol* 1985; 43: 113-22.

111 Muller K, Bendtzen K. 1,25-Dihydroxyvitamin D3 as a natural regulator of human immune functions. *J Investig Dermatol Symp Proc* 1996; 1: 68-71.

112 Pinette KV, Yee YK, Amegadzie BY *et al.* Vitamin D receptor as a drug discovery target. *Mini Rev Med Chem* 2003; 3: 193-204.

113 Tobler A, Gasson J, Reichel H *et al.* Granulocyte-macrophage colony-stimulating factor. Sensitive and receptor-mediated regulation by 1,25-dihydroxyvitamin D3 in normal human peripheral blood lymphocytes. *J Clin Invest* 1987; 79: 1700-5.

114 Kang S, Yi S, Griffiths CE *et al.* Calcipotriene-induced improvement in psoriasis is associated with reduced interleukin-8 and increased interleukin-10 levels within lesions. *Br J Dermatol* 1998; 138: 77-83.

115 Michel G, Gailis A, Jarzebska-Deussen B *et al.* 1,25-(OH)2-vitamin D3 and calcipotriol induce IL-10 receptor gene expression in human epidermal cells. *Inflamm Res* 1997; 46: 32-4.

116 Penna G, Adorini L. 1 Alpha,25-dihydroxyvitamin D3 inhibits differentiation, maturation, activation, and survival of dendritic cells leading to impaired alloreactive T cell activation. *J Immunol* 2000; 164: 2405-11.

117 Adorini L, Penna G, Giarratana N *et al.* Tolerogenic dendritic cells induced by vitamin D receptor ligands enhance regulatory T cells inhibiting allograft rejection and autoimmune diseases. *J Cell Biochem* 2003; 88: 227-33.

118 Gottlieb AB, Chao C, Dann F. Psoriasis comorbidities. *J Dermatolog Treat* 2008; 19: 5-21.

119 Christophers E. Comorbidities in psoriasis. *Clin Dermatol* 2007; 25: 529-34.

120 Gottlieb AB, Dann F, Menter A. Psoriasis and the metabolic syndrome. *J Drugs Dermatol* 2008; 7: 563-72.

121 Sommer DM, Jenisch S, Suchan M *et al.* Increased prevalence of the metabolic syndrome in patients with moderate to severe psoriasis. *Arch Dermatol Res* 2006; 298: 321-8.

122 Akhyani M, Ehsani AH, Robati RM *et al.* The lipid profile in psoriasis: a controlled study. *J Eur Acad Dermatol Venereol* 2007; 21: 1330-2.

123 Palomer X, Gonzalez-Clemente JM, Blanco-Vaca F *et al.* Role of vitamin D in the pathogenesis of type 2 diabetes mellitus. *Diabetes Obes Metab* 2008; 10: 185-97.

124 Tai K, Need AG, Horowitz M *et al.* Vitamin D, glucose, insulin, and insulin sensitivity. *Nutrition* 2008; 24: 279-85.

125 Procopio M, Magro G, Cesario F *et al.* The oral glucose tolerance test reveals a high frequency of both impaired glucose tolerance and undiagnosed Type 2 diabetes mellitus in primary hyperparathyroidism. *Diabet Med* 2002; 19: 958-61.

126 Taylor WH, Khaleeli AA. Prevalence of primary hyperparathyroidism in patients with diabetes mellitus. *Diabet Med* 1997; 14: 386-9.

127 Luong K, Nguyen LT, Nguyen DN. The role of vitamin D in protecting type 1 diabetes mellitus. *Diabetes Metab Res Rev* 2005; 21: 338-46.

128 Mauricio D, Mandrup-Poulsen T, Nerup J. Vitamin D analogues in insulin-dependent diabetes mellitus and other autoimmune diseases: a therapeutic perspective. *Diabetes Metab Rev* 1996; 12: 57-68.

129 Hewison M, Gacad MA, Lemire J *et al.* Vitamin D as a cytokine and hematopoetic factor. *Rev Endocr Metab Disord* 2001; 2: 217-27.

130 Norman PE, Powell JT. Vitamin D, shedding light on the development of disease in peripheral arteries. *Arterioscler Thromb Vasc Biol* 2005; 25: 39-46.

131 Lemire JM. Immunomodulatory actions of 1,25-dihydroxyvitamin D3. *J Steroid Biochem Mol Biol* 1995; 53: 599-602.

132 Millard TP, Antoniades L, Evans AV *et al.* Bone mineral density of patients with chronic plaque psoriasis. *Clin Exp Dermatol* 2001; 26: 446-8.

133 Nymann P, Kollerup G, Jemec GB *et al.* Decreased bone mineral density in patients with pustulosis palmaris et plantaris. *Dermatology* 1996; 192: 307-11.

134 Frediani B, Allegri A, Falsetti P *et al.* Bone mineral density in patients with psoriatic arthritis. *J Rheumatol* 2001; 28: 138-43.

135 Chapuy MC, Arlot ME, Duboeuf F *et al.* Vitamin D3 and calcium to prevent hip fractures in the elderly women. *N Engl J Med* 1992; 327: 1637-42.

136 Chel VG, Ooms ME, Popp-Snijders C *et al.* Ultraviolet irradiation corrects vitamin D deficiency and suppresses secondary hyperparathyroidism in the elderly. *J Bone Miner Res* 1998; 13: 1238-42.

137 Lilliu H, Pamphile R, Chapuy MC *et al.* Calcium-vitamin D3 supplementation is cost-effective in hip fractures prevention. *Maturitas* 2003; 44: 299-305.

138 Landin-Wilhelmsen K, Wilhelmsen L, Wilske J *et al.* Sunlight increases serum 25(OH) vitamin D concentration whereas 1,25(OH)2D3 is unaffected. Results from a general population study in Goteborg, Sweden (The WHO MONICA Project). *Eur J Clin Nutr* 1995; 49: 400-7.

139 Wassberg C, Backvall H, Diffey B *et al.* Enhanced epidermal ultraviolet responses in chronically sun-exposed skin are dependent on previous sun exposure. *Acta Derm Venereol* 2003; 83: 254-61.

140 Chadwick CA, Potten CS, Nikaido O *et al.* The detection of cyclobutane thymine dimers, (6-4) photolesions and the Dewar photoisomers in sections of UV-irradiated human skin using specific antibodies, and the demonstration of depth penetration effects. *J Photochem Photobiol B* 1995; 28: 163-70.

141 Campbell C, Quinn AG, Angus B *et al.* Wavelength specific patterns of p53 induction in human skin following exposure to UV radiation. *Cancer Res* 1993; 53: 2697-9.

142 Hoffmann K, Kaspar K, Altmeyer P *et al.* UV transmission measurements of small skin specimens with special quartz cuvettes. *Dermatology* 2000; 201: 307-11.

143 Farr PM, Diffey BL, Steele MC. A preliminary study on the in vivo transmission of light through psoriatic plaques. *Photodermatol* 1984; 1: 87-90.

144 Krutmann J, Morita, A. and Elmets, A. Mechanisms of photo(chemo)therapy. *Springer pp. 56-59* 2001.

145 Trautinger F. Mechanisms of photodamage of the skin and its functional consequences for skin ageing. *Clin Exp Dermatol* 2001; 26: 573-7.

146 Clingen PH, Arlett CF, Cole J *et al.* Correlation of UVC and UVB cytotoxicity with the induction of specific photoproducts in T-lymphocytes and fibroblasts from normal human donors. *Photochem Photobiol* 1995; 61: 163-70.

147 Dalziel KL. Aspects of cutaneous ageing. *Clin Exp Dermatol* 1991; 16: 315-23.

148 Diffey BL, Farr PM. An appraisal of ultraviolet lamps used for the phototherapy of psoriasis. *Br J Dermatol* 1987; 117: 49-56.

149 Parrish JA. Ultraviolet phototherapy of psoriasis. *Pharmacol Ther* 1981; 15: 313-20.

150 Goeckerman WH. The treatment of psoriasis. *Northwest Med* (1925); 24 pp. 229-31.

151 Honigsmann H. Phototherapy for psoriasis. *Clin Exp Dermatol* 2001; 26: 343-50.

152 Ingram JT. The approach to psoriasis. *Br Med J* 1953; 2: 591-4.

153 Parrish JA, Fitzpatrick TB, Tanenbaum L *et al.* Photochemotherapy of psoriasis with oral methoxsalen and longwave ultraviolet light. *N Engl J Med* 1974; 291: 1207-11.

154 Wolff K, Honigsmann H, Gschnait F *et al.* [Photochemotherapy of psoriasis: clinical experiences with 152 patients (author's transl)]. *Dtsch Med Wochenschr* 1975; 100: 2471-7, 1497.

155 Feldman SR, Garton R, Averett W *et al.* Strategy to manage the treatment of severe psoriasis: considerations of efficacy, safety and cost. *Expert Opin Pharmacother* 2003; 4: 1525-33.

156 Fischer T, Alsins J. Treatment of psoriasis with trioxsalen baths and dysprosium lamps. *Acta Derm Venereol* 1976; 56: 383-90.

157 Parrish JA, Jaenicke KF. Action spectrum for phototherapy of psoriasis. *J Invest Dermatol* 1981; 76: 359-62.

158 Green C, Ferguson J, Lakshmipathi T *et al.* 311 nm UVB phototherapy--an effective treatment for psoriasis. *Br J Dermatol* 1988; 119: 691-6.

159 Karvonen J, Kokkonen EL, Ruotsalainen E. 311 nm UVB lamps in the treatment of psoriasis with the Ingram regimen. *Acta Derm Venereol* 1989; 69: 82-5.

160 van Weelden H, De La Faille HB, Young E *et al.* A new development in UVB phototherapy of psoriasis. *Br J Dermatol* 1988; 119: 11-9.

161 Berneburg M, Rocken M, Benedix F. Phototherapy with narrowband vs broadband UVB. *Acta Derm Venereol* 2005; 85: 98-108.

162 Coven TR, Burack LH, Gilleaudeau R *et al.* Narrowband UV-B produces superior clinical and histopathological resolution of moderate-to-severe psoriasis in patients compared with broadband UV-B. *Arch Dermatol* 1997; 133: 1514-22.

163 Dawe RS, Cameron H, Yule S *et al.* A randomized controlled trial of narrowband ultraviolet B vs bath-psoralen plus ultraviolet A photochemotherapy for psoriasis. *Br J Dermatol* 2003; 148: 1194-204.

164 Pasic A, Ceovic R, Lipozencic J *et al.* Phototherapy in pediatric patients. *Pediatr Dermatol* 2003; 20: 71-7.

165 Walters IB, Burack LH, Coven TR *et al.* Suberythemogenic narrow-band UVB is markedly more effective than conventional UVB in treatment of psoriasis vulgaris. *J Am Acad Dermatol* 1999; 40: 893-900.

166 Diffey BL. Towards optimal regimens for the UVB phototherapy of psoriasis: a mathematical model. *Acta Derm Venereol* 2004; 84: 259-64.

167 Dawe RS, Wainwright NJ, Cameron H *et al.* Narrow-band (TL-01) ultraviolet B phototherapy for chronic plaque psoriasis: three times or five times weekly treatment? *Br J Dermatol* 1998; 138: 833-9.

168 Cameron H, Dawe RS, Yule S *et al.* A randomized, observer-blinded trial of twice vs. three times weekly narrowband ultraviolet B phototherapy for chronic plaque psoriasis. *Br J Dermatol* 2002; 147: 973-8.

169 Wainwright NJ, Dawe RS, Ferguson J. Narrowband ultraviolet B (TL-01) phototherapy for psoriasis: which incremental regimen? *Br J Dermatol* 1998; 139: 410-4.

170 Zanolli M. Phototherapy treatment of psoriasis today. *J Am Acad Dermatol* 2003; 49: S78-86.

171 Duthie MS, Kimber I, Norval M. The effects of ultraviolet radiation on the human immune system. *Br J Dermatol* 1999; 140: 995-1009.

172 Sethi G, Sodhi A. Role of p38 mitogen-activated protein kinase and caspases in UV-B-induced apoptosis of murine peritoneal macrophages. *Photochem Photobiol* 2004; 79: 48-54.

173 Krueger JG, Wolfe JT, Nabeya RT *et al.* Successful ultraviolet B treatment of psoriasis is accompanied by a reversal of keratinocyte pathology and by selective depletion of intraepidermal T cells. *J Exp Med* 1995; 182: 2057-68.

174 Ozawa M, Ferenczi K, Kikuchi T *et al.* 312-nanometer ultraviolet B light (narrow-band UVB) induces apoptosis of T cells within psoriatic lesions. *J Exp Med* 1999; 189: 711-8.

175 Okamoto H, Horio T, Maeda M. Alteration of lymphocyte functions by 8-methoxypsoralen and long-wave ultraviolet radiation. II. The effect of in vivo PUVA on IL-2 production. *J Invest Dermatol* 1987; 89: 24-6.

176 Kripke ML, Morison WL, Parrish JA. Systemic suppression of contact hypersensitivity in mice by psoralen plus UVA radiation (PUVA). *J Invest Dermatol* 1983; 81: 87-92.

177 Aubin F, Mousson C. Ultraviolet light-induced regulatory (suppressor) T cells: an approach for promoting induction of operational allograft tolerance? *Transplantation* 2004; 77: S29-31.

178 Weitzen ML, Bonavida B. Mechanism of inhibition of human natural killer activity by ultraviolet radiation. *J Immunol* 1984; 133: 3128-32.

179 Kasahara S, Aizawa K, Okamiya M *et al.* UVB irradiation suppresses cytokine production and innate cellular immune functions in mice. *Cytokine* 2001; 14: 104-11.

180 Schneider LA, Hinrichs R, Scharffetter-Kochanek K. Phototherapy and photochemotherapy. *Clin Dermatol* 2008; 26: 464-76.

181 Schneider LA, Bloch W, Kopp K *et al.* 8-Isoprostane is a dose-related biomarker for photo-oxidative ultraviolet (UV) B damage in vivo: a pilot study with personal UV dosimetry. *Br J Dermatol* 2006; 154: 1147-54.

182 Potten CS, Chadwick CA, Cohen AJ *et al.* DNA damage in UV-irradiated human skin in vivo: automated direct measurement by image analysis (thymine dimers) compared with indirect measurement (unscheduled DNA synthesis) and protection by 5-methoxypsoralen. *Int J Radiat Biol* 1993; 63: 313-24.

183 Greinert R, Boguhn O, Harder D *et al.* The dose dependence of cyclobutane dimer induction and repair in UVB-irradiated human keratinocytes. *Photochem Photobiol* 2000; 72: 701-8.

184 Brenneisen P, Blaudschun R, Gille J *et al.* Essential role of an activator protein-2 (AP-2)/specificity protein 1 (Sp1) cluster in the UVB-mediated induction of the human vascular endothelial growth factor in HaCaT keratinocytes. *Biochem J* 2003; 369: 341-9.

185 Snellman E, Lauharanta J, Reunanen A *et al.* Effect of heliotherapy on skin and joint symptoms in psoriasis: a 6-month follow-up study. *Br J Dermatol* 1993; 128: 172-7.

186 Colipa. International Sun Protection Factor Test Metho. In: *The European Cosmetic Toiletry and Perfumery Association*, Vol. 12. 2006.

187 McKinlay AFaD, B. L. A Reference Action Spectrum for Ultraviolet Induced Erythema in Human Skin. *CIE-J* 1987; *6,*: *17-22*.

188 Leszczynski K JK, Ylianttila L, Visuri R, Blumthaler M. Report of the WMO/STUK intercomparison of erythemally-weighted solar UV radiometers. In: Vol. Spring/Summer. Geneva:: World Meteorological Organization (WMO). 1995.

189 Seckmeyer G BA, Bernard G, Blumthaler M, Booth C R., Lantz K, McKenzie R L. Instruments to Measure Solar Ultraviolet Radiation. Part 2: Broadband Instruments Measuring Erythemally Weighted Solar Irradiance. . In, Vol. WMO/GAW Report No. 164. Geneva: Geneva: World Meteorological Organization (WMO). 2008.

190 Mayer B, Kylling A. Technical note: The libRadtran software package for radiative transfer calculations - description and examples of use. *Atmos. Chem. Phys.* 2005; 5: 1855-77.

191 NASA. Total ozone mapping spectrometer. 2006.

192 Holick MF. Environmental factors that influence the cutaneous production of vitamin D. *Am J Clin Nutr* 1995; 61: 638S-45S.

193 Lehmann B, Knuschke P, Meurer M. The UVB-induced synthesis of vitamin D3 and 1alpha,25-dihydroxyvitamin D3 (calcitriol) in organotypic cultures of keratinocytes: effectiveness of the narrowband Philips TL-01 lamp (311 nm). *J Steroid Biochem Mol Biol* 2007; 103: 682-5.

194 Leenutaphong V, Sudtim S. A comparison of erythema efficacy of ultraviolet B irradiation from Philips TL12 and TL01 lamps. *Photodermatol Photoimmunol Photomed* 1998; 14: 112-5.

195 Holick MF, Chen TC. Vitamin D deficiency: a worldwide problem with health consequences. *Am J Clin Nutr* 2008; 87: 1080S-6S.

196 Holick MF. Sunlight, UV-radiation, vitamin D and skin cancer: how much sunlight do we need? *Adv Exp Med Biol* 2008; 624: 1-15.

197 Barth J, Gerlach B, Knuschke P *et al.* Serum 25(OH)D3 and ultraviolet exposure of residents in an old people's home in Germany. *Photodermatol Photoimmunol Photomed* 1992; 9: 229-31.

198 Holick MF, Matsuoka LY, Wortsman J. Age, vitamin D, and solar ultraviolet. *Lancet* 1989; 2: 1104-5.

199 Chen TC, Chimeh F, Lu Z *et al.* Factors that influence the cutaneous synthesis and dietary sources of vitamin D. *Arch Biochem Biophys* 2007; 460: 213-7.

200 Devgun MS, Paterson CR, Johnson BE *et al.* Vitamin D nutrition in relation to season and occupation. *Am J Clin Nutr* 1981; 34: 1501-4.

201 Armas LA, Dowell S, Akhter M *et al.* Ultraviolet-B radiation increases serum 25-hydroxyvitamin D levels: the effect of UVB dose and skin color. *J Am Acad Dermatol* 2007; 57: 588-93.

202 Porojnicu AC, Bruland OS, Aksnes L *et al.* Sun beds and cod liver oil as vitamin D sources. *J Photochem Photobiol B* 2008; 91: 125-31.

203 Olds WJ, McKinley AR, Moore MR *et al.* In vitro model of vitamin D(3) (Cholecalciferol) synthesis by UV radiation: Dose-response relationships. *J Photochem Photobiol B* 2008; 93: 88-93.

204 McCarty CA. Sunlight exposure assessment: can we accurately assess vitamin D exposure from sunlight questionnaires? *Am J Clin Nutr* 2008; 87: 1097S-101S.

205 Barbagallo J, Spann CT, Tutrone WD *et al.* Narrowband UVB phototherapy for the treatment of psoriasis: a review and update. *Cutis* 2001; 68: 345-7.

206 Storbeck K, Holzle E, Schurer N *et al.* Narrow-band UVB (311 nm) versus conventional broad-band UVB with and without dithranol in phototherapy for psoriasis. *J Am Acad Dermatol* 1993; 28: 227-31.

207 Larko O. Treatment of psoriasis with a new UVB-lamp. *Acta Derm Venereol* 1989; 69: 357-9.

208 Schiener R, Behrens-Williams SC, Pillekamp H *et al.* Calcipotriol vs. tazarotene as combination therapy with narrowband ultraviolet B (311 nm): efficacy in patients with severe psoriasis. *Br J Dermatol* 2000; 143: 1275-8.

209 Snellman E, Jansen CT, Lauharanta J *et al.* Solar ultraviolet (UV) radiation and UV doses received by patients during four-week climate therapy periods in the Canary Islands. *Photodermatol Photoimmunol Photomed* 1992; 9: 40-3.

210 Thieden E, Agren MS, Wulf HC. The wrist is a reliable body site for personal dosimetry of ultraviolet radiation. *Photodermatol Photoimmunol Photomed* 2000; 16: 57-61.

211 Diffey BL, Larko O, Swanbeck G. UV-B doses received during different outdoor activities and UV-B treatment of psoriasis. *Br J Dermatol* 1982; 106: 33-41.

212 Herlihy E, Gies PH, Roy CR *et al.* Personal dosimetry of solar UV radiation for different outdoor activities. *Photochem Photobiol* 1994; 60: 288-94.

213 Lips P. Vitamin D deficiency and osteoporosis: the role of vitamin D deficiency and treatment with vitamin D and analogues in the prevention of osteoporosis-related fractures. *Eur J Clin Invest* 1996; 26: 436-42.

214 DeLuca HF. The vitamin D story: a collaborative effort of basic science and clinical medicine. *Faseb J* 1988; 2: 224-36.

215 Smith EL, Walworth NC, Holick MF. Effect of 1 alpha,25-dihydroxyvitamin D3 on the morphologic and biochemical differentiation of cultured human epidermal keratinocytes grown in serum-free conditions. *J Invest Dermatol* 1986; 86: 709-14.

216 Kragballe K, Wildfang IL. Calcipotriol (MC 903), a novel vitamin D3 analogue stimulates terminal differentiation and inhibits proliferation of cultured human keratinocytes. *Arch Dermatol Res* 1990; 282: 164-7.

217 Binderup L, Bramm E. Effects of a novel vitamin D analogue MC903 on cell proliferation and differentiation in vitro and on calcium metabolism in vivo. *Biochem Pharmacol* 1988; 37: 889-95.

218 Armas LA, Hollis BW, Heaney RP. Vitamin D2 is much less effective than vitamin D3 in humans. *J Clin Endocrinol Metab* 2004; 89: 5387-91.

219 Holick MF, Siris ES, Binkley N *et al.* Prevalence of Vitamin D Inadequacy Among Postmenopausal North American Women Receiving Osteoporosis Therapy. *Obstet Gynecol Surv* 2005; 60: 658-9.

220 Chesney RW, Rosen JF, Hamstra AJ *et al.* Absence of seasonal variation in serum concentrations of 1,25-dihydroxyvitamin D despite a rise in 25-hydroxyvitamin D in summer. *J Clin Endocrinol Metab* 1981; 53: 139-42.

221 Lips P, Wiersinga A, van Ginkel FC *et al.* The effect of vitamin D supplementation on vitamin D status and parathyroid function in elderly subjects. *J Clin Endocrinol Metab* 1988; 67: 644-50.

222 Bouillon RA, Auwerx JH, Lissens WD *et al.* Vitamin D status in the elderly: seasonal substrate deficiency causes 1,25-dihydroxycholecalciferol deficiency. *Am J Clin Nutr* 1987; 45: 755-63.

223 Adams JS, Clemens TL, Parrish JA *et al.* Vitamin-D synthesis and metabolism after ultraviolet irradiation of normal and vitamin-D-deficient subjects. *N Engl J Med* 1982; 306: 722-5.

224 Landin-Wilhelmsen K, Wilhelmsen L, Lappas G *et al.* Serum intact parathyroid hormone in a random population sample of men and women: relationship to anthropometry, life-style factors, blood pressure, and vitamin D. *Calcif Tissue Int* 1995; 56: 104-8.

225 Kragballe K. *Vitamin D in Dermatology*. 2000.

226 Shultz TD, Fox J, Heath H, 3rd *et al.* Do tissues other than the kidney produce 1,25-dihydroxyvitamin D3 in vivo? A reexamination. *Proc Natl Acad Sci U S A* 1983; 80: 1746-50.

227 Bikle DD, Halloran BP, Riviere JE. Production of 1,25 dihydroxyvitamin D3 by perfused pig skin. *J Invest Dermatol* 1994; 102: 796-8.

228 Vanhooke JL, Prahl JM, Kimmel-Jehan C *et al.* CYP27B1 null mice with LacZreporter gene display no 25-hydroxyvitamin D3-1alpha-hydroxylase promoter activity in the skin. *Proc Natl Acad Sci U S A* 2006; 103: 75-80.

229 Lehmann B. The vitamin D3 pathway in human skin and its role for regulation of biological processes. *Photochem Photobiol* 2005; 81: 1246-51.

230 Lehmann B, Sauter W, Knuschke P *et al.* Demonstration of UVB-induced synthesis of 1 alpha,25-dihydroxyvitamin D3 (calcitriol) in human skin by microdialysis. *Arch Dermatol Res* 2003; 295: 24-8.

231 Lehmann B, Knuschke P, Meurer M. UVB-induced conversion of 7-dehydrocholesterol to 1 alpha,25-dihydroxyvitamin D3 (calcitriol) in the human keratinocyte line HaCaT. *Photochem Photobiol* 2000; 72: 803-9.

232 van de Kerkhof PC. Biological activity of vitamin D analogues in the skin, with special reference to antipsoriatic mechanisms. *Br J Dermatol* 1995; 132: 675-82.

233 Hosomi J, Hosoi J, Abe E *et al.* Regulation of terminal differentiation of cultured mouse epidermal cells by 1 alpha,25-dihydroxyvitamin D3. *Endocrinology* 1983; 113: 1950-7.

234 Merke J, Schwittay D, Furstenberger G *et al.* Demonstration and characterization of 1,25-dihydroxyvitamin D3 receptors in basal cells of epidermis of neonatal and adult mice. *Calcif Tissue Int* 1985; 37: 257-67.

235 Sahota O, Mundey MK, San P *et al.* The relationship between vitamin D and parathyroid hormone: calcium homeostasis, bone turnover, and bone mineral density in postmenopausal women with established osteoporosis. *Bone* 2004; 35: 312-9.

236 Vieth R. Vitamin D supplementation, 25-hydroxyvitamin D concentrations, and safety. *Am J Clin Nutr* 1999; 69: 842-56.

237 Landin-Wilhelmsen K, Wilhelmsen L, Bengtsson BA. Postmenopausal osteoporosis is more related to hormonal aberrations than to lifestyle factors. *Clin Endocrinol (Oxf)* 1999; 51: 387-94.

238 Fogelman I, Blake GM. Different approaches to bone densitometry. *J Nucl Med* 2000; 41: 2015-25.

239 Riggs BL. Pathogenesis of osteoporosis. *Am J Obstet Gynecol* 1987; 156: 1342-6.

240 Cummings SR, Nevitt MC, Browner WS *et al.* Risk factors for hip fracture in white women. Study of Osteoporotic Fractures Research Group. *N Engl J Med* 1995; 332: 767-73.

241 Johnell O, Gullberg B, Kanis JA *et al.* Risk factors for hip fracture in European women: the MEDOS Study. Mediterranean Osteoporosis Study. *J Bone Miner Res* 1995; 10: 1802-15.

242 Cummings SR, Melton LJ. Epidemiology and outcomes of osteoporotic fractures. *Lancet* 2002; 359: 1761-7.

243 Aniansson A, Zetterberg C, Hedberg M *et al.* Impaired muscle function with aging. A background factor in the incidence of fractures of the proximal end of the femur. *Clin Orthop Relat Res* 1984: 193-201.

244 Gunnes M, Lehmann EH, Mellstrom D *et al.* The relationship between anthropometric measurements and fractures in women. *Bone* 1996; 19: 407-13.

245 Kanis JA, Johansson H, Oden A *et al.* A family history of fracture and fracture risk: a meta-analysis. *Bone* 2004; 35: 1029-37.

246 Fox KM, Cummings SR, Powell-Threets K *et al.* Family history and risk of osteoporotic fracture. Study of Osteoporotic Fractures Research Group. *Osteoporos Int* 1998; 8: 557-62.

247 Seeman E, Cooper ME, Hopper JL *et al.* Effect of early menopause on bone mass in normal women and patients with osteoporosis. *Am J Med* 1988; 85: 213-6.

248 Kanis JA. Estrogens, the menopause, and osteoporosis. *Bone* 1996; 19: 185S-90S.

249 Kleerekoper M, Brienza RS, Schultz LR *et al.* Oral contraceptive use may protect against low bone mass. Henry Ford Hospital Osteoporosis Cooperative Research Group. *Arch Intern Med* 1991; 151: 1971-6.

250 Scoutellas V, O'Neill TW, Lunt M *et al.* Does the presence of postmenopausal symptoms influence susceptibility to vertebral deformity? European Vertebral Osteoporosis Study (EVOS) Group. *Maturitas* 1999; 32: 179-87.

251 Cooper C, Barker DJ, Wickham C. Physical activity, muscle strength, and calcium intake in fracture of the proximal femur in Britain. *Bmj* 1988; 297: 1443-6.

252 Silman AJ, O'Neill TW, Cooper C *et al.* Influence of physical activity on vertebral deformity in men and women: results from the European Vertebral Osteoporosis Study. *J Bone Miner Res* 1997; 12: 813-9.

253 Frandin K, Grimby G, Mellstrom D *et al.* Walking habits and health-related factors in a 70-year-old population. *Gerontology* 1991; 37: 281-8.

254 Greendale GA, Barrett-Connor E, Edelstein S *et al.* Lifetime leisure exercise and osteoporosis. The Rancho Bernardo study. *Am J Epidemiol* 1995; 141: 951-9.

255 van der Klift M, de Laet CE, McCloskey EV *et al.* Risk factors for incident vertebral fractures in men and women: the Rotterdam Study. *J Bone Miner Res* 2004; 19: 1172-80.

256 Rundgren A, Mellstrom D. The effect of tobacco smoking on the bone mineral content of the ageing skeleton. *Mech Ageing Dev* 1984; 28: 273-7.

257 Kanis JA, Johnell O, Oden A *et al.* Smoking and fracture risk: a meta-analysis. *Osteoporos Int* 2005; 16: 155-62.

258 Johansson C, Mellstrom D. An earlier fracture as a risk factor for new fracture and its association with smoking and menopausal age in women. *Maturitas* 1996; 24: 97-106.

259 Kanis JA, Johansson H, Oden A *et al.* A meta-analysis of prior corticosteroid use and fracture risk. *J Bone Miner Res* 2004; 19: 893-9.

260 Huang C, Ross PD, Lydick E *et al.* Factors associated with joint pain among postmenopausal women. *Int J Obes Relat Metab Disord* 1997; 21: 349-54.

261 Jones SM, Harris CP, Lloyd J *et al.* Lipoproteins and their subfractions in psoriatic arthritis: identification of an atherogenic profile with active joint disease. *Ann Rheum Dis* 2000; 59: 904-9.

262 Mallbris L, Granath F, Hamsten A *et al.* Psoriasis is associated with lipid abnormalities at the onset of skin disease. *J Am Acad Dermatol* 2006; 54: 614-21.

263 Burger D, Dayer JM. High-density lipoprotein-associated apolipoprotein A-I: the missing link between infection and chronic inflammation? *Autoimmun Rev* 2002; 1: 111-7.

264 Furumoto H, Arai H, Kuramitsu Y *et al.* Lipoproteins modulate growth and differentiation of cultured human epidermal keratinocytes. *Electrophoresis* 2002; 23: 161-6.

265 Girolomoni G, Gisondi P. Psoriasis and metabolic comorbidities: the importance of well-designed prospective studies. Commentary. *Dermatology* 2008; 217: 222-4.

266 Eckel RH, Grundy SM, Zimmet PZ. The metabolic syndrome. *Lancet* 2005; 365: 1415-28.

267 Wannamethee SG, Shaper AG, Lennon L *et al.* Metabolic syndrome vs Framingham Risk Score for prediction of coronary heart disease, stroke, and type 2 diabetes mellitus. *Arch Intern Med* 2005; 165: 2644-50.

268 Gisondi P, Tessari G, Conti A *et al.* Prevalence of metabolic syndrome in patients with psoriasis: a hospital-based case-control study. *Br J Dermatol* 2007; 157: 68-73.

269 Tilg H, Moschen AR. Adipocytokines: mediators linking adipose tissue, inflammation and immunity. *Nat Rev Immunol* 2006; 6: 772-83.

270 Barth J, Kohl V, Hanefeld M. [Behavior of lipids, other serum parameters and cardiovascular functions in ultraviolet therapy]. *Hautarzt* 1994; 45: 702-7.

271 Neimann AL, Shin DB, Wang X *et al.* Prevalence of cardiovascular risk factors in patients with psoriasis. *J Am Acad Dermatol* 2006; 55: 829-35.

272 Das UN. Is angiotensin-II an endogenous pro-inflammatory molecule? *Med Sci Monit* 2005; 11: RA155-62.

273 Huskic J, Alendar F, Matavulj A *et al.* Serum angiotensin converting enzyme in patients with psoriasis. *Med Arh* 2004; 58: 202-5.

274 Ucak S, Ekmekci TR, Basat O *et al.* Comparison of various insulin sensitivity indices in psoriatic patients and their relationship with type of psoriasis. *J Eur Acad Dermatol Venereol* 2006; 20: 517-22.

275 Carr DB, Utzschneider KM, Hull RL *et al.* Intra-abdominal fat is a major determinant of the National Cholesterol Education Program Adult Treatment Panel III criteria for the metabolic syndrome. *Diabetes* 2004; 53: 2087-94.

276 Libby P. Inflammation and cardiovascular disease mechanisms. *Am J Clin Nutr* 2006; 83: 456S-60S.

277 Austin RC, Lentz SR, Werstuck GH. Role of hyperhomocysteinemia in endothelial dysfunction and atherothrombotic disease. *Cell Death Differ* 2004; 11 Suppl 1: S56-64.

278 Clarke R, Daly L, Robinson K *et al.* Hyperhomocysteinemia: an independent risk factor for vascular disease. *N Engl J Med* 1991; 324: 1149-55.

279 Thambyrajah J, Townend JN. Homocysteine and atherothrombosis--mechanisms for injury. *Eur Heart J* 2000; 21: 967-74.

280 Malerba M, Gisondi P, Radaeli A *et al.* Plasma homocysteine and folate levels in patients with chronic plaque psoriasis. *Br J Dermatol* 2006; 155: 1165-9.

281 Vanizor Kural B, Orem A, Cimsit G *et al.* Plasma homocysteine and its relationships with atherothrombotic markers in psoriatic patients. *Clin Chim Acta* 2003; 332: 23-30.

282 Grissom CB, Chagovetz AM, Wang Z. Use of viscosigens to stabilize vitamin B12 solutions against photolysis. *J Pharm Sci* 1993; 82: 641-3.

283 Salmon S, Maziere JC, Santus R *et al.* UVB-induced photoperoxidation of lipids of human low and high density lipoproteins. A possible role of tryptophan residues. *Photochem Photobiol* 1990; 52: 541-5.

284 Kelly GS. Seasonal variations of selected cardiovascular risk factors. *Altern Med Rev* 2005; 10: 307-20.

285 Carbone LD, Rosenberg EW, Tolley EA *et al.* 25-Hydroxyvitamin D, cholesterol, and ultraviolet irradiation. *Metabolism* 2008; 57: 741-8.