# Path To Beauty

## ADVANCED SKIN CARE GUIDE

Nicole Taylor & Karen Miller

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# Chapter 1. Introduction

Good health plays an important role in nutrition and skin quality. Everyone desires to live longer and healthier lives while maintaining a youthful appearance. The dining table is the chief way from the food producer to eater. Farmer who harvests different season fruits and vegetables to manufacturer and markets tocopherols, polyphenols and plant extracts, There are many other ways to reach the consumer. Nutritional products are interlinked to health, wellness, and needs of consumer. Marketing power behind these products are pharmacological activity of Nutraceuticals which are necessary for living.
Vitamins and essential fatty acids and their tremendous impact on skin health are very important. To concentrate not only on what we know but what we do not know to meet consumer needs and to elucidate not only the potential health benefits that certain foods bring to various tissues and organs is very vital. Food and good nutrition to overall health and appearance is necessary for all human beings. Everybody is very much aware of his or her appearance. Skin is the largest organ of the human body. Skin is very necessary for thermoregulation, protection, metabolism, and sensation. Nutrients are essential for normal skin functions, and presence and function still attract many scientists. Vitamins and amino acids, many proactive agents as well with beneficial pharmacological activity that belong to the wider category of Nutraceuticals.

Many scientists and nutritionist have done their work in this field which are discussed in each chapter.

- Zouboulis and Elewa reviewed the multiple activities of that are used as rapeutic agents for various skin diseases.
- Reichrath and Trémezaygues undertook to discuss multifaceted duty of vitamin D and organically necessary metabolites.
- Burke eloquently reviews the photoprotection that vitamins C and E offer against induced photodamage.
- Richelle's team arranged information on antioxidants in skin research to date, the carotenoids.
- Lademann's team complements the vast range of antioxidant activities and their benefits to the skin, revealing in addition ways to assess their important activities.
- Petra Winkler reviewed several minerals and their influence on skin.
- Boyle and coworkers offered puzzling relation of probiotics and skin with a clinical research.
- Larsen Smith) worked on relation of the glycemic load and acne symptoms through development in insulin metabolism.
- Rawlings distills inscribed atopic dermatitis as relates to nutrition with essential fatty acid metabolism.
- Anthonavage identifies research areas unchartered by nutritionists and food scientists with respect to hair biology.
- Stamatas and Kollias captured enormous potential that is available to scientists and health professionals of imaging and spectroscopic techniques.
- Zouboulis's team tackled immense job of nutritional clinical studies in dermatology.

# Chapter 2. Vitamin A and the Skin

Vitamin A is essential for normal differentiation and maintenance of epithelial tissues in skin and mucous membranes, retinaldehyde, reproduction and embryonic morphogenesis. Retinoid are used in negotiating therapeutic for hyperkeratotic and parakeratotic skin diseases. Acne, acne-related disorders and hand eczema and used as prophylaxis for epithelial skin tumors in immune-suppressed patients and solution for no melanoma skin cancers and cutaneous T-cell lymphoma.

**2.1 Introduction:** Retinoids have biological activities similar to those of naturally occurring vitamin A but not necessarily the same chemical structure. They are used as therapy and or as local applications for various skin diseases and tumors, in the cosmetic field for acne, seborrhea, psoriasis, epithelial tumors, and hand eczema.

**2.2 Naturally Occurring Retinoids:** Natural retinoids include vitamin A and its metabolic derivatives retinaldehyde and retinoic acid. The normal concentration of vitamin A in plasma is 0.35–0.75 mg/ml. Retinoic acid is produced by in vivo oxidation of retinol. Its two isoforms are all-trans retinoic acid and 13-*cis* retinoic acid, with normal plasma concentrations of 0.55–1.20 and 0.80–2.40 ng/mL, respectively. Retinoic acid can fully substitute for retinol, except for maintaining reproduction. Daily requirement of vitamin A is 0.8–1.0 mg (2,400–3,000 IU), which can be found in ten medium-sized eggs or 100 g of butter. Hypervitaminosis occurs with intake of more than 18,000–60,000 IU vitamin A per day for children and 50,000–1,00,000 IU for adults.

**2.3 Synthetic Retinoids:** The synthetic retinoids are chemical modifications of naturally occurring vitamin A and are chemically different compounds with the capability to secure retinoid nuclear receptor proteins.

**2.4 Absorption, Distribution, and Metabolism:** The oral bioavailability of retinoids can be enhanced by fatty acids, which stop binding of retinoids with albumin and improve the clinical effect. The metabolism of retinoids arise in liver. It involves oxidation and chain shortening to produce biologically inactive metabolites. Retinoids are excreted through feces and urine.
Isotretinoin is observed after 30 min in blood and maximum clusters are reached 2–4 h after oral intake. Half-life elimination rate of isotretinoin ranges from 7 to 37 h, and its known metabolites is 11–50 h. Prime metabolites of isotretinoin in blood are 4-oxo- and 4-hydroxy-isotretinoin. 99% of isotretinoin in plasma is bound to plasma proteins and commonly albumin. Serum albumin has a critical function as a retinoid-binding protein to lessen concentration of active retinoids and limiting biological effects on sebaceous gland cells. Acitretin is eliminated Acitretin is eliminated more rapidly than etretinate. Etretinate is highly lipophilic, binds strongly to albumin, kept in adipose tissue and is released slowly whereas the half-life of acitretin is only 2 days. Usage of acitretin to etretinate happen only in case of alcohol utilization. Bexarotene in plasma is 99% bound to plasma proteins. It is discharged by hepatoiliary system, and its terminal half-life is 7–9 h.

**2.5 Mechanism of Action**

***2.5.1 Retinoid Receptors and Gene Regulation:*** Retinoids enter cell by non-receptor-mediated endocytosis, meet with cytosolic proteins and bind to nuclear receptors. The retinoid nuclear receptors are members of steroid thyroid hormone receptor superfamily. Retinoid A receptors (RAR) bind all-trans retinoic acid and 9-*cis* retinoic acid with high affinity and bind to 13-*cis* retinoic acid. Retinoid X receptors (RXR) also bind 9-*cis* retinoic acid and discern for bexarotene which is a specific RXR ligand. 14-Hydroxy-retro-retinol does not initiate retinoid receptors. Retinoid effects happen by influence of retinoid receptors with other signal transduction structure.

***2.5.2 Effect on Epidermal Growth and Differentiation:*** Retinoids encourage cell proliferation in normal epithelia and normalize it in hyperproliferative conditions. Retinoids induce and modulate the expression growth factors and their receptors. Keratinocyte proliferation by retinoids is mediated by organizing cyclic adenosine monophosphate (cAMP), epidermal growth factor (EGF), protein kinase C (PKC) and transforming growth factor-a (TGFa). Retinoids down-regulate cell growth and is mediated by a TGFb2-regulated inhibition of the EGF binding to its receptor. Retinoids have also a keratolytic effect through shifting the terminal keratinocyte differentiation toward a nonkeratinizing mucosa.

***2.5.3 Effects on Sebaceous Gland Activity***.

***2.5.4 Immunomodulatory and Anti-inflammatory Properties:*** Isotretinoin, etretinate, and acitretin manifested inhibit angiogenesis both in vitro and in vivo most probably through RAR-a. Retinoids encourage humoral and cellular resistance by strenghting antibody production, increasing blood T-helper cells and preventing Langerhans cells depletion from epidermis by ultraviolet light. Unease of neutrophil migration in psoriatic skin, LTB4-induced trek of neutrophils and inhibition of nitric oxide and tumor necrosis factor-a (TNFa) production.

**2.6 Therapeutic Uses**

***2.6.1 Psoriasis and Related Disorders:*** The orally managed aromatic retinoids are used to treat psoriasis both initial and continuity. Retinoids have a synergistic effect with other psoriasis treating modal quality. The daily dose of acitretin is 0.5 mg/kg body weight. Taking acitretin with meals that have some fat increases the blood absorption two- to fivefold. Retinoids being metabolized in the liver interact with ketoconazole and phenytoin but not with oral contraceptives.

***2.6.2 Disorders of Keratinization:*** Etretinate and acetretin are better than isotretinoin because of the sebumdrying property. Seriousness of Darier's disease, ichthyosis vulgaris, congenital ichthyosis and palmoplantar keratodermas is swayed with retinoids.

***2.6.3 Seborrhea, Acne, and Acneiform Disorder:*** Isotretinoin proved to be the most effective sebostatic retinoid both in vivo and in cell cultures and is best retinoid for treating severe acne. It shows low binding affinity for intracellular retinoidbinding proteins and heavy sebostatic activity. It undergoes a specific and selective intracellular isomerization process into tretinoin, which in turn binds to RARs. It decreases sebum production, number of proliferating sebocytes, size of sebaceous gland, inhibits sebocyte differentiation in vivo, vitro and directly suppresses abnormal desquamation of sebaceous follicles. It acts in a receptor-independent technique by impacting cellular signaling pathways through direct

protein reciprocal activity. It minimize monocyte and neutrophil chemotaxis and migration to trekking of epidermis and minimizing excessive inflammation that causes marking.

**2.6.3.1 Dosing, Therapeutic Effect, and Monitoring:** The required dose is 0.5 mg/kg/day, an initially high dose for 3 months and maintenance requires a lower dose. A cumulative dose of more than 150 mg/kg administered over 6–12 months has been considered necessary to ensure a long-lasting remission. Longer treatment duration might be needed in patients with extrafacial lesions, low-dose therapy, or severe acne. Treatment of acne should start at 0.5 mg/kg, and that it should be used only for severe acne that is not responding to antibiotics or topical therapy; it should not be used as a first-line treatment. Only 30 days of oral isotretinoin can be supplied to female patients at a time.

**2.6.3.2 Retinoid Local Therapy in Acne:** Tretinoin, isotretinoin, motretinide, adapalene and tazarotene are used in treatment of acne. Retinaldhyde, retinol, and retinyl esters are used in cosmetic preparations. Topical retinoids were found to perform their therapeutic action by increasing follicular epithelium turnover, reversing abnormal desquamation of the sebaceous duct. Maintenance therapy is required for a chronic disease such as acne. Topical adapalene is also adequate for maintenance treatment.

***2.6.4 Retinoids in Skin Cancer***

**2.6.4.1 Prevention of Keratinocyte Skin Cancers:** Retinoids are used for chemoprevention and chemosuppression in many diseases and syndromes with high susceptibility of nonmelanoma skin cancer development. Retinoids regulate MMPs, TGFb, cyclin-dependent kinase 1, P16, and P21. They are capable of regulating tumor stroma production and control tumor progression and invasion RAR-related antiproliferative and proapoptotic signals may also be involved. Isotretinoin plus interferon-a for treatment of hostile skin SCC showed high rates of tumor and second primary tumors.

**2.6.4.2 Therapy of Other Skin Cancers:** Melanoma was nonsensitive to retinoids. Bexarotene gel was introduced as a monotherapy for treatment of CTCL in early stages which were refractory or intolerant to at least two other treatment modalities for more than 6 months. Alitretinoin, as a 0.1% gel, was initiated as adjuvant topical regimen for Kaposi sarcoma associated with AIDS.

***2.6.5 Chronic Hand Eczema:*** Eczema of the hands is a relatively common disease in 6–8% of the population. Alitretinoin given at well-tolerated doses induces substantial clearing of chronic hand dermatitis in patients mulished to usual topical treatment. Complete or near-complete disappearance of disease signs and symptoms was reported for 53% of patients treated with the highest alitretinoin dose (40 mg/day). The response was reported with all types of chronic hand eczema. Treatment is generally tolerated, with the typical retinoid adverse effects at the highest dose. Bexarotene, applied topically in 1% concentration also proved to be effective for treatment of chronic hand eczema.

**2.7 Adverse Reactions and Tolerability**

***2.7.1 Mucocutaneous Complications:*** Issue with retinoids is skin and mucous membrane dryness, including cheilitis, which occurs in about 90% of cases. Mucocutaneous xerosis (30%), nosebleeds (15%), mild hair loss, augmented skin fragility, and palmoplantar desquamation (80%) are additional adverse effects. The use of skin emollients, artificial eye

tears, vaseline for the inner part of the nose, and moisturizers for the lips are a necessity from the start of the treatment to avoid the dry skin complications.

***2.7.2 Ocular and Neurological Complications:*** Decreased tear production and lipid content of the tear film lead to dryness of the eyes and keratitis and corneal erosions can occur. The use of contact lenses is contraindicated and artificial tears are mandatory. Other issues comprise blurred vision, declined vision, photophobia, shrinked dark adaptation, papilledema, corneal opacities, and retinal dysfunction. Blepharoconjunctivitis, which may be complicated with *Staphylococcus* infection, occurs in 20–50% of cases. Pseudotumor cerebri is the most important neurological side effect. The most frequent central nervous system adverse effect associated with oral isotretinoin is headache.

***2.7.3 Serum Lipids, Gastrointestinal Side Effects, Liver Function, and Endocrine Adverse Effects:*** Increased serum lipids—hypertriglyceridemia (20–40%) being more common than hypercholesterolemia with decreased high-density lipoprotein and increased low-density lipoproteinemia as well as mild transaminase increase are characteristic metabolic side effects of retinoids. Lipid profile changes are more likely to occur in patients with predisposing factors as obesity, familial hyperlipidemia, nicotine abuse, and diabetes as well as in patients using b-blockers and contraceptive pills. Chronic liver toxicity is although acutely elevated liver enzymes is not uncommon mostly occurring with etretinate. Reintroduction is nominated with enzyme level normalization with 25% of the original dose. Bexarotene causes significant central hypothyroidism and hyperlipidemia in most patients managed with thyroid replacement and hypolipidemic drugs. Hypothyroidism is in part due to increased peripheral thyroid hormone metabolism.

***2.7.4 Long-Term Toxicity: Bone Changes:*** The long-term bone toxicity occurs mostly with vitamin A chronic toxicity. The changes include hyperostosis and periostosis, demineralization, thinning of the bones, and premature epiphysial closure. Radiography of significantly symptomatic joints is recommended with long-term therapy. Yearly radiography of the ankle or thoracic spine is optional.

***2.7.5 Arthralgias and Myalgias:*** Diffuse idiopathic skeletal hyperostosis-like hyperostosis, lesions mimicking seronegative spondyloarthropathy, arthralgia, myalgia, stiffness, true myopathy, muscular damage, rhabdomyolysis, and museuloskeletal pain can occur under retinoid treatment, especially isotretinoin. Creatine phosphokinase, a specific marker of muscle destruction, has been found to be elevated, occasionally by up to 100 times the normal value. Oral acitretin has been found to cause peripheral nerve dysfunction, particularly of sensory fibers, which in rare cases leads to clinically evident sensory disturbances. Less clear is the causal relation between acitretin and benign intracranial hypertension or myopathy. Intense physical exercise and concurrent treatment with neurotoxic or myotoxic drugs should be avoided during treatment with oral retinoids. When arthralgias and myalgias occur, it is recommended that the dose be decreased by 25% until the symptoms resolve.

***2.7.6 Teratogenicity:*** Retinoids freely cross the placenta, causing severe fatal fetal malformations including craniofacial deformities, bone and cardiovascular abnormalities, and endocrine malfunctions. The pregnancy-preventing program for female patients during their childbearing period is strictly applied to isotretinoin therapy. This includes a medically

supervised pregnancy test before, during, and 5 weeks after therapy begins. For female patients, only 30 days of oral isotretinoin can be supplied at a time.

# Chapter 3. Relevance of the Cutaneous Vitamin D Endocrine System for Skin Physiology and Treatment of Skin Diseases

**Léa Trémezaygues and Jörg Reichrath**

**Core Messages**

• Vitamin D can be absorbed from the diet or synthesized in the skin from 7 dehydrocholesterol under the influence of ultraviolet B radiation.
• The keratinocyte is the only cell type known today that is able to synthesize the biologically active vitamin D metabolite 1,25(OH)2D3 from 7-dehydrocholesterol.
• Dose-dependent effects vitamin D analogues on cell proliferation and differentiation as well as effects on immunomodulatory apoptosis, antioxidative and cytoprotective effects.
• 1.25(OH)2D3 and numerous of its analogues are used in the treatment of psoriasis and other skin diseases.

**3.1 Introduction:** The skin is the only organ that is capable of synthesizing vitamin D after exposure to sunlight. On the other hand, ultraviolet (UV) exposure is one of the major risk factors for epithelial skin cancer. Thus, there is a conflict between the positive and negative properties of sunlight.

**3.2 Vitamin D Synthesis in Skin:** Vitamin D, the precursor of the biologically active vitamin D metabolite 1.25- dihydroxyvitamin D [1.25(OH)2D] can be absorbed from the diet or synthesized in skin under the influence of UVB radiation from 7-dehydrocholesterol (7-DHC). Nine enzymatic reactions are involved in the photochemical cutaneous synthesis of vitamin D. Vitamin D2 can be found in plants, vitamin D3 is photochemically synthesized under the influence of UVB radiation in the skin of animals and humans. The biologically active vitamin D metabolite 1.25(OH)2D, or calcitriol, which circulates in the blood is synthesized from vitamin D by a well-characterized biochemical reaction cascade. Production of 1.25-(OH)2D3 in the kidney is regulated by a feedback mechanism of the hormone itself as well as by parathyroid hormone, calcium, and cytokines such as interferon-g (IFNg) and tumor necrosis factor-a (TNFa). The reduction of 1.25(OH)2D3 to 24,25-dihydroxycholecalciferol in the kidney and consecutive reactions are effectuated by a third cytochrome P450 enzyme, 1,25-dihydroxyvitamin D-24-hydroxylase (CYP24A1).

**3.3 Biological Effects of 1.25-(OH)2D3 in the Skin**

***3.3.1 Inhibition of Proliferation and Induction of Differentiation in Keratinocytes:***

Numerous in vitro and in vivo studies have demonstrated dose-dependent effects of vitamin D analogues on cell proliferation and differentiation. At low concentrations (<10−8 M), calcitriol promotes proliferation of keratinocytes in vitro. In psoriatic skin, immunohistochemical and biochemical analysis have clearly shown antiproliferative and

differentiation-inducing effects in epidermal keratinocytes along with treatment with vitamin D analogues in vivo. In lesional psoriatic skin, the clinical improvement correlates with an increase of VDR mRNA in calcitriol-treated skin areas.

***3.3.2 Immunmodulatory Effects in the Skin:*** Various cell types involved in immunological reactions not only express VDR, they possess the enzymatic machinery for local synthesis of calcitriol. Impressive therapeutic effects were seen after application of vitamin D analogues for diseases that are related with the function of T cells or dendritic cells. Vitamin D analogues suppress in vitro immunoglobulin E (IgE) production and IgE-mediated cutaneous reactions.

***3.3.3 Regulation of Apoptosis:*** 1.25-Dihydroxyvitamin D3 has the potential to induce the neutral Mg2+-dependent sphingomyelinase, which hydrolyzes sphingomyelin to ceramid. In contrast, pharmacological concentrations of 1.25-dihydroxyvitamin D3 (³10−6 M) do induce apoptosis. Concentrations of 1.25-dihydroxyvitamin D3 around 10−11 M stimulate cell proliferation. Higher concentrations have a dosedependent antiproliferative effect.

***3.3.4 Cytoprotective Effects:*** All types of skin cancer, basal cell carcinoma (BCC) and squamous cell carcinoma (SCC), is induced by UV exposure. The total UV dose accumulated in the exposed skin area during a lifetime is responsible for the genesis of SCCs. The origin of BCCs is presumed to be both in the accumulated UV dose in the exposed skin area over a lifetime and in higher intermittent UV doses. Excessive UV exposure, especially UVB with a wavelength range of 290–320mn, is consequently a major risk factor in the development of skin cancer. Sufficient doses of UVB are needed for the photochemical synthesis of vitamin D in the skin.
Cutaneous vitamin D synthesis may act as an evolutionary highly conserved natural protection system against the hazardous effects of UVB radiation in the skin. The cytoprotective effects of 1,25-dihydroxyvitamin D3 in UVB-irradiated keratinocytes using morphological and colorimetric vitality Assays. 1.25-Dihydroxyvitamin D3 doses of 10−6–10−8 M were necessary to obtain a considerable cytoprotective effect Using an enzyme-linked immunosorbent assay (ELISA) that detects DNA fragmentation, they showed that pretreatment of the keratinocytes with 1,25-dihydroxyvitamin D3 (1 mM) over 24 h suppresses UVB-induced apoptosis up to 55–70%. Pretreatment of keratinocytes with 1,25-dihydroxyvitamin D3 (1 mM) reduces the mitochondrial cytochrome c release—a marker of UVB-induced apoptosis—up to 90%. Two important mediators of the UV answer—activation of the -Jun-NH2- terminal kinase and production of IL-6—are also reduced about 30% by a pretreatment with 1,25-dihydroxyvitamin D3 (1 mM). Even the UVB-induced cleavage of PARP is inhibited by 24 h of pretreatment of the cells with 1,25-dihydroxyvitamin D3 (1 mM).

Metallothionein MT is an antioxidant and acts as a radical catcher after UV irradiation. 1.25-dihydroxyvitamin D3 protects keratinocytes by induction of antiapoptotic proteins such as Bcl-2 and activation of the MEK/ERK and PI-3 K/Akt metabolic pathways before apoptosis. 1.25-dihydroxyvitamin D3 (10−7 M) has a photoprotective effect in WST-1- and crystal violet-based assays after irradiation of the keratinocytes with ascending doses of UVB light (100–1,000 J/cm2). The photocarcinogenesis of skin cancer is mainly due to mutations resulting from insufficiently repaired DNA photoproducts. 1.25-dihydroxyvitamin D3 prevents human keratinocytes from the induction of CPDs after UVB irradiation Gupta et al.

Reduced immunoreactivity for gH2AX caused by ionizing radiation after pretreating the cells with 1,25-dihydroxyvitamin D3.

***3.3.5 Antioxidative Effects:*** 1.25-Dihydroxyvitamin D3 has a photoprotective effect on keratinocytes in vitro. It induces production of the protein metallothionein in keratinocytes. This represent an important mechanism, protecting the cells against the UVB-induced synthesis of reactive oxygen radicals.

**3.4 Clinical Studies of Vitamin D and Its Analogues in Psoriasis and Other Skin Diseases:**
The importance of 1.25(OH) 2D3 and its analogues for the treatment of psoriasis resulted from two independent lines of investigation. In 1985, Morimoto and Kumahara reported that a patient who was treated orally with 1a-hydroxyvitamin D3 for osteoporosis had a drastic remission of psoriatic skin lesions. Numerous vitamin D analogues, including calcitriol, calcipotriol, tacalcitol, maxacalcitol, and beocalcidiol, are effective and safe for topical treatment of psoriasis. Calcipotriol is more effective for topical treatment of psoriasis than betamethasone 17-valerate ointment.
Calcipotriol cream has been reported to be as effective as and cosmetically more favorable than coal tar in a small, observer-blinded trial in patients with psoriasis. Recently, twice daily calcipotriol ointment was compared with once-daily shortcontact dithranol cream therapy in a randomized controlled trial (RCT) of supervised treatment of psoriasis in a daycare setting. This multicentre RCT, which was performed in six centers in The Netherlands, included 106 patients with chronic plaque psoriasis. No statistically significant differences were found between the calcipotriol and the dithranol group in any of the QOL domains or scales of the Skindex-29 and the SF-36 at the end of treatment.
Effective application of calcipotriol cream is mean percentage reduction in PASI. The reduction in PASI with twice-daily application of calcipotriol cream did not differ from that with application of calcipotriol cream in the morning plus clobetasone butyrate cream in the evening (53.7%) and was significantly lower than that with application of calcipotriol cream in the morning plus betamethasone valerate cream in the evening (57.5%). In that study, the efficacy and safety of two dosing regimens of becocalcidiol ointment in the treatment of plaque-type psoriasis were evaluated. Vitamin D analogues are effective in the treatment of psoriatic skin lesions in children and in human immunodeficiency virus (HIV)-infected patients.

**3.5 Treatment**
***3.5.1 Scalp Psoriasis:*** In a double-blind, calcipotriol solution is an effective topical treatment for scalp psoriasis. A total of 49 patients were treated twice a day over a 4-week period and 60% of patients treated with calcipotriol showed clearance or marked improvement of their scalp psoriasis versus 17% in the placebo group. No side effects were reported.

***3.5.2 Nail Psoriasis:*** Nail psoriasis has been reported in up to 50% of patients with psoriasis. Nails in general are difficult to treat and respond slowly. Calcipotriol ointment is effective in the treatment of nail psoriasis.

***3.5.3 Face and Flexures:*** Use of calcipotriol ointment is not recommended on face and flexures due to its irritant nature, most patients tolerate vitamin D analogues on these sites. The tolerability and efficacy of calcitriol 3 mg/g and tacrolimus 0.3 mg/g ointment in chronic plaque psoriasis affecting facial and genitofemoral regions was analyzed. In this double-

blind, parallel, 6 week study, 50 patients were randomized in a 1:1 ratio to apply calcitriol or tacrolimus twice a day. Tacrolimus was significantly more effective than calcitriol based on a significant reduction of mean TAS (67% vs. 51%; $P < 0.05$) as well as more patients achieving complete or almost complete clearance by the PGA (60% vs. 33%; $P < 0.05$). Both calcitriol 3 mg/g and tacrolimus 0.3 mg/g are safe and well-tolerated therapeutic agents for treating psoriasis in sensitive areas.

***3.5.4 Skin Lesions in Children:*** Topical application of calcitriol ointment is an effective, safe, reliable therapy to cure psoriatic skin lesions in children.

***3.5.5 Psoriatic Lesions in HIV Patients:*** Holick treated an HIV-positive patient suffering from psoriatic skin lesions with topical and oral calcitriol and patient responded no evidence of enhanced HIV disease. Other case reports also demonstrated the efficacy and safety of vitamin D analogues in the treatment of psoriasis.

***3.5.6 Combination of Vitamin D Analogues with Other Therapies:*** The efficacy of topical treatment with vitamin D analogues against psoriasis can be augmented by combining it with other therapies, including very low dose oral cyclosporine (2 mg/kg/day), oral acitretin, topical dithranol, topical steroids, and UVB or narrow-band UVB phototherapy. Addition of calcipotriol ointment to oral application of acitretin was demonstrated to produce a significantly better treatment response achieved with a lower cumulative dose of acitretin. Combined topical treatment with calcipotriol ointment (50 mg/g) and betamethasone ointment was recently shown to be slightly more effective and caused less skin irritation than calcipotriol used twice a day. Therapy of psoriasis with combined topical calcipotriol and narrow-band UVB has been highly effective for treating psoriatic plaques. The therapeutic effects of vitamin D analogues were not clinically inactivated by subsequent irradiation with PUVA or NB-UVB phototherapy.

***3.5.7 Other Skin Disorders with Vitamin D Analogues:*** Vitamin D3 was used in dermatology in large pharmacological doses for the treatment of scleroderma, psoriasis, lupus vulgaris, and atopic dermatitis. Attempts of vitamin D treatment in dermatology were rapidly abandoned because of severe vitamin D intoxication that caused hypercalcemia, hypercalciuria, and kidney stones.

**3.5.7.1 Vitamin D and Ichthyosis:** A double-blind, bilaterally paired, comparative study showed the effectiveness of topical treatment with calcipotriol ointment on congenital ichthyose. Patient suffering from ichthyosis bullosa of Siemens who was treated with calcipotriol did not show any change in severity on the calcipotriol-treated side compared to the vehicle-treated side.

**3.5.7.2 Vitamin D and Scleroderma:** Oral administration of calcitriol 1.0–2.5 mg/day alleviates skin involvement is probably by inhibiting fibroblast proliferation and dermal collagen deposition.

**3.5.7.3 Vitamin D and Vitiligo:** Vitamin D analogues may be efficient and safe in the treatment of vitiligo.

**3.5.7.4 Vitamin D and Skin Cancer:** Antiproliferative and prodifferentiating effects of vitamin D analogues in many VDR-expressing tumor cell lines, including malignant melanoma, SCC, and leukemic cells are strong. Active vitamin D analogues block proliferation and tumor progression of epithelial tumors in Rats. Inhibition of tumor growth of human malignant melanoma and colonic cancer xenografts was also mice, but only at high doses of calcitriol.

**3.5.7.5 Vitamin D and Other Skin Diseases:** Positive effects of topical treatment with vitamin D analogues in a variety of skin diseases like transient acantholytic are demonstrated. Dermatosis, inflammatory linear verrucous epidermal nevus, disseminated superficial actinic porokeratosis, pityriasis rubra pilaris and epidermolytic palmoplantar keratoderma.

**Summary:**

• Sunscreen alone is not sufficient to provide optimal protection from ultraviolet induced and other environmental free-radical damage to the skin.
• The antioxidants vitamins C and E have been extensively researched and proven to protect the skin against photodamage.
• When formulated correctly and at a high enough concentration, vitamins C and E are more protective when applied topically to the skin than if taken orally.
• An effective formulation requires the natural molecular form of the antioxidant in a composition that maintains stability.

**4.1 Introduction:** Our skin is the largest organ of our body and the organ most exposed to the environment. Our skin suffers from ultraviolet A (UVA) and B (UVB) exposure as well as from urban pollutants. Research has recently demonstrated a synergistic enhancement of oxidative damage to the skin when the skin is exposed to UVA.

# Chapter 4. Photoprotection of the Skin with Vitamins C and E: Antioxidants and Synergies

**Karen E. Burke**

Environmental irritants, including cigarette smoke can damage which leads to unattractive premature aging of skin and cancers of the skin. Devastating is the fact that one in four potentially *lethal melanomas* occurs in individuals younger than 40 years. 90% of skin cancers and all photoaging are caused by sun exposure. fewer than 33% of the population apply sunscreen regularly despite the current increased publicity promoting the need for protection.

First, even when applied properly at 2 mg/cm2, broad-spectrum sunscreens decrease free radical damage by only 55%. Second, even when conscientious individuals apply sunscreen generously, they can usually apply only one-fourth of the amount required to attain the full SPF. The Federal Drug Administration (FDA) has specified required daily amounts (RDA) of all vitamins to prevent deficiency diseases such as scurvy. The RDA of vitamin C is only 65 mg/day and of vitamin E 30 IU/day. Supplements of vitamin C at 1,000–3,000 mg/day and of d-a-tocopherol (natural vitamin E) at 400 IU/day are needed to attain significant photoprotection—equivalent to 100 oranges and 44 tablespoons of sunflower oil respectively, per day. For optimal protection against sunburn, tanning, and skin precancers and cancers, vitamin C must be taken for at least 5 days and vitamin E for at least 3–4 weeks before exposure and continued daily, especially during summer months.

The challenge is to create stable formulations that give effective transcutaneous absorption of the active form, as discussed in detail in the following sections. This application over time allows repair and actually acts to reverse prior Vitamin C (l-ascorbic acid) is the body's major aqueous-phase antioxidant and is vital for life. Humans and other primates. Even minimal UV exposure of 1.6 minimal erythema dose (MED) decreases the level of vitamin C to 70% of the normal level, and exposure to 10 MED decreases the vitamin C to only 54%. Exposure to ozone at a dose of 10 parts per million in city pollution decreases the level of epidermal vitamin C by 55%.

Active l-ascorbic acid is such an excellent antioxidant that it is inherently unstable, turning brown as it is oxidized to dihydroascorbic acid when exposed to air. To achieve photoprotection and other benefits to the skin with topical vitamin C, the formulation must contain l-ascorbic acid in a high enough concentration (at least 10%), be stable, and be at an acidic pH—less than the pKa of vitamin C. Topical absorption has been proven by radioactive labeling studies in pigs. Concentrations of 5%, 10%, 15%, 20%, or 25% vitamin C were tested: 20% resulted in the highest skin levels, with maximized concentration in the skin after 3 days of once-daily application. Vitamin C does not absorb light in the UV spectrum, so vitamin C is itself not a sunscreen. However, as an antioxidant vitamin C deactivates UV-induced free radicals and decreases UVB erythema by 52% Laser resurfacing causes redness for at least 3–4 months after treatment. With vitamin C applied before and after laser surgery, this redness of inflammation was markedly decreased afterward, and healing took

only 2 months. The mechanism of this anti-inflammatory action has been researched in vitro with human cells in vitamin C-enriched media.
Newborn fibroblasts and elderly fibroblasts (obtained from biopsies of individuals >90 years old) were grown in vitro with and without 10% l-ascorbic acid added to the culture medium. Fibroblast proliferation rate and synthesis of collagen per cell were measured. Newborn fibroblasts synthesize a larger percentage of collagen than elderly cells; but, again, when elderly cells were exposed to vitamin C in vitro, they produced more collagen than the normal, newborn fibroblasts. In contrast to the increased synthesis of collagen, other in vitro studies suggested that vitamin C may inhibit elastin biosynthesis by fibroblasts. This might be advantageous in reducing the solar elastosis due to photodamage. Postmenopausal women who applied 5% vitamin C to one arm and half of the neck with placebo to the other side showed an increase in mRNA of collagens I and III. Tissue levels of the inhibitor of metalloproteinase-1 (MMP-1) were also increased, thus decreasing UV-induced collagen breakdown. However, mRNA levels of elastin, fibrillin, and tissue inhibitor of MMP-2 remained unchanged. Histology showed elastic tissue repair. Other studies demonstrated a decrease in the crepey, laxity of forearm skin with restoration of a younger skinfold pattern after 6 months of once-daily treatment with 15% vitamin C.
The periorbital wrinkles on the right were markedly decreased by treatment with vitamin C treatment with 15% topical vitamin C on the right and placebo cream on the left, the right periorbital wrinkles were clearly reduced, and the skin acquired a healthy, more youthful glow. With these lipids, vitamin C helps the natural moisturization of the skin as it enhances the protective barrier function.

**4.3 Vitamin E:** It is the most important lipid-soluble, membrane-bound antioxidant in the body. As a free-radical quencher, vitamin E deactivates these aggressive radicals and terminates damaging chain reactions, protecting primarily the fatty components of cell membranes. Vitamin E is synthesized only in plants, so all animals require a nutritional source. The highest levels in the body are in fatty tissue where vitamin E is stored. Vitamin E is delivered to the skin by sebum. As the stratum corneum is the outermost defense of the body, this layer is the first to absorb the oxidative stress of sunlight and pollution.
The lipophilic structure of vitamin E enhances absorption and makes it cosmetically attractive for application as a moisturizer.
Several forms of vitamin E exist in natural dietary sources. The form found in mammalian tissues with by far the greatest biological activity is pure, nonesterified RRR-a-tocopherol. Humans use predominantly a-tocopherol because a specific a-tocopherol transfer protein selectively transfers a-tocopherol into lipoproteiea. The other natural forms are b, g, and d, which contain only one or two methyl groups on the 6-chromal ring. Relative to a form, the b, g, and d RRR-tocopherols give only 42%, 72%, and 40%, respectively, of the protection against post-UV edema. Synthetic vitamin E is “dl” or “all-*rac*,” a mixture of eight stereoisomers.
The skin has only a limited ability to cleave the esterified forms of vitamin E to the active free tocopherol form, so the antioxidant potential of the esters is minimal. The all-*rac* form of vitamin E has been reported to cause allergic contact dermatitis and erythema multiforme when applied topically. No such adverse reaction has been reported with d-a-tocopherol. Even various forms of topical vitamin E that are less metabolically potent when applied topically than the nonesterified d-a-tocopherol have demonstrated protection from the acute UV-induced damage of inflammation and hyperpigmentation (tanning) as well as protection from the chronic UV-induced damage of actinic keratosis and skin cancer. In one

mouse model, topical a-tocopheryl succinate and a-tocopheryl acetate not only failed to inhibit UVB-induced immunosuppression and carcinogenesis but appeared to enhance carcinogenesis. In a 44-week mouse study, both d-a-tocopherol and d-a-tocopheryl succinate were proven effective in protecting against all acute and chronic UV-induced damage, with d-a-tocopherol most effective for all parameters.
Clinically, vitamin E reverses photoaging dramatically, decreasing unattractive wrinkles and solar lentigos. Histological improvement in all parameters of photoaging was noted, with a marked decrease in hyperkeratosis and epidermal hypertrophy, repair of damaged dermal collagen and elastin, and clearing of dermal inflammation after treatment with retinoic acid or with d-a-tocopherol.

**4.4 Vitamin C with Vitamin E:** Vitamins C and E act synergistically in cells to provide antioxidant protection: Vitamin E is located in the cellular membranes. With a lower redox potential, vitamin C can reduce the oxidized vitamin E, thereby regenerating vitamin E activity and eliminating the need for nutritional replacement. Formulating l-ascorbic acid (15%) with a-tocopherol (1%) was found to give fourfold protection against UV-induced erythema in porcine Skin. A decrease in the number of damaged “sunburn cells” was seen histologically as was a decrease in thiamine dimer formation compared to twofold protection with either vitamin alone. Mixing hydrophilic vitamin C with lipophilic vitamin E has the additional advantage of stabilizing each.

**4.5 Vitamin C with Vitamin E and Ferulic Acid:** Ferulic acid is found ubiquitously and at high concentrations in plants, where it crosslinks polysaccharides and proteins during lignin cell wall synthesis. Ferulic acid is a potent antioxidant, so it further protects membranes from lipid peroxidation and neutralizes alkoxyl and peroxyl radicals. It has also been shown to interact synergistically with ascorbic acid. Ferulic acid was found to provide stability of more than 90% for l-ascorbic acid and 100% for a-tocopherol. Addition of ferulic acid (optimally 0.5%) to the formulation of vitamin C (15%) + vitamin E (1%) doubled the photoprotection against solar-simulated irradiation of skin from fourfold to approximately eightfold, as measured by both erythema and a decrease in sunburn cell formation.
Enhanced photoprotection was further demonstrated immunohistochemically by inhibition of UV-induced formation of thymine dimer mutations and of UV-induced evaluation by a real-time polymerase chain reaction demonstrated suppression of UV-induced cytokine mRNA formation. The mice were treated once daily (5 days/week) for 2 weeks prior to exposure to UVB and throughout the experiment of 35 weeks. One group of 15 mice—vitamin C (15%) + vitamin E (1%) + ferulic aid (0.5%)—were treated with vehicle serum and the other with antioxidant serum. Amazingly, only one tumor was seen in the antioxidant-treated group, whereas the vehicle-treated group had 195 tumors. 40 weeks of observation the group of 15 vehicle-treated mice had 67 tumors cumulatively and the 15 d-a-tocopherol-treated mice had 36 tumors. With vitamins C and E in microsomes or in microemulsions are equally impressive in protecting against UV-induced sunburn, tanning, and skin cancer.

- Dietary carotenoid bioavailability is rather low in comparison to other macronutrients.
- Co-consumption of dietary fat is essential for carotenoid bioavailability.
- Fruits and vegetables contain bioactive agents, including carotenoids that protect not only plants but also humans against solar ultraviolet (UV) radiation damage.
- Systemic photoprotection via a dietary supply of carotenoids may contribute significantly to skin health.

• The knowledge acquired so far about the role of dietary carotenoids in photoprotection is still in its infancy and needs to be further investigated.

# Chapter 5. Carotenoids and Skin

**(Sagar K. Thakkar, Angus M. Moodycliffe, and Myriam Richelle)**

**5.1 Introduction:** In mammals, the skin is the largest organ system. Collectively and individually, all layers of the skin work toward the primary goal of protecting internal tissues and vital organs from day-to-day environmental challenges.

The synthesis and storage of lipophilic molecules such as vitamin D are essential functions of the skin. On a daily basis, skin is exposed to various chemical and physical agents that may harm its integrity and lead to a variety of skin pathologies including skin cancer. Visible signs of premature skin aging are attributed to the repeated exposure of the skin to environmental insults, including solar ultraviolet (UV) radiation, leading to the generation of oxidative free-radical molecules, which can damage cellular lipids, proteins, and DNA, — thereby influencing cell survival or death. For centuries, humans have knowingly or unknowingly been protecting their skin against the detrimental effects of solar UV exposure. Deficiencies of vitamin A, vitamin C, riboflavin, niacin, pyridoxine, vitamin E, zinc, selenium, and certain fatty acids have been shown to cause skin anomalies.

**5.2 Dietary Carotenoids:** Carotenoids comprise a family of highly lipophilic pigments that are synthesized by all photosynthetic organisms and some nonphotosynthetic micro-organisms, but not animals. Carotenoids have a polyisoprenoid structure with a long chain of conjugated double bonds and nearly bilateral symmetry around the central double bond. Generally the *trans* isomer is the predominant form in nature, but exposure to light, heat, and acid during the processing of plant foods after harvest and cooking may induce isomerization. Carotenoids are divided into two major classes: carotenes and xanthophylls. Carotenes, also known as hydrocarbon carotenoids, contain only hydrogen and carbon atoms. The most common dietary carotenes include the acyclic lycopene and its biosynthetic downstream cyclic products b-carotene and a-carotene. Xanthophylls are also known as oxycarotenoids due to the presence of at least one oxygen molecule in the structure. Xanthophylls are more hydrophilic than carotenes. Orange fruits and vegetables contain b-carotene, a-carotene, and b-cryptoxanthin, which are the three most common provitamin A carotenoids. Structures of the most abundant dietary carotenoids collard greens which contain carotenoids and are excellent sources of b-carotene, lutein, and zeaxanthin, have their colors masked by the presence of chlorophyll.
Red fruits and vegetables are rich sources of lycopene. The main purpose of the existence of these pigments in plants is to preserve the photosynthetic complex by protecting it against damage by solar UV.  Over the last few decades, more than 600 carotenoids found in nature have been well characterized. However, humans consume only about 50 carotenoids from various sources in their diet, and approximately half of them are found in human plasma. It is evident that the bioavailability of carotenoids in plasma must precede its distribution to hepatic and extrahepatic tissues including skin.

**5.3 Carotenoids Bioavailability and Biodistribution to Skin**
***5.3.1 Carotenoids Bioavailabilit:*** Bioavailability of carotenoids is defined as a transfer of dietary carotenoids and their metabolites to the lymphatic or portal circulation for

distribution to hepatic and extrahepatic tissues for biological functioning, metabolism, or storage. Uptake by intestinal mucosal cells in itself cannot be categorized as bioavailability because mucosal cells may be sloughed off into the lumen before carotenoids or their metabolites can cross the basolateral surface.
Absorption of carotenoids from a meal requires several processes, including (1) release of carotenoids from a food matrix, (2) incorporation into lipid droplets, (3) transfer to mixed bile salt micelles in the lumen of the small intestine, (4) uptake of carotenoid molecules across the apical surface of intestinal mucosal cells from bile salt micelles, (5) incorporation of carotenoids and their metabolites into chylomicrons, and (6) efflux of chylomicrons across the basolateral membrane into the lymphatic circulation.
The presence of dietary lipid stimulates bile flow from the gallbladder, which facilitates emulsification of lipid droplets and other lipophilic molecules in the small intestine. Carotenoid uptake into intestinal epithelial cells has been assumed to occur carotenoids, *Vit A* Vitamin A, *BCO1* b-carotene 15,15¢-monooxygenase 1; *SRB-I* scavenger receptor type B class I passively. Several apical membrane transporters, including Nieman-Pick C1-Like 1 (NPC1L1), scavenger receptor type B class I (SRB-I), and ATP binding cassette transporter subfamily A (ABCA1), have been hypothesized to participate directly or indirectly in the transport. Among the above-indicated transporters, SRB-I may be of primary interest, yet its role in uptake of carotenoids remains ambiguous. Provitamin A carotenoids are partly converted by the central cleavage enzyme b-carotene 15.15¢-monooxygenase 1 (BCO1) to vitamin A primarily in the form of retinyl esters in the intestinal mucosal cells. Carotenoids appear to selectively accumulate in tissues expressing a high density of LDL receptors.

***5.3.2 Factors Influencing Carotenoid Bioavailability:*** The bioavailability of carotenoids is dependent on physiochemical properties of the carotenoid type of processing of raw food, presence or absence of compounds that promote or inhibit their absorption, pathophysiological status of the gut.

**5.3.2.1 Physicochemical Properties of Carotenoids:** Carotenoid speciation also appears to affect bioavailability. Oxycarotenoids are more hydrophilic owing to the presence of one or more polar functional groups and are located on the surface of oil droplets. Hydrocarbon carotenoids are more hydrophobic in comparison and are embedded in the core acid esters. Small intestinal simulations have demonstrated that hydrolytic cleavage of ester bond by carboxyl ester lipase, a broad-specificity pancreatic enzyme, precedes the preferential uptake of the nonesterified form of carotenoids by small intestinal cells.

**5.3.2.2 Food Matrix and Processing:** Carotenoids may be bound to proteins in the food matrix. Food processing alters the matrix by disrupting the cell wall and membrane-bound organelles, loosening the linkages between carotenoids and proteins or fiber, partially dissolving the crystalline carotenoids in oil, and increasing its surface area. Lycopene concentrations in human serum increased only when tomatoes were subjected to 1 h of boiling in the presence of oil. Processing of plant foods also induces isomerization of carotenoids, thus increasing the levels of *cis* isomers. Although *cis* isomers of b-carotene are also precursors of vitamin A, isomerization of a provitamin a carotenoid reduces the retinol activity equivalence (RAE) as compared to their transanalogues.

**5.3.2.3 Interactions with Other Dietary Components:** Consumption of carotenoids in our diet is usually associated with other macronutrients and micronutrients in the diet, which

may act as promoters or inhibitors of carotenoid bioavailability. The association of carotenoids with dietary proteins has been shown to decrease their absorption in ferrets. Citrus pectin and wheat bran are likely to bind to bile salts and decrease the formation of micelles and ultimately the absorption of carotenoids. Carotenoid absorption from a salad meal was enhanced by addition of avocado or avocado oil.
The synthesis and secretion of chylomicrons by enterocytes is increased by dietary fat. High doses of pure b-carotene and lutein have been reported to antagonize the absorption of one another. The mechanisms for preabsorptive interactions among carotenoids are not well understood. Proposed interactions have been suggested to occur during their incorporation into bile salt micelles, uptake by intestinal epithelial cells, and incorporation into chylomicrons within enterocytes.

**5.3.2.4 Health Status of the Host:** The digestive health status and overall health status of an individual, also influences the absorption of carotenoids. Increased gastric pH appears to decrease b-carotene absorption. Gastrointestinal conditions that cause fat malabsorption may also lead to decreased carotenoid absorption. Parasitemia in general has been negatively associated with plasma concentrations of carotenoids. The presence of parasites in the intestine impaired carotenoid absorption or utilization.

***5.3.3 Carotenoids Biodistribution to Skin:*** Once carotenoids clear the intestinal epithelial barrier and reach the systemic circulation via lymphatics, deposition occurs in hepatic and extrahepatic tissues including skin. Carotenoids stored in hepatic tissues can also be mobilized for distribution to extrahepatic tissues in times of need. Skin derived from subjects undergoing abdominoplasty suggests that apolar carotenoids and their isomers constitute approximately 70% of the total pool. Although stereoisomers of carotenoids mainly exist as the all-*trans* configuration in nature, *cis* isomers are also present in skin. Recent data from our laboratory suggests that isomerization begins in small-intestinal epithelial cells and may even occur in other tissues. Xanthophylls esters are also present in skin as mono- or di-fatty acid esters in picomolar ranges.
Carotenoids exhibit a concentration gradient in the layers of skin with a higher amount in the dermis and lower levels in the stratum corneum. Consumption of diets or supplementation containing more than 30 mg carotenoids per day for more than 4 weeks may result in yellowish discoloration of the skin, called carotenodermia.

**5.4 Techniques to Analyze Carotenoids in Skin:** Specimens from which are then analysed by high performance liquid chromatography (HPLC) for the presence of carotenoids. Chemical analyses by HPLC for carotenoids in skin biopsies from cadavers, abdominoplasty subjects, and a few healthy subjects have been reported. The complexity of the matrix of the skin biopsies makes extraction of carotenoids for HPLC analysis a challenging task. Researchers have started to focus on the use of Raman spectroscopy for noninvasively assessing the concentration of carotenoids in skin. Two laboratories reported a good correlation between the amount of carotenoids in plasma compared to that measured by Raman spectroscopy in skin.

**5.5 Dietary Carotenoids Protect Skin Against Some of the Damaging Effects of UV Exposure**
***5.5.1 Damaging Effects of UV on Skin:*** Ultraviolet radiation present in sunlight represents one of the most important environmental hazardous physical agents that the skin encounters on a daily basis throughout a person's lifetime. Depending on the amount and

form of the UV radiation and the skin type of the individual exposed, UV irradiation may cause tissue injury and cutaneous inflammation signifying sunburn. Chronic exposure to solar UV is considered the major etiological factor for the development of nonmelanoma skin cancer, which occurs primarily on sun-exposed areas of the body. The underlying cause of sunburn is direct and indirect damage to specific cellular targets. Damage to DNA, the activation of several inflammatory pathways, and the release of inflammatory mediators by keratinocytes are thought to trigger this reaction, ultimately leading to vasodilation and edema.
It is now appreciated that there is a linkage between a history of repeated, severe sunburn and an increased risk for melanoma and nonmelanoma skin cancer. Various strategies are followed for the protection of skin against UV-dependent damage. Limiting sun exposure, protective clothing, and the use of sunscreens are generally recommended. Systemic photoprotection through endogenous supply of nutritional bioactive agents such as carotenoids also provides important protection of the skin against the damaging effects of UV irradiation.

***5.5.2 Carotenoids are Scavengers of UV-Induced Reactive Oxygen Species that Damage DNA:*** Upon UV exposure, reactive oxygen species (ROS), which are constantly generated in skin, may be rapidly neutralized by nonenzymatic and enzymatic antioxidant substances. In consequence, cellular macromolecules are protected from oxidation and a pro-oxidant/antioxidant balance is maintained, resulting in cell and tissue stabilization. If the antioxidant defense is exhausted, these ROS oxidize lipids, proteins, or DNA, leading to the formation of oxidized products such as lipid hydroperoxides, protein carbonyls, or 8-hydroxyguanosine, respectively and result in cell damage.
Carotenoids are efficiently distributed to skin and might participate to the antioxidant capacity of the skin. Supplementation with lutein and zeaxanthin significantly decreases skin lipid peroxidation, as measured by malondialdehyde levels.

***5.5.3 Dietary Carotenoids Reduce Sunburn Development:*** The sensitivity of an individual to erythematogenic UV exposure is determined by two methods: (1) the minimum erythema dose (MED), defined as the threshold dose required to cause perceptible reddening of the skin 24 h after exposure or (2) the change of skin color assessed by chromametry. With the latter technique, erythema is assessed by a change of chromametry a-values after and before irradiation. Decreasing Da values in comparison to those at week 0 (set to 100%) reflects protection against UV-induced erythema. In human intervention studies, the photoprotective effect of a nutritional intervention refers to UV-induced erythema as an early observable immediate response. It is measured either as increased MED or as a reduction of erythema intensity after UV exposure.
The photoprotective effect is observed only in studies providing a daily dose of 20 mg for a minimum of 10 weeks of supplementation. Daily intake of 10 mg of lycopene was provided from three sources: a synthetic lycopene, a tomato extract (Lyc-o-Mato), or a drink containing solubilized Lyc-o-Mato. The synthetic lycopene showed a trend but it did not reach statistical significance. Supplements derived from tomato-based products contain a number of other constituents, including other carotenoids such as phytofluene and phytoene, which are precursors of lycopene in the biosynthetic pathway. These compounds may well contribute to the photoprotective effects as they absorb in the UV range.
Erythema development was diminished in subjects whose diets were supplemented with b-

carotene (24 mg/day) or a carotenoid mixture consisting of b-carotene, lutein, and lycopene (8 mg each/day) for 12 weeks.

***5.5.4 Dietary Carotenoids and Skin Cancer:*** Basal cell carcinoma (BCC) and squamous cell carcinoma (SCC) are the most commonly occurring skin cancers in white populations. According to estimates of the National Cancer Institute, 40–50% of Americans who live to age 65 develop skin cancer at least once, and the risk of developing additional tumors is high. In animal studies, b-carotene protects against skin cancer induced by chemicals and UV radiation. Based on epidemiological studies, no association was found between dietary carotenoids and BCC or SCC. B-carotene supplementation failed to decrease the risk of nonmelanoma skin cancer among men with low baseline plasma b-carotene. B-carotene (30 mg/day) had no effect on the incidence of solar keratosis, a premalignant skin cancer, in a randomized controlled study in 1,600 participants. The daily use of b-carotene (30 mg/day, *n* = 1621) for 4.5 years did not reduce the incidence of BSC or SCC.

**5.6 Future Perspectives on Systemic Photoprotection by Carotenoids:** Sunburn is just one phenomenon related to exposure to UV radiation. Erythema formation can be readily quantified and is being used successfully as a noninvasive parameter for assessing the biological response to UV exposure. Whether erythema formation is the most suitable surrogate endpoint for long-term degenerative diseases such as skin cancer, photodermatoses, or photoaging is not clear and needs to be scrutinized in further work. It is not clear whether endogenous carotenoids can positively affect skin health by also interfering with UV-induced pathways that lead to DNA mutations, immune suppression, and skin cancer development. Metabolites or oxidation products of parent carotenoids such as retinoids or apocarotenals are the ultimate active agents. The mechanisms that lead to the incorporation of micronutrients such as carotenoids into different parts of the skin are also not yet known. The fact that there are large disparities between skin areas in terms of embedded micronutrients.
The concept of endogenous dietary skin photoprotection is that it provides a systemic maintenance level at sensitive dermal and epidermal target layers—beyond those reached by topical and temporary coverage through the use of sunscreen. An increased lifelong overall systemic protection via dietary supply may contribute significantly to skin health and complement the use of sunscreens in protecting the skin against the damaging effects of solar UV exposure.

# Chapter 6. Antioxidants and Skin

**Juergen Lademann, Maxim E. Darvin, and Ulrike Heinrich**

**6.1 Introduction** Free radicals are constantly formed in human skin as a result of cellular metabolism, environmental conditions, inflammation and sun irradiation. They play an important role in signaling processes inside the cell and between cellular systems. The human body also utilizes free radicals to destroy viruses and bacteria. Although in the past comprehensive studies elucidated the radical production in human skin by ultraviolet (UV) light A high concentration of free radicals can cause oxidative cell damage, skin aging, and cancer. With the antioxidative network, the human body has developed a protective system against the destructive action of free radicals. In recent years it has been established that carotenoids function as antioxidants in lipophilic systems.
Fruit and vegetables contain high amounts of antioxidants. Their efficacy can be improved when they are combined with carotenoids and vitamins, such as a-tocopherol or vitamin C. The interaction of the antioxidants with the free radicals. Antioxidants form protective chains in the human skin and protect each other against the destructive action of free radicals. This does not apply to molecules such as DNA alone but also to cellular structures such as the elastic fibers in elastin and collagen. Enhancement of the antioxidative network of human skin is recommended. The interaction between antioxidants and free radicals has been a subject of intensive research for decades, this topic has recently received fresh impetus by the development of methods that permit antioxidants, specifically carotenoids, to be detected in the human skin noninvasively and online. The methods mainly applied here are reflection spectroscopy and resonance Raman spectroscopy. In the past, such investigations had been possible only by performing biopsies or obtaining blood samples, which subsequently had to be subjected to time-consuming and expensive conventional methods, such as high-pressure liquid chromatography (HPLC).

**6.2 Positive and Negative Effects of Antioxidants:** In ancient
times it had been known that intake of fruit and vegetables is important for staying healthy, but it had not been possible to demonstrate that healthy food positively influences the course of disease as well. Based on this positive experience, intensive efforts had been undertaken to extract antioxidants from fruit and vegetables or to produce them synthetically.
The therapy of cancer patients, was supported by treatment with antioxidants. Some studies revealed that the patients of the verum group died earlier than those of the placebo group as a consequence of carotenoid uptake. The results of these studies have severely damaged the reputation and importance of supportive treatment with antioxidants, specifically with b-carotene. The situation is quite different when it comes to cosmetic treatments. Most cosmetic products, therefore, contain significantly lower concentrations of antioxidants than products for supportive treatment with antioxidants. Excessive uptake of antioxidants is prevented by a natural saturation effect of the human organism. For useful supplementation with antioxidants one needs to pay attention to good compatibility, high bioavailability, and synergies. Moreover, the intake must be adjusted to daily needs.

**6.3 Polyphenols: Cocoa and Green Tea:** Several antioxidants given systemically or topically as enriched food and supplements have been demonstrated to provide photoprotection. They include flavanols, carotenoids, tocopherols, and vitamin C. Flavonoids comprise a group of secondary plant constituents widespread in nature and available from dietary sources such as cocoa, green tea, soy, berries, or other fruit. The alleged effects have been linked to the antioxidant properties of flavonoids, but they also exhibit other biological activities.

In vitro and animal studies have provided evidence that tea flavanols, when applied orally or topically, ameliorate adverse skin reactions following UV exposure, including skin damage, erythema, and lipid peroxidation  Topical application of green tea polyphenols to human skin have inhibited the UVB-induced erythema response and decreased formation of cyclobutane pyrimidine dimers in skin, found in both epidermis and dermis. Pretreatment of skin with green tea extracts led to a lower number of sunburned cells after exposure to solar-simulated radiation with a minimum erythema dose (MED) of 2 and protected epidermal Langerhans cells from UV damage.

Photoprotective effects and parameters of skin condition were measured. UV-induced erythema following exposure of selected skin areas to 1.25 MED was significantly decreased in the high-polyphenol group after 6 and 12 weeks of treatment. Ingestion of high-polyphenol cacao powder led to an increase in the blood flow of cutaneous and subcutaneous tissues. Supplementation with high-polyphenol cacao powder results in a significant increase in skin density and thickness.  Cacao polyphenols further affect cosmetically relevant parameters of skin surface and hydration.

The major flavanol monomer in both drinks was epicatechin: 61 mg/dl in the high-flavanol product and 6.6 mg/dl in the low flavanol product. Dermal blood flow and oxygen saturation were examined by laser Doppler flowmetry and spectroscopically at 1 mm skin depth at 0, 1, 2, 4, and 6 h. At the same time points, the plasma levels of total epicatechin  were measured by means of HPLC. Subsequent to the intake of high-flavanol cocoa, dermal blood flow was significantly increased (by 1.7-fold) at 2 h and oxygen saturation was elevated 1.8-fold. No statistically significant changes were found with intake of low-flavanol cocoa. Maximum plasma levels of total epicatechin were observed 1 h after ingestion of the high-flavanol cocoa drink, 11.6 ± 7.4 nmol/l at baseline and 62.9 ± 35.8 nmol/l at 1 h.

Photoprotection parameters, skin structure, and function were measured at baseline (weeks 0, 6, and 12. Following exposure of the skin areas to 1.25 MED of radiation from a solar simulator, UV-induced erythema was significantly decreased in the intervention group.

**6.4 Antioxidants Protect Human Skin from Premature Aging**

Various studies in which hundreds of volunteers had been investigated with the noninvasive Raman spectrometer disclosed that those volunteers who exhibited a high carotenoid level looked younger than they actually were an optical skin profile. The measured area was 1 × 2 cm. Based on the skin surface measurements, skin roughness was measured. The results showed a clear correlation between the antioxidant level in the human skin and skin roughness, which is determined by the depth and density of the furrows and wrinkles. The correlation factor R2 was 0.7.  A high concentration of antioxidants, including carotenoids, in human skin can efficiently neutralize free radicals before they can develop their harmful effect.  The increase in skin density and thickness is caused by a fortifying effect on the collagen structure of the skin, which in turn has a positive effect on its elasticity and vigor. The ingestion of a nutritional supplement with various natural carotenoids, combined with selenium and vitamin E, over a period of 12 weeks resulted in a significant increase

in skin density and thickness.

• Several minerals such as zinc, iron, and copper have an impact on the skin.
• Adequate intake of minerals positively influences the overall appearance of the skin.
• Deficiency of certain minerals results in certain manifestations in skin, hair, and nails and it is associated with skin diseases.
• Dermatological effects are observed when the minerals are either orally supplemented or topically applied.
• Mineral supplements should be taken with care because excessive intake of minerals can lead to intoxication.

**7.1 Introduction:** Although only approximately 4% of the human body mass consists of minerals, they play a significant role. Minerals are involved in maintenance of electrical neutrality, osmotic pressure, solubility, buffer systems, nervous conditions, and metabolism via constituents of enzymes; they are also components of bones and teeth. Because these substances take an active part in the metabolism, humans need a regular supply of these essential nutrients.

# Chapter 7. Minerals and the Skin

**Petra Winkler**

**7.2 Zinc:** With approximately 1.5–2.5 g, zinc is the second most abundant trace element in the human body. It can be found in all living cells and body fluids. Amounts of 6–20% of the body stores have been described to be found in the skin, mainly as metalloenzymes, where zinc is needed because of the highly proliferative nature of the tissue. A continuous zinc supply is necessary as the body is not able to store the mineral and make use of it in case of an impending deficiency.

### *7.2.1 Functions*

• Cofactor in more than 200 enzymes and metalloproteins: e.g., RNA polymerase, alcohol dehydrogenase, various enzymes of DNA synthesis.
• Involved in the metabolism of numerous hormones and the metabolism of proteins, fat, and carbohydrates.
• Cell growth and differentiation.
• Stabilization of the structures of DNA, RNA, and membranes.
• Involvement in collagen synthesis and thereby essential for the construction and degradation of connective tissue, ligaments, and tendons.
• Production and regulation of cellular and humoral immune reactions.
• Cytoprotection against organic toxins, heavy metals, radiation, and endotoxins from pathogen bacteria.

### *7.2.2 Deficiency*

Although plasma zinc concentration is the most reliable measure of the total bodily storage, it is not always an accurate reflection. In fact, there is no test that can make this determination with accuracy. The range of normal values is broad (90 ± 20 mg/dl). Zinc levels in hair also vary greatly and can be elevated falsely by traces of tightly bound zinc. Zinc deficiency due to malnutrition was discovered in rural areas of the Middle East, where diets consist mainly of cereals, which contain phytates that chelate zinc and prevent absorption, and a low intake of meat. It is found independent of socioeconomic status and can also occur in those who believe they are eating healthy diets. Moreover, delayed wound healing and higher susceptibility to infections as a consequence of an impaired immune system may occur. As the zinc concentration begins to fall, an acral dermatitis begins to develop, which starts off as dry scaly, eczematous plaques in perioral, perineal, and acral areas. The rash is characteristically symmetrical and is accompanied by progressive alopecia, including loss of eyelashes in some cases. Felons can appear on the hands and feet and erythema-like dermatitis on the palms. Almost all cases of zinc deficiency can be cured after 2–3 weeks of taking oral zinc supplements.

***7.2.3 Impact on the Skin:*** Many skin disorders have been associated and treated with zinc and oral zinc therapy has been shown to be effective in treating inflammatory conditions.

**7.2.3.1 Zinc and Skin Appearance:** Zinc has been shown to protect the skin cell membrane. A negative correlation was observed between zinc intake and skin evenness as well as the hydration of facial areas.

**7.2.3.2 Zinc in the Healing Wound:** Orally supplemented or topically applied zinc in the form of zinc oxide or calamine has been used in the treatment of skin wounds. It is still unclear how much additional zinc a wound needs for healing process to be enhanced or to what extent topical applied zinc is absorbed. Not only is zinc a constituent of enzymes that play a central role in reconstruction of a wound's matrix but effect of zinc was on these integrins, affecting cellular mobility during the proliferative phase of wound healing.

**7.2.3.3 Zinc and Acne:** As patients with acne were reported to have low levels of serum zinc acne itself seems to be a symptom of zinc deficiency. It may be due to overproduction of male hormones, which was observed in cases of zinc deficiency. Zinc works to clear the skin by reducing oil production and may be effective in controlling the formation of acne lesions or help those already present to heal quicker by its anti-inflammatory effect.

**7.2.3.4 Zinc and Psoriasis:** Zinc involvement in the pathogenesis of psoriasis is not clear but it seems that oral zinc supplementation has no effect on the skin manifestation of psoriasis.

**7.2.3.5 Zinc and Hair Loss:** There is a widespread belief that zinc deficiency can cause hair loss in humans, which is seen only with severe zinc deficiency.

**7.2.3.6 Zinc and Herpes Simplex Infections:** Oral zinc treatment may be considered as part of overall immune-enhancing therapy for patients with recurrent herpes simplex infections.

**7.2.3.7 Topically Applied Zinc:** Diaper Dermatitis Zinc oxide, often combined with castor oil, has a special place in the treatment of diaper dermatitis. The anti-inflammatory and antimicrobial activity of zinc oxide can help alleviate the symptoms. Dandruff Zinc pyrithione is an antifungal agent that is effective as an antidandruff agent and for seborrheic dermatitis. Zinc itself may also have an impact beyond the antifungal action. Its biologically active metabolite including stimulation of sebaceous gland activity and could be an additional benefit of the use of zinc-containing antidandruff treatments. With regard to acne, topically applied zinc has been proven to have positive effects due to its antimicrobial and anti-inflammatory effects as well as reduction of hyperproliferation and high sebum production. Zinc is bacteriostatic against *Propionibacterium acnes*, one of the bacteria involved in causing acne. Zinc inhibits chemotaxis and may also directly inhibit bacterial lipases from converting sebum triglycerides to fatty acids. Comparison of the zinc containing antibiotic treatment with one without zinc demonstrated the zinc-containing product to have greater lesion reduction efficacy. Although used zinc sulfate, zinc monoglycerolate or zinc oxide-glycine may also be beneficial. Zinc oxide, however, seems not to be effective because it does not release a sufficient amount of zinc ions to exert an antiviral effect.

**7.3 Iron:** It is the most abundant trace element in the body. The adult human body contains approximately 2–5 g of iron, about 60% of which is bound to hemoglobin, 25% to ferritin and hemosiderin, and about 15% to myoglobin and enzymes.

***7.3.1 Functions***

• Transport of oxygen in the form of hemoglobin in erythrocytes.
• Storage of oxygen as myoglobin in muscle cells.
• Generation of energy in mitochondrial cytochromes.
• Constituent in groups of enzymes (e.g., cytochrome P450 system).
• Production of neurotransmitters in the brain.
• Factor in collagen synthesis.
• Hydroxylation of proline and lysine.

***7.3.2 Deficiency:*** Iron deficiency represents a public health problem and frequency ranges from <10% to high 70% among various ethnic and socioeconomic groups. Symptoms caused by iron deficiency include fatigue, lack of energy, loss of appetite and increased susceptibility to infections. From dermatological perspective iron-deficiency state can lead to dry skin, brittle hair, hair loss, rutted fingernails, inflammation of oral mucosa and tongue, pruritus, chronically sustained inflammation, dermatitis herpetiformis, photodermatitis and changes in the overall appearance such as pallor and blue sclerae.

***7.3.3 Impact on the Skin***

**7.3.3.1 Pruritus:** Causes of puritus is iron deficiency with or without accompanying anemia. Men and women who have low serum haemoglobin and ferritin concentrations suffer significantly more often from pruritus than people with normal iron values.

**7.3.3.2 Iron and Hair:** In the presence of iron deficiency, the hair appears lusterless, dry, brittle, and spliced. It may be due to impaired keratin production. The main cause appears to be depleted iron stores accompanied by suboptimal intake of the essential amino acid l-lysine. The role of iron as an etiological factor in diffuse hair loss in nonanemic women with an iron deficiency was demonstrated. The percentage of hair in the telogen phase in women with CTE after supplementation with 72 mg iron and 1.5 g l-lysine daily for 6 months. Although the exact level of serum ferritin that should be reached is still not fully resolved, 70 mg/L seems to be the level to achieve. According to a preliminary analysis of data, a daily iron supplement of 24–48 mg seems to be needed by menstruating women.

**7.3.3.3 Iron and Nails:** Brittle and rutted fingernails are often observed with moderate iron deficiency. In advanced cases, the nails have an even, plain, spoon-shaped, convex structure, predominantly, but not exclusively, the index and middle fingers.

**7.4 Copper:** It is with 50–150 mg, third in abundance in the human body after iron and zinc.

***7.4.1 Functions***

• Compartment of several vital metalloenzymes that are involved, for example, in blood clotting, detoxification of free radicals, and oxidation–reduction.
• Involvement in iron metabolism.
• Constituent of tyrosinase, which functions in melanin production.
• Essential for the production of mature collagen and elastin as well as hair maturation

Copper deficiency is rare, and taking copper supplements can be dangerous as the dose for intoxication is relatively low. A tolerable upper intake level of 5 mg/day was derived; but this upper level is not applicable during pregnancy or lactation because of inadequate data relating to this critical life stage.

***7.4.2 Deficiency:*** Copper deficiency can result from long-term parenteral nutrition or chronic malabsorption. Cerebral degeneration and severe arterial disease are the most important systemic manifestations and usually lead to death at 3–4 years of age. Slight copper deficiency leads to anemia with leukopenia and neutropenia, increased sensitivity to oxidative damage, hypercholesterolemia, hypertriglyceridemia, glucose intolerance, impaired immune reaction, poor wound healing, osteoporosis, faintness, and fatigue. Although one might expect changes in skin elasticity given the role of copper in elastin formation, it is not generally observed.

**7.4.2.1 Impact on the Skin**

Topical use of copper–peptide complex can provide facial skin antiaging effects. Copper-containing creams have been found to firm the skin and help restore some elasticity. Evaluations via clinical grading of efficacy parameters, high-resolution digital imaging, and subject self-assessments revealed statistically significant and visible improvement of overall appearance, skin radiance, under-eye bags, under-eye wrinkles, under-eye dark circles, sagging, cheek wrinkles, pigmentation, radiance, fine lines, as well as global lifting and firming when the preparation was applied at least once a day.

**7.5 Selenium:** The total body pool of selenium has been estimated to be 5–15 mg in adults.

***7.5.1 Functions***

- Antioxidant as a constituent of glutathione peroxidase.
- Part of an antioxidant system that protects cell membranes and structural membranes from lipid peroxidation.
- Immunomodulation via regulation of immunoglobulin G (IgG) production and stimulation of leukocyte activity.
- Regulation of thyroid hormone metabolism.

Because of the increase in self-medication with selenium, several cases of acute selenium intoxication occurred because of falsely specified preparations. Intake of 250 mg selenium as a single dose or multiple doses of 27–31 mg resulted in acute toxicity with nausea, vomiting, nail changes, dryness of hair, hair loss, tenderness and swelling of fingertips, fatigue, irritability, and garlicky breath. A tolerable upper intake level of 300 mg/day was derived for adults.

***7.5.2 Deficiencies:*** No specific selenium deficiency condition has been described in humans. Obvious deficiency symptoms occur in Keshan disease, which is related to a combination of coxsackie virus and low selenium uptake, causing inflammation of the heart muscle, leading to cardiomyopathy. Manifestations are recognized in skin and hair in the form of depigmentation, which is also observed with long-term deficiency due to long-term parenteral nutrition.

***7.5.3 Impact on the Skin***

**7.5.3.1 Selenium in Acne:** Because the selenium-dependent glutathione peroxidase enzyme activity is low in acne patients, it has been theorized that selenium would be of value. The combination led to reduced severity of persistent acne and reduced scarring, especially in those with low baseline glutathione peroxidase activity.

**7.5.3.2 Selenium and Psoriasis:** Observations showed low selenium concentration and depressed glutathione peroxidase activity in patients with psoriasis, especially in those who had been suffering from the disease for 3 years or more. Patients who responded to treatment had a significant increase in their plasma selenium level. However, supplementation with inorganic selenomethionine did not lead to clinical improvement.

**7.5.3.3 Topical Application:** Selenium sulfide is an antifungal agent that is effective as an antidandruff agent**.** Topical application of selenomethionine prevents UV-induced erythema and is discussed as providing facial anti-wrinkle effects.

**7.6 Chromium:** It is ubiquitous. Chromium ions are constituents of the glucose tolerance factor that is important for protein, glucose, and lipid metabolism.

***7.6.1 Functions***

• Responsible for production of certain enzymes.
• Influences carbohydrate, lipid, and protein metabolism.
• Effect on insulin action, part of the "glucose tolerance factor".
• Helps to fight acne and reduce infections.

Currently, there is no formal RDA for chromium. Good sources of chromium are meat, liver, eggs, seafood, whole grain products, oat flakes, lettuce, green beans, broccoli, tomatoes, potatoes, prunes, nuts, cacao, and mushrooms. Chromium in food is not easily absorbed. Some research has indicated that almost 90% of the population could be deficient.

***7.6.2 Impact on the Skin:*** Chromium has been shown to reduce skin infection rates and thus may help decrease the symptoms of acne. Moreover, it is estimated that 90% of people suffering from acne also exhibit excessive or unstable blood glucose levels. The human body recognizes the presence of insulin very quickly, and chromium takes part in insulin metabolism.

**7.7 Silicon** is present in biological material as a silanate, an ether (or ester-like) derivative of silicic acid, which may play a role in the structure of glycosaminoglycans and their protein complexes. Despite the fact that silicon is involved in important processes, there is little known about its mode of action or physiological requirements

***7.7.1 Functions***

• Along with calcium, involved with growth and maintenance of strong bones.
• Formation and structure element of connective tissue, ligaments, and tendons.
• Involvement in cell metabolism and division.
• Growth of hair, skin, and fingernails.
• Favors biological moisturization of the dermis and epidermis because of the ability to maintain water bound in tissues.

***7.7.2 Dietary Recommendation:*** Even though measurable responses of humans to variations in dietary silicon intake have been demonstrated, no estimated average requirement or adequate intake has been set because the data available are still insufficient. Good sources of silicon are brown rice, cereal products, leafy green vegetables, root vegetables, seafood, and soybeans because it is made from grains, beer also is a dietary source of silicon.

***7.7.3 Deficiency:*** Signs of silicon deficiency have not been described for humans. The nature of silicon deficiency in animals has led to the speculation that the lack of silicon is involved in several human disorders, including atherosclerosis, osteoarthritis, and hypertension as well as the aging process.

***7.7.4 Impact on the Skin:*** Orthosilicic acid, a bioavailable form of silicon that has been shown to stimulate collagen synthesis in skin fibroblasts and mechanical properties as well as on hair and nail brittleness in women with photodamaged facial skin after supplementation with 10 mg daily for 20 weeks.

• Our cutaneous and intestinal microbiota have a close relationship with skin health, so manipulation of these microbiota may have health benefits for the skin.
• Nevertheless, there are promising data suggesting that probiotics administered by either route may have significant beneficial effects on the skin.
• Orally ingested probiotic bacteria have been widely investigated for a potential role in treating or preventing the common skin disease eczema.
• Current evidence suggests that orally administered probiotic bacteria are not effective for the treatment of established eczema.

**8.1 Introduction:** Approximately 90% of cells in each human are constituents of the large intestinal microbiota, so we are in at least one sense more microbe than human. This intense colonization is no evolutionary accident but provides survival advantages for both microbes and humans.

# Chapter 8. Probiotics and Skin

**Robert J. Boyle, Sampo J. Lahtinen, and Mimi L.K. Tang**

The use of probiotic bacteria to alter the intestinal microenvironment and therefore impact on immune function is one approach to the management of skin disease. Probiotic preparations have largely been administered via the oral route, but more recently topical use has been investigated. The latter approach derives from the importance of our cutaneous microbiota—we typically have 104–106 microbes/cm2 on our skin, including bacteria and fungi—and aims to influence immune cells or epithelial cells in the skin. Although the composition of the skin microbiota in an individual is less stable than the intestinal microbiota, it varies significantly between healthy and unhealthy skin, even within the same individual.

**8.2 Probiotics:** Beneficial microbes in the form of *Streptococcus Thermophilus* and *Lactobacillus bulgaricus* in fermented milk have been ingested by humans for thousands of years in the belief that fermented products have health benefits. Lactic acid bacilli may have beneficial health effects and attributed his own longevity to regular ingestion of beneficial microbes. Probiotics are most commonly defined as "live microorganisms which when administered in adequate amounts confer a health benefit on the host". Some of the best characterized probiotics have also been shown to adhere strongly to intestinal epithelium in both in vitro and in vivo studies. Probiotics must also be resistant to gastric acid digestion and to bile salts to reach the intestine intact, and they should be nonpathogenic. Most probiotics are strains of *Bifidobacterium* or *Lactobacillus* species.
Nonbacterial microorganisms such as yeasts from the genus *Saccharomyces* have also been used as probiotics for many years. Probiotics have been advocated for the prevention and/or treatment of a diverse range of disorders, from acute gastroenteritis to intestinal neoplasia. a meta-analysis of randomized controlled trials (RCTs) has shown that many probiotics are effective in preventing antibiotic-associated diary. They include the yeast *Saccharomyces boulardii,* the bacterium *Lactobacillus acidophilus* in combination with *L. bulgaricus, L. rhamnosus* strain GG.
The treatment of irritable bowel syndrome (IBS) and various types of inflammatory bowel disease (IBD). Studies in Crohn's disease have often failed to show clinical benefit, whereas studies of other forms of IBD, such as ulcerative colitis or pouchitis, have demonstrated potential benefit. Finally, there is good evidence that probiotics added to enteral feeds are able to prevent an intestinal complication of premature birth, at least in infants weighing >1,000 g at birth. Probiotic therapy has also been explored in skin diseases, particularly for the treatment and prevention of eczema.

**8.3 Eczema:** There has been a rapid rise in the prevalence of allergic and autoimmune disorders in recent decades. The intestinal microbiota represents the greatest microbial exposure throughout life, and acquisition of the early intestinal microbiota is the newborn infant's first major microbial challenge. Intestinal microbiota development has been shown to play an important role in immune regulation and the induction and/or maintenance of tolerance to environmental and self-antigens . Manipulation of the intestinal microbiota during infancy may therefore provide an approach to the prevention or treatment of allergic

skin conditions. It is unclear whether probiotics are able to modulate early development of intestinal microbiota when administered early in life.

***8.3.1 Probiotics for Treating Eczema:*** Probiotic bacteria have been quite widely investigated for effects on the treatment of eczema with or without associated food allergy, in infants and children. Eczema have evaluated *Lactobacillus* species either alone or in combination with other probiotic bacteria. Infants and children reported improvement in eczema (SCORAD or symptoms) following treatment with *Lactobacillus rhamnosus* GG (LGG), *Bifidobacterium lactis* Bb-12, or *B. breve* M-16 V. Probiotic treatment resulted in more rapid alleviation of the eczema. The impact of eczema on the family, and topical corticosteroid use were not significantly different between the active and placebo groups. Recent systematic review evaluating the use of probiotics for treating eczema concluded that probiotics do not appear to be an effective treatment, and there is insufficient evidence to support their use for this condition. Significant heterogeneity was noted between studies, and we found evidence that it may be explained by the use of different probiotic strains. *RCT* randomized controlled trial, *CMA* cow's milk allergy, *eHF* extensively hydrolysed formula, *LGG Lactobacillus rhamnosus* GG, *CFU* colony-forming units, *SCORAD* Scoring Atopic Dermatitis, *QoL* quality of life could still be effective. In contrast to our meta-analysis, a recent meta-analysis by Michail et al. reported a significant difference favoring probiotics in reducing the SCORAD score of children with eczema (mean change from baseline was −3.01 with the 95% CI −5.36 to −0.66, $P = 0.01$), and children with moderately severe disease were more likely to benefit. The treatment was also associated with reduced *Staphylococcus aureus* colonization of skin. Future studies evaluating topical probiotics for the management of eczema will be of great interest.

***8.3.2 Probiotics for Preventing Eczema:*** In all, 13 RCTs evaluating various probiotic bacteria used alone or in combination with other. Three of the ten prenatal/postnatal treatments showed no statistically significant reduction in rates of eczema or IgE-associated eczema at age 1 or 2 years. In contrast to the findings for probiotics administered prenatally and postnatally, most of the studies that evaluated postnatal without prenatal treatment with various probiotic bacteria reported no beneficial effects on the development of eczema associated eczema at 12 months. Meta-analysis of these four studies revealed no evidence that postnatal treatment without a prenatal component reduces the risk of eczema.
First, we should note that most of the combined prenatal/postnatal treatments were effective in reducing eczema and/or IgE-associated eczema during the first 1–2 years of life, whereas most of the treatments involving only a postnatal component of therapy failed to reduce the risk for eczema and/or IgE-associated eczema.
Second, administration of a probiotic solely to women from 36 weeks of pregnancy to 3 months postpartum without direct infant supplementation was sufficient to reduce the cumulative incidence and prevalence of eczema at 2 years indicating that direct infant probiotic supplementation during early life may not be an absolute requirement for protective effects.
The greatest protective effects of LGG treatment in their study were seen in breast-fed infants for whom probiotic was administered to their mothers during pregnancy and breastfeeding, without direct administration to the infant until after 3 months of age. Both strains were associated with immunomodulatory effects in cord blood and breast milk, but only one was clinically effective. The very different nature of the interventions used in each

trial published to date, combined with the inconsistent results of studies using the same intervention.

**8.4 Probiotics for Other Skin Applications: Oral and Topical:** The use of oral probiotics for promoting immune responses in the skin has been explored in relation to UV light-induced skin damage, where one clinical study suggested that probiotic treatment may have a role in promoting recovery from such damage in those most susceptible to it. There is little evidence that this mode of treatment would lead to any long-term modulation of the skin microbiota, it may nevertheless lead to long-term immune or skin barrier changes. The optimal approach for topical probiotic treatment may be the use of probiotic metabolites because to oral administration where such metabolites may be degraded in the upper gastrointestinal tract.

**8.5 Conclusion:** Studies suggest a potential role for selected probiotics in the prevention of eczema. Our own meta-analyses suggest that a prenatal component of treatment is important for beneficial effects. Studies of probiotics for other skin conditions are at a much earlier stage of development but show promise, particularly in the augmentation of wound healing.

**Part II Clinical Crossover between Nutrition and Dermatology**

- Nutrition may play a role in acne development, especially a diet with high glycemic load.
- Given that acne has a complex etiology of several factors, it seems unlikely that one nutrient could be responsible for its clinical manifestations.
- Retinoids and carotenoids, which are, respectively, metabolites and precursors of vitamin A, target skin cells.
- The role of dairy foods in acne development has not been fully validated, and proper clinical studies are needed to address this role.
- Few clinical studies have successfully addressed the relationship between diet and acne.

**9.1 Introduction:** Nutrition and diet affect overall health and general well-being. Acne is one of the most common dermatological conditions, affecting millions of people worldwide. It is generally accepted that excess sebaceous lipids, hormones, bacteria, and hyperproliferation of follicular cells are the major etiological factors for acne.

# Chapter 9. Diet and Acne

**Apostolos Pappas**

The current status of the relation between diet and acne is unclear and under debate. On one hand, the American Academy of Dermatology (AAD) published recommendations in 2007 suggesting that caloric restriction has no benefit in the treatment of acne and that there is insufficient evidence to link the consumption of certain "food enemies" to acne. One of his main principles and teachings was "Let food be your medicine, and let medicine be your food." Before continuing with the review of the publications of the last few years, it is appropriate to quote the conclusions of that review. It turns out that there are no meta-analyses, randomized controlled clinical studies, or well-designed scientific trials that follow evidence-based guidelines for providing solid proof in dealing with this issue.

**9.2 Could Certain Essential Nutrients Affect Acne?** It is obvious that Hippocrates' ancient but wise statement should hold some truth when applied to acne, given that the most efficacious current therapies for acne are retinoids. Oral administration of isotretinoin and topical application of its isomers and natural retinoids are used as anti-acne therapies. 13-*cis*-retinoic acid (RA) is the only drug that targets all four pathogenic factors of acne and is the most efficient so far in regard to sebum suppression. Vitamin A deficiency causes abnormal visual adaptation to darkness (night blindness) but also dramatically affects cutaneous biology.
The susceptibility of keratinocytes to the antiproliferative effects of vitamins A and D has been documented  They control metabolism, inactivation, activation, and elimination of specialized skin cells. The body's endocrine system is demonstrated by the fact that the skin is the site where active vitamin D metabolites, such as 1,25(OH)2D3, are synthesized.
In keratinocytes, 1,25(OH)2D3 regulates growth and differentiation; and for this reason vitamin D analogues have been developed for the treatment of psoriasis, an aggressive hyperproliferative skin disease. These pathways involve metabolites of polyunsaturated fatty acids (PUFAs). Numerous studies have revealed that clinical imbalances of specific essential fatty acids are associated with a variety of skin problems, such as dry, itchy, scaly skin, which is a hallmark sign of fatty acid deficiency. Sebum analysis demonstrates that these essential fatty acids and their derivatives comprise small amounts of surface lipids.  A recent nutritional clinical study in two groups of women who consumed flaxseed or borage oil for 12 weeks revealed that the daily ingestion of 2.2 g ALA or 2.2 g linoleic and g-linolenic acid, respectively, demonstrated skin benefits.  Another class of nutrients that derives from the diet includes minerals such as zinc, copper, selenium, and iron, which known to influence anti-inflammatory and proinflammatory enzymes.

**9.3 Could Diet Affect Acne Status:** The fact that Western diets are often deficient in the longer-chain omega-3 s and their precursor ALA raises an additional issue for this discussion. It is known that the ratio of omega-6 to omega-3 fatty acids in a typical western diet ranges from about 10:1 to 20:1. The omega-6 fatty acids are thought to induce more proinflammatory mediators and have been associated with the development of inflammatory acne. In general, our Western diet is not just deprived of omega-3 s but is also

a diet rich in refined carbohydrates. It has been reported that people living in the Kitavan Islands and the Aché hunter-gathers of Paraguay do not suffer from acne and that it is associated with their low-glycemic diet, associated low-glycemic-index diet with reduced acne risk.

**9.4 Status and Studies:** That review summarized the few poorly designed studies, more than three decades old, which contained few objective data or analyses. In general, those studies were inconclusive due to various methodological limitations. Could they be responsible for acne as milk exposes us to the hormones that cows produce during pregnancy? Given also the fact that hormones play an obvious role in acne, sebum production may be influenced by androgens and hormonal mediators, including sex hormone-binding globulin (SHBG) and insulin-like growth factor-1 (IGF-I), all of which may be influenced by dietary factors.
An important point is that the dermatologists should not ignore the vast amount of literature on the inverse association of milk or calcium to obesity.
Certainly any "milk animosity" tendency that has been created should be mediated.
Interestingly, some studies suggested that milk consumption could potentially alter insulin production. Even if milk is responsible for elevated insulin levels, it is noteworthy that higher dairy intake, especially low-fat products, may lower the risk of type 2 diabetes in men and women. Although milk has a low glycemic index, the insulin response is comparable to that seen with high-glycemic-index foods. Indeed, insulin and a high glycemic index are perhaps the two most biochemically and clinically associated factors with acne.
Acne and insulin sensitivity were assessed after participants followed either a low glycemic load (25% calories from protein, 45% from low-glycemic-index carbohydrates) or a control (typical high glycemic load) diet. Randomly assigned participants were enrolled to the dietary intervention or to the control group and were followed for 12 weeks. Blinded dermatologists assessed the number of acne lesions, starting at baseline and then every 4 weeks. Participants on the low-glycemic-load diet experienced greater reductions in total lesion counts and inflammatory lesions than did those in the control group. In all, 31 of the acne patients completed sebum tests as part of the larger 12-week, parallel design dietary intervention trial. follicular sebum outflow and composition of skin surface triglycerides were assessed by instrumental methods. The association of acne and high-glycemic-load diets, there is weak evidence for an association between dairy product ingestion and acne.

**9.5 Conclusion:** The low glycemic diet induces relatively low amounts of insulin to keep blood glucose levels within the normal range, in contrast to a high glycemic diet, which requires more insulin to maintain glucose levels. This could lead to insulin resistance, which in turn causes numerous health problems including high blood pressure, heart disease, obesity, and diabetes.
Dermatologists should not ignore nutritional studies, and perhaps nutritionists should understand better the complexity of the skin and sebum production. These specialties should work together to elucidate the "nature of the beast" as it is obvious that much more research and clinical studies are needed to reveal the potential effects of diet or nutrients on acne. Is diet the sole reason, or are other lifestyle and environmental conditions, such as stress, sun exposure, and air pollution, important? To prevent acne by dietary manipulation may not be possible. There are scientifically plausible reasons—beyond the consumers' perceptions—to believe that nutrition can affect acne.

• Acne affects 80–90% of individuals during puberty, which is a period associated with a normal decline in insulin sensitivity.
• Acne remains relatively unknown among non-Westernized societies existing on low-glycemic-load diets.
• Low-glycemic-load diets may represent a unique dietary strategy for alleviating acne symptoms through improving insulin metabolism.

**10.1 Introduction:** Acne has long been thought to be associated with the consumption of certain foods. Diets high in carbohydrates and sweets tended to make acne worse, with chocolate thought to be the most offending factor. Allergenic skin tests were unable to identify the culprit foods suggesting that foods may aggravate acne through an indeterminate mechanism.

# Chapter 10. Glycemic Load and Acne

**Robyn Smith and Neil Mann**

The diet and acne connection finally fell from favor in 1969 when a study found no difference in acne after the daily consumption of a chocolate versus a placebo bar containing equivalent amounts of fat and sugar. The notion to “eat more” to prevent deficiency was in clear opposition to the idea of avoiding certain foods to prevent acne. However, in 1969 scientists were just beginning to become aware of the role of diet in the etiology of chronic disease states such as cardiovascular disease and diabetes. By the late 1970s, dietary advice had shifted from preventing dietary deficiencies to recommendations aimed at avoiding excessive consumption of food components.
Because the glycemic index (GI) was introduced to quantify the blood glucose-raising potential for a given sample of food containing 50 g of available carbohydrate. According to early scientific principles of medicine, to test the hypothesis that a dietary component is implicated in disease, one should demonstrate that (1) the diet of persons with disease is significantly different from those without the disease; (2) the signs and symptoms should be known to be or plausibly suspected of being caused by the dietary imbalance; and (3) correction of the dietary imbalance should result in alleviation of the signs and symptoms.

**10.2 Acne Pathophysiology: Could Acne Be a Metabolic Disease?** Acne is considered as a disease of adolescence, affecting 80%–90% of individuals aged 12–15 years. Clinical observation indicates that this condition can also affect prepubescent children and adults. Acne often begins during adrenarche (8–12 years), and incidence rates increase with pubertal maturation.  Acne has shown to correlate better with the proportion of testosterone to sex hormone binding globulin (SHBG), an indicator of testosterone bioavailability. Other biological factors, such as insulin and insulin- like growth factor (IGF)-I, can also stimulate sebum production and growth of keratinocytes in cell cultures
These physiological traits may influence one or more of the pathogenic processes involved in acne development, including (1) increased sebum production, (2) hyperproliferation and differentiation of follicular keratinocytes, (3) microbial colonization by *Propionbacterium acnes*, and (4) inflammation.
Acne clinically presents during puberty, which is a transient period of normal insulin resistance. Acne incidence follows the rise and fall of pubertal insulin resistance more closely than the change in androgen levels, as androgen concentrations remain elevated following acne regression during the late teenage years. Perhaps the strongest evidence for an association of acne and insulin resistance comes from the fact that acne is a common feature of women with polycystic ovary syndrome (PCOS), a condition characterized by hyperinsulinemia and hyperandrogenism. Clinical observation suggests that insulin resistance is the underlying feature in PCOS, as it generally precedes
and gives rise to hyperandrogenism.

**10.3 Secular Trends of Advancing Pubertal Age and Acne: Evidence for a Role of Diet?**
The reasons for the change in insulin sensitivity at the various hormonal stages of life are unknown. It has been suggested that insulin resistance of puberty may relate more to changes in growth hormone release than changes in body mass. This is in support of earlier

observations during the 1970s of the emergence of acne in Eskimos groups following the introduction of Western foods . The higher rates of acne in Eskimo groups paralleled the increase in annual per-capita consumption of refined sugar and flour, and the per-capita consumption of protein from animal sources showed an inverse relation. Only since the relatively recent exposure to refined high-GI carbohydrates have the Eskimos demonstrated faster growth earlier puberty, and dramatic increases in the incidence of obesity, diabetes, and heart disease.

At present, there are relatively few data available on the classification of foods according to their insulin response, although the correlation between glycemic and insulinemic responses is reported to be high ($r$ = 0.74 and 0.90, respectively when applying the glycemic load concept to whole diets, the glycemic load may be reduced by decreasing total carbohydrate intake or by selecting foods using the glycemic index concept. Traditionally, the Eskimo diet Glycemic index classification: low < 55; moderate 56–69; high > 70 b Values for the insulin response per gram serving weight was sourced from Holt et al. would have been low in glycemic load due to low intakes of carbohydrate and the consumption of low-GI foods. Consequently, reducing the dietary glycemic load may represent a unique dietary strategy to alleviate acne via a reduction in hyperinsulinemia and its hormonal sequelae.

### 10.4 Clinical Evidence of a Therapeutic Effect of Low Glycemic Load Diets in Acne Vulgaris

A recent randomized controlled trial found that a low glycemic load (GL) diet that mimics the diets of acne-free populations may alleviate acne symptoms and hormonal markers of acne. When compared to controls, participants on the low-GL diet demonstrated significant improvements in insulin sensitivity and hormonal markers of acne. These changes may also relate to the modest weight loss (2.5 kg) that occurred with the reduction in dietary glycemic load. This suggests that weight loss mediated the reduction in insulin resistance and its associated hyperinsulinemia, which may be important in the clinical regression of acne.

Dietary intervention trials suggest that low-GL diets can alleviate acne symptoms, possibly through improving insulin metabolism and decreasing the bioavailability of testosterone and IGF-I. These endocrine changes may influence the desquamation of follicular keratinocytes and sebum production, two primary factors involved in the development of an acne lesion.

- A recent randomized controlled trial found that a low-glycemic-load diet that mimics the diets of acne-free populations.
- Low-glycemic-load diets may alleviate acne by decreasing insulin demand and influencing mediators such as sex-hormone binding globulin, insulin-like growth factor-I, and insulin-like growth factor-binding proteins.
- It remains to be objectively determined whether weight loss associated with low-glycemic-load diets is the principal factor for the alleviation of acne.

# Chapter 11. Essential Fatty Acids and Atopic Dermatitis

**Anthony Vincent Rawlings**

Approximately 20% tends to have predominance among women and is a less severe condition. The transitional nature of intrinsic AD has led many groups to conclude that a skin barrier defect plays a role in the development of AD. In 80% of subjects with intrinsic AD, IgE levels are subsequently elevated, and patients develop extrinsic AD. However, the remaining 20% of patients never develop a high IgE level and continue with the intrinsic AD phenotype  barrier function might drive disease activity rather than be an epiphenomenon associated with the immunological processes involved in this disease. Newborn infants with family histories of atopic tendencies, reported that transepidermal water loss (TEWL) values were normal, but they became elevated once AD lesions developed. Skin surface hydration on the flexor forearms of the infants who developed AD, however, tended to be lower than that of the non-AD infants after 1 month.
External factors also influence barrier function. Environmental agents that can perturb barrier function include soap, detergents, olive oil, excessive use of topical corticosteroids, bacterial infection, inhalant allergens such as house dust mites, and all other topical formulations used to treat AD. Exposure to soap and detergents has been recognized as an exacerbating environmental factor in AD for four decades. The detrimental effects arise through damage to the lipid lamellae, washout of innate antimicrobial compounds and natural moisturizing factor (NMF), increased pH, and changes in proteases levels and their activities.

***11.1.2 Atopic Dermatitis and Aberrations in Stratum Corneum Structure and Function:***
Elevated TEWL and reduced skin hydration are signs of impaired SC function and are associated with AD. Clearly, the SC thickness and corneocyte size predominantly control the tortuosity of the SC, although this is also influenced by the swelling of the SC due to the presence of NMF, whereas the corneocyte covalently bound and free intercellular lipids consisting of predominantly ceramides, fatty acids, and sterols provide the waterproofing of the SC. Naturally, all these mechanisms are influenced detrimentally in AD. Generally, there was an increase in the ratio of the sphingosine-containing ceramides to the phytosphingosine-containing ceramides, which appears to be characteristic of hyperproliferative skin disorders also studied the deficiency of corneocyte protein-bound omega-hydroxyceramides in atopic dermatitis, which are present at 46–53 wt% of total protein-bound lipids in healthy skin. Their levels were reduced to 23–28% and 10–25% in nonlesional and lesional areas, respectively. These changes resulted in an increase in the corneocyte-bound omega-hydroxy fatty acid fraction and smaller increases in the fatty acid fraction.
AS position; containing C16–18 and C22, 24, 26 a-hydroxy fatty acids. This seemed to be specific for AD, as only a single peak was found in samples from senile xerosis, seborrheic eczema, and psoriasis tissues. Nevertheless, they did not observe increases in the cholesterol fraction. The levels of CER EOS, EOH, EOP, NP, and NH to be lower in atopic subjects. They also found that the larger ceramide species of >50 carbon atoms of CER NS,

NDS, NH, AS, and AH had lower expression, whereas the smaller species of CER NS, NDS, and AS tended to be expressed at higher levels. Positive correlations with improved barrier function were observed with CER NDS, NH, AH, AP, EOS, EOH, and EOP. A disturbance of extrusion of lamellar bodies and fusion of intercellular lipids has been reported by in the dry noneczematous skin of AD patients. The most compelling evidence for the role of excess serine protease activity due to reduced levels of SC-derived serine protease inhibitors in the pathogenesis of AD in humans comes from Netherton syndrome (NS). NS includes AD as one of its manifestations. Mutations in the serine protease inhibitor Kazaltype 5 (SPINK5) gene, which encodes the lymphoepithelial Kazal-type 5 serine protease inhibitor (LEKTI), have been linked to NS. Mutations in the SPINK5 gene have also been associated with AD. Reduced levels of LEKTI leads to a thinner SC because of uncontrolled serine protease degradation of the corneodesmosomes. In the AD animal mode, serine protease activity occurred throughout the entire SC. Additionally, corneocytes appeared to detach prematurely between the SC and the underlying nucleated cell layers. In AX, the presence of immature corneocytes may also contribute to decreased skin barrier function via their detrimental effect on the tortuosity of the stratum corneum. Differences in the presence of these CEs reflect aberrant keratinocyte proliferation and differentiation together with abnormal expression of transglutaminases. Selected reductions and increases in SC lipids lead to a weak hexagonally packed lipid barrier. The role of the LPP needs to be further defined, but changes are expected in lesional skin, as CER EOS linoleate levels are known to be decreased in subjects with AD.

**11.2 Importance of Omega-6 EFA in Atopic Dermatitis:** Burr and Burr were the first to define essential fatty acid deficiency (EFAD). Equally, examining blood samples, did not find a linoleic acid deficiency but did find a deficiency of their downstream metabolites: g linolenic acid (GLA), dihomo-g-linolenic (DGLA), and AA. The hypothesis that the fatty acid profiles in subjects with AD reflected lower activities of the desaturase enzymes.
The delta-5 and delta-6 desaturases are considered the rate-limiting enzymes in the formation of long-chain polyunsaturated fatty acids (PUFAs). The genes have been identified as FADS1 and FADS2 Linoleic acid is converted by delta-6 desaturation to GLA, which is followed by elongation to DGLA. The delta-5 desaturase converts DGLA to AA reported an alternate pathway via elongation of linoleic acid to eicosadienoic acid followed by a delta-8 desaturation to DGLA.
It was utilized in reversing epidermal hyperproliferation and increasing ceramide synthesis in guinea pigs induced into an EFAD state using a hydrogenated coconut diet (HCO) for 8 weeks. Guinea pigs were fed HCO (14 g/kg) to induce the deficiency state. To reverse the EFAD, the animals were fed for 10 weeks with primrose oil (PO), borage oil (BO), or equal quantities of BO+PO (BS) at doses of 60 g/kg. The combination treatment was designed to provide a level of GLA similar to that of PO but in a different position on the triglyceride backbone. All three GLA-containing diets suppressed epidermal thickening induced by the HCO diet, although the BO diet was most effective. Upon examination of thymidine incorporation into epidermal DNA, the GLA diets were effective but the BO diet was best. GLA was not detected in epidermal lipid fractions, but DGLA was. DGLA showed greater incorporation into epidermal phospholipids and ceramides in the order BO>BS>PO.
These results suggest that the absolute level of GLA in oils determines the accumulation of DGLA in epidermal phospholipids and ceramides and that the presence of GLA in the sn-2 position of the triglyceride oil mediates this whereas the presence of LA in the sn-1 position of the triglyceride facilitates the accumulation of linoleic acid in epidermal ceramides. The

content of other anti-inflammatory and antiproliferative metabolites of these lipids reflect their incorporation into the epidermal lipid fractions. Finally, stimulation of epidermal ceramide synthesis was greater for the borage oil diet alone, in the order BO>BS>PO.
On the SO diet, only 0.7 g of DGLA/100 g of total epidermal fatty acids was found in the skin. In comparison, the PO diet delivered 2.9 g, the BS diet delivered 11.7 g, and the BO diet delivered 15.2 g. These gave DGLA/LA ratios of 0.9%, 5.0%, 17.4%, and 27.5%, respectively. it is highly dependent on the blood for its supply of GLA and subsequently its further metabolites. Although the precise mechanism of action of GLA has not been determined in these studies, it is believed to be a direct effect of GLA itself on the keratinocyte differentiation process or due to the effects of its downstream metabolites.

**11.3 Effect of Omega-6 EFAs on Atopic Dermatitis**

Provision of appropriate essential fatty acids (EFAs) should be expected to correct the skin defects attributable to the EFA deficiency in AD. If in the presence of atopic eczema there is a reduced rate of conversion of linoleic to GLA, it makes sense to provide oils enriched with GLA. However, these are not drugs but nutrients, and the doses usually applied to humans are low compared to the dosages used in animal studies. Several oral intervention studies have been conducted to assess the effects of EFAs on the alleviation of AD.
It is possible that these positive effects are lost as a less healthy diet begins to be adopted or there is poor skin care leading to skin barrier problems. Also demonstrated that EPO was effective compared to SSO in Indian subjects. The enhanced oral bioavailability allowed the GLA to have a greater effect on epidermal differentiation. Indeed, relative to placebo, skin barrier function was significantly improved after consumption of the product. Respectively, the relative improvement in TEWL to baseline was 10.8% (7.65 vs. 6.82 g/m²/h at 8 weeks; $P < 0.05$); and relative to placebo it was 7.7% and 9.1% (9.1 vs. 8.4 g/m²/h and 8.7 vs. 7.9 g/m²/h at 12 weeks, $P < 0.05$). Evening primrose oil proved to have a stabilizing effect on the SC barrier, but this was apparent only with the water-in-oil emulsion, not the amphiphilic emulsion. The choice of vehicle is therefore an extremely important factor in the efficacy of topically applied evening primrose oil. In normal, healthy adults, topical borage and sunflower seed oils have been shown to be beneficial in winter when the levels of CER EOS linoleate are reduced. After treatment, the levels of CER EOS linoleate were normalized. Presumably in these studies, the linoleate is delivered to the epidermis and used as a precursor in the CER EOS biosynthetic pathway. However, the hydrolyzed EFAs from the oils may also be acting as PPAR agonists to induce epidermal differentiation, as discussed earlier.

# Chapter 12. Hair Biology and Nutritional Influences

**Michael Anthonavage**

**Core Messages**
• Hair biology is closely related to skin health and in many instances correlates well with it.
• Hair biology is affected by many factors and is useful for indicating how well the body copes with nutritional, psychological, and pathological stress over periods of time.
• Hair fiber structure, follicle health, follicle cycling, and pigmentation can be examined individually or as a whole when assessing hair health as it relates to nutritional status.

**12.1 Introduction:** Human hair emanating from the hair follicle is considered a biological tissue composed primarily of keratins, which serve the body in a variety of ways. It involves so many aspects of biological science including embryology, cell biology, molecular biology, stem cell biology, tissue engineering, and nutritional biology. Healthy hair is certainly an indicator of one's general well-being, particularly during the reproductive years, but hair loss itself is not a life-threatening event.

**Hair Biology and Nutritional Influences:** Hair integrity, growth and function rely on balance as do all biological systems. That balance is maintained largely in part by the homeostatic mechanisms of survival of those cells and tissues that comprise the organ. In humans, there are surprisingly few scientific studies correlating nutrition and hair health because the models for hair growth and health are primarily other species of mammals, such as mice and primates.

**12.2 Healthy Hair:** It is easy to recognize and is generally full bodied, shiny, lustrous, and free of flakes and damage. Unhealthy hair can result from over processing and, more importantly, inadequate nutrition or disease processes.

**12.3 Hair Biology:** The average human scalp has approximately 100,000 hairs emanating from hair follicles. Hair is one of the fastest growing tissues in the body. It acts as a sensory tool and conduit for excretion of sebaceous oils. Hair is also important for insulation and warmth in terms of thermoregulation and has been shown to harbor stem cell populations and immune cells, which are important for the regenerative capacity of the skin and innate immunity.

**Functions of the hair shaft:**
• Decoration, social communication, and camouflage.
• Protects against trauma and insect penetration.
• Protects against electromagnetic radiation.
• Provides a sensory "antenna" to feel the environment.
• Insulates against heat loss and heat gain.
• Mechanism of cleansing skin surface of squames, dirt, and parasites.
• Mechanism of outward transport of environmental signals: sebum, pheromones.

**Functions of pilosebaceous follicle:**

- Produces and moors the shaft.
- Provides epithelial and dermal reservoir for normal renewal and reparative response.
- Provides sensory apparatus for detecting shaft movement.
- Provides melanocyte reservoir for shaft pigmentation and epidermal repigmentation.
- Produces and releases sebum for shaft processing and epidermal surface protection.
- Provides reservoir of Langerhans cells.

Normally in the human scalp up to 90% of the hair follicles are in the anagen phase, 10–14% are in the telogen phase, and 1–2% are in the catagen phase. The length of the cycle can vary on different parts of the body. The outside of the hair follicle is not vascularized and receives nutrients from surrounding interstitial spaces through diffusion from local capillary beds in the dermis. These spaces are drained and perfused by a series of lymphatic vessels. The importance of all these vessels ensures a continuous rate of perfusion of both nutrients and waste that ultimately affect the heath of the follicular tissue.

**12.4 Influence of Age, Sex, and Ethnicity:** In men, facial and pubic hair follicle biology is regulated in part by androgens, and these changes are considered some of the hallmarks of puberty. Hormonal changes in the physical properties of hair have also been noted in postmenopausal women where the ratio of estrogens to androgens is altered resulting in thicker more noticeable facial hair.

**12.5 Influence of Adipose Tissue:** Fat tissue is highly vascularized and hormonally active as it responds quickly to excess caloric intake and a sedentary lifestyle. Adipose tissue is also a source of a variety of inflammatory mediators such as tumor necrosis factor-a (TNFa) and matrix metalloproteinases (MMPs), which can ultimately affect homeostasis of the surrounding tissue.

**12.6 Apoptosis and Hair Health:** Apoptosis is a nonpathological process of cellular death brought on by a wide array of conditions, one of which is mitochondrial insufficiency. A mosaic of respiratory chain deficiencies in a subset of cells in various tissues is typically found in aged humans.

**12.7 Stress:** Stress plays a critical role in hair biology, particularly transient hair loss. Emotional stress has long been discussed controversially as a cause of hair loss. If psychoemotional stress does affect hair growth, it would be mediated via definable neurorendocrine and/or neuroimmunological signalling pathway.

**12.8 Circulation and Perfusion of Nutrients**: The rate of blood flow through the skin is variable in the human body owing to its primary role in regulating body temperature, which is based on both internal metabolism and external temperature. Hair growth and maintenance are in part linked to vitamin B and folate assimilation through the gastrointestinal tract. These include erythrocyte and serum folate concentrations within the normal range, serum vitamin B12 levels of 300–1,000 ng/l, haemoglobin levels >13.0 g/dl, and serum ferritin concentrations of ³70 ng/ml. Other factors affecting nutrient bioavailability are associated with external stresses such as UV radiation and smoke, pharmaceutical drug use, and pathologies associated with the hair shaft and skin.

**12.9 Hair Fiber Stress:** As the external portion of the hair is a keratin fiber structure susceptible to external effects—whether mechanical, physical, or chemical—excessive sun exposure and heat from processing are the most frequent causes of hair shaft structural impairment. Photochemical impairment of the hair includes degradation and loss of hair proteins as well as degradation of melanin in the hair. Ultraviolet B (UVB) radiation is responsible for hair protein loss, and UVA radiation is responsible for color changes.

**12.10 Influence of Pharmaceutical Drugs and Herbal Extracts:** Pharmaceutical drugs, chemotherapy, and the components of medicinally used herbals can cause unhealthy hair formation and hair loss. Oral contraceptives and hormone replacement therapy, progestin, and progesterone, respectively, have been documented to cause hair loss as have retinoids and angiotensin-converting enzyme (ACE) inhibitors and androgens. Compounding the problem is the fact that there are limited studies available for documenting such approaches. When such studies are available, they are usually performed in murine models, which have not been shown to correlate well with human hair growth or hair maintenance.

**12.11 Disease and Inflammation:** Many disease states associated with abnormal skin and hair follicle biology focus on immunity, infection, and inflammation. A careful examination of shed hair reveals the etiology of a variety of alopecias due to systemic immune processes. Telogen effluvium is a condition preceded by severe systemic stress occurring at least 2 months prior to the loss of normal hair. Psoriasis is a chronic autoimmune disease affecting the skin resulting in thickened scaly patches of affected areas that may include the scalp. An inappropriate immune response and the interaction between epithelial cells, called keratinocytes, in the epidermis. Keratinocytes are derived from stem cells that originate from the epidermal compartment and/or cells from within the bulge area of the follicle. Infections on or in the skin usually indicate an imbalance in the skin's innate immunity. Tinea capitis is a fungal infection of the scalp, usually caused by *Microsporum* or *Trichophyton* species of dermatophytes. It usually occurs in prepubertal patients. The most severe form of tinea capitis is a kerion—a fluctuant, boggy lesion with overlying hair loss. Tinea capitis can result in widespread hair loss with increased fragility of the hairs and frequent breakage, suggesting inappropriate or altered hair fiber synthesis.

**12.12 Historical Link Between Hair and Nutrition:** Many skin problems originate from extrinsic factors but also have an underlying dietary/nutritional component. The relation between nutrients and skin comes from the incidence of skin problems as a result of nutritional deficiencies. Other examples of dietary components that have an effect on skin are nicotinic acid, riboflavin, thiamine, and pyridoxine. Of particular note is the report that pyridoxine deficiency produces dermatological findings with features similar to that of essential fatty acid deficiency. Deficient consumption of several vitamins and essential fatty acids clearly results in cutaneous manifestations. The effects of oral supplementation with relatively high doses of vitamins, trace minerals, and fatty acids have indicated the possibility that dietary factors can modulate skin function and possibly hair health.

**12.13 Hair Color:** Hair color can also be an indicator of nutritional status, especially in cancer patients and children. The incidence of melanoma is higher in fair-skinned than in dark skinned individuals. There is in vitro evidence to support the notion that fair skin or poorly tanning skin contains more saturated fatty acids and therefore is more subject to UV-induced oxidative stress. Additionally, the Western diet, which is rich in omega-6 polyunsaturated fatty acids, is more associated with the development of a particular type of skin cancer called cutaneous melanoma. The precise mechanism(s) by which melanin content is reduced in hair and the role of aromatic amino acid availability in hair color change does warrant further investigation.

**12.14 Protein and Amino Acids:** The influence of protein on hair follicle biology in humans emanates primarily from studies on energy malnutrition, particularly in areas of the world where starvation and poor diet are prevalent. Gas chromatography studies have shown that it takes more than 4 weeks for both essential and nonessential amino acids to be detected in hair using stable isotopic abundance analysis after a group of young women were subjected to additional meat intake versus a group who had had meat removed from their diet. The phenotype of a hair root's response to protein malnutrition has been studied widely in children with kwashiorkor and marasmus conditions. The amino acids proline and lysine along with ascorbic acid from the diet are linked to a healthy dermis, where hair follicles reside. With increased age, dermal proteins begin to degrade.
This highlights the importance of serum ferritin levels in regard to hair loss. In women without systemic inflammation or other underlying disorders, serum ferritin levels £30 ng/ml are strongly associated with telogen hair loss

**12.15 Androgens, Insulin Resistance, and Lipids:** Healthy hair is not just about preventing hair loss; healthy hair may also play a role in preventing inappropriate or unwanted hair growth, or hirsutism, which is more of a symptom than a disease. The term hirsutism is usually applied to women when hair starts to grow in areas typically seen in men.
One of the primary enzymes involved in androgen metabolism is 5a-reductase, which converts testosterone to dihydrotestosterone. The latter is biologically active on androgen-sensitive hair tissue. Interestingly, there are a number of commonly used food/herbal products that have 5a-reductase properties. 5a-Reductase has two isoforms that are found in two distinct areas of the skin. Isoform type 1 is found in keratinocytes and sebaceous glands, and type 2 is found in hair follicles. Flavonoids from natural products that were potent inhibitors of the type 1 5a-reductase include myricetin, quercitin, baicalein, and fisetin. Biochanin A, daidzein, genistein, and kaempferol are flavonoids that are much better inhibitors of the type 2 isozyme than the type 1 isozyme. Hirsutism is a finding that can lead to subsequent metabolic diagnoses such as the metabolic syndrome. Metabolic syndrome describes a cluster of risk factors associated with being overweight that can lead to an increase in cardiovascular risk and further obesity. Because diet and insulin resistance are closely linked, there may be a plausible link to controlling hirsutism.
Dietary fats and fatty acids have been shown to modulate skin especially in hyperinflammatory conditions such as eczema and atopic dermatitis. Sebum's lipid content is unique to humans, and it has been demonstrated that essential fatty acids, a-tocopherol, and coenzyme Q10 are secreted to the skin through this mechanism, thus reinforcing the link between diet and skin. Foods and seed extractions rich in these fatty acids are

becoming quite popular with the average consumer and may play a role in acne. What these lipids have to do with hair formation is still not well understood, but they have recently been associated with cicatricial alopecia.
A key desaturase found in sebaceous glands, called stearoyl-coenzyme A (CoA) desaturase, is an endoplasmic reticulum-bound enzyme that catalyzes the delta 9-*cis* desaturation of saturated fatty acyl-CoAs, with the preferred substrates being palmitoyl-CoA and stearoyl-CoA. Stearoyl CoA desaturase activity leads to the production of the monounsaturated fatty acids palmitoleate and oleate, both of which are bactericidal against Gram-positive (but not Gram-negative) organisms in vitro. PPARs regulate sebocyte differentiation and promote hair follicle growth through prodifferentiating effects in keratinocytes in normal and inflammatory conditions. Owing to their diverse function and activation schemes, PPAR biology depends not only on dietary fatty acids but also on the ligands produced during inflammation.

**12.16 Trace Elements:** Trace element nutrition is the most studied aspect of hair as it relates to nutrition. Zinc, copper, magnesium, and selenium have been examined for their effects on hair biology. In alopecia areata, serum zinc levels are significantly decreased in subjects who are resistant to standard treatment when compared to control subjects. Of particular note, copper and magnesium levels were elevated in this study but not significantly. It is an essential micronutrient for human metabolism; it is a cofactor for more than 100 enzymes, facilitates protein folding, and helps regulate gene expression. Zinc is one of the oldest medicines in recorded history, with its use having been reported on 3000-year-old papyrus. Patients with malnutrition, alcoholism, inflammatory bowel disease, and malabsorption syndromes are at increased risk of zinc deficiency. Manifestations of zinc deficiency in the skin include nail dystrophies and hair abnormalities.
Copper is the key mineral in an enzyme called lysyl oxidase, which participates in the crosslinking of elastin fibers in the dermis. A combination of collagen and elastin is essential for tissues such as blood vessels and for a functional matrix in the dermis. Mice with a spontaneous recessive mutation in the lysyl oxidase gene show growth retardation, cyclic and progressive hair loss, hyperplastic epidermis, abnormal hair follicles, cardiac muscle degeneration, and reduced amounts of collagen and elastin in the skin and heart. Selenium along with zinc has been getting much attention in the cold and flu market as they are said to reduce the symptoms of the common cold. High doses of ingested selenium have been documented to have adverse effects in natural killer cell activity, hepatotoxicity, gastrointestinal disturbances, and nail and hair loss.

**12.17 Lipophilic Vitamins:** Key lipophilic vitamins to health are b-carotene, cholecalciferol, and tocopherol. b-Carotene is cleaved to form retinal, which in turn gives rise to retinol and retinoic acid, both of which play vital roles in epidermal biology including cell proliferation and epidermal differentiation. In vitro and in vivo studies have demonstrated that of the retinoids tested those most effective in altering the levels of cellular retinoic acid-binding protein in the skin were also capable of significantly altering hair cycle dynamics. There appears to be a relation between the ability of retinoids to increase cellular retinoic acid-binding protein, increase 3H-thymidine incorporation, and alter the dynamics of the hair cycle. It is noteworthy that vitamin D receptor knockout mice express a hair follicle cycling defect and a hyperproliferative phenotype resulting in disordered skin structure, epidermal thickening, and aloped.

**12.18 Hydrophilic Vitamins:** The hydrophilic vitamins, such as the family of B vitamins, have important for normal skin homeostasis. Cyanocobalamin, or B12, occurs in two metabolically active forms: (1) methylcobalamin, which is linked to DNA, protein, and lipid metabolism, and (2) coenzyme B12, which has a role in carbohydrate and fat metabolism. Cyanocobalamin deficiency is associated with a reduction in glutathione, which normally inhibits tyrosinase. Tyrosinase activity is fundamental to the production of melanin in the skin and hair.
Deficiency here may be caused by insufficient dietary uptake of biotin, drug–vitamin interactions, and perhaps an increase in biotin catabolism during pregnancy and in smokers. Symptoms of biotin deficiency in skin include dermatitis and hair loss. Biotin is one of the most widely used vitamins for hair as it is said to promote healthy hair growth and protect against dryness presumably through long-chain fatty acid incorporation into the cuticle. It also increases the elasticity of the hair's cortex, preventing breakage. There is evidence that impaired fatty acid metabolism secondary to reduced activities of the biotin-dependent carboxylases (especially acetyl-CoA carboxylase) plays an etiological role in the dermatological manifestations of biotin deficiency.

**12.19 Tools for Studying Hair Loss:** Certain tools are best suited for diagnosis in medical practice, whereas others can only be used to monitor hair growth under treatment in clinical arenas. The techniques can be classified as either semiinvasive or noninvasive. Each has its pros and cons and should be combined with accurate nutritional assays in the serum to correlate the cause and effect of the underlying problem in the hair and its follicle.

**12.20 Final Thoughts:** Malnutrition results from a deficiency of one or more basic nutrients and may be caused by insufficient dietary intake, malabsorption, poor utilization of nutrients, an increase in metabolic breakdown or catabolism, each of which can contribute to poor hair health in some form or fashion. To establish the cause of aberrant hair biology, one requires a history to identify known triggers, biochemical investigations to exclude endocrine, nutritional or autoimmune etiologies, and in many cases histology to identify the earliest stages of alopecia. The duration of hair loss upon presentation helps predict those patients in whom further investigation will have the greatest yield. Dietary influences on hair cycling and healthy hair growth are documented. Locked within the metamorphosing hair follicles in the balding scalp are all the secrets of growth and differentiation. Searching for these secrets should transcend the eagerness to "re-grow" hair on a bald scalp, an achievement which is of no great consequence. When we know these answers, we shall have the key, not to hair growth alone, but to all growth, which is, after all, the basis of all biological phenomena 24.

# Chapter 13. Detecting and Monitoring Nutrients on Skin Using Noninvasive Methods

**Georgios N. Stamatas and Nikiforos Kollias**

**Core Messages**

• Due to metabolic processes inside the body, most nutrients cannot be detected directly on skin.
• Based on their Raman spectral fingerprint, lycopene and b-carotene can be detected noninvasively on skin.
• Nutritional lipid uptake can be detected on the lips using infrared spectroscopy.
• Immediate and long-term effects of nutrition may be quantitatively evaluated using noninvasive optical methods.

**13.1 Introduction:** Technological progress in optics and electronics during the last couple of decades has allowed the development of sensitive analytical methods. Although originally such methods were designed for analysis of small samples, many quickly were adapted with probes that could come in contact with a living tissue.
We discuss how optical noninvasive methods can be applied to detect and monitor the evolution of concentrations of nutrients on the skin and the acute and long-term effects of nutrients that can be manifested as alterations in skin physiological parameters. When trying to detect nutrients or their effects in the skin, we need to consider how they might arrive there or why we might expect them or their metabolites to localize in the skin. For lipophilic molecules, we might expect to find an increased concentration in the stratum corneum (SC) or in subcutaneous fat, each of which presents challenges for detection. These problems might be worked out by exploring techniques that would allow probing of large areas of the SC to increase the path length through the absorber or by using near-infrared (IR) radiation to probe to greater depths. Microspectroscopy of histological sections might prove useful for determining the site in the tissue where the chromophores might concentrate. The distribution of chromophores is not constant over the skin surface area but varies. Therefore, when designing suitable detection methods, we need to consider such variations in regions of high or low concentration of the molecule of interest.

**13.2 Short Description of Optical Noninvasive Methods**

***13.2.1 Brief Introduction to Skin Optics:*** Interactions of electromagnetic radiation (light) in the ultraviolet (UV) B (280–315 nm), UVA (315–400 nm), visible (400–700 nm), and IR (700 nm–300 mm) parts of the spectrum. The fundamental principle is that light is allowed to interact with the tissue and is then collected and analyzed. Light interacts with the components of the skin in ways that can alter: (1) its intensity (light absorption by skin chromophores); (2) its spectral composition (fluorescence, Raman scattering, higher-order effects such as second harmonic generation and multiphoton fluorescence; and (3) its direction of travel (Mie and Rayleigh scattering, fluorescence, Raman scattering, higher

order effects). Following multiple scattering events, part of the light is remitted back out of the skin tissue. This light can be collected and analyzed for its intensity and spectral composition. An exception to the above fundamental principle is the method of skin chemiluminescence (CL) analysis. This method is used to quantify the amount of light that spontaneously is emitted from the skin during oxidation reactions and therefore does not require any incident light.

***13.2.2 Spectroscopic Methods:*** The spectroscopic methods usually involve a light source, a means of delivering the light to the skin and collecting the remitted light and a spectrometer that analyzes the collected light. The intensity spectral profile of the remitted light can be analyzed to give apparent concentrations of the skin chromophores and an indication of the light-scattering strength of the tissue. The method can be used to evaluate skin pigmentation, erythema (redness), and blanching reactions. The excitation (source) light is always at shorter wavelengths than the emission. In skin, tryptophan and other aromatic amino acids have characteristic fluorescence signals that can be detected. Particularly tryptophan fluorescence has been used as a marker of epidermal cell proliferation. Attenuated total reflection–Fourier transform infrared (ATR-FTIR) spectroscopy is used to analyze absorptions in the mid-IR part of the spectrum, indicating the presence of specific chemical bond vibrations. Identification of these bonds can provide information about concentration and molecular orientation of molecules that contain these bonds.

The ATR method limits the interrogated tissue volume to a couple of micrometers, which in skin corresponds to the uppermost layers of the SC. ATR-FTIR has been used in the analysis of SC lipids and secreted sebaceous lipids and therefore the collected information is the average over the illuminated tissue volume. Raman spectroscopy can be coupled to a microscope objective in a confocal arrangement that allows focusing the light beam on a small volume (1 × 1 × 5 mm). Chemiluminescence is the phenomenon of spontaneous light emission from the skin due to oxidative reactions and recombination of free radicals. CL has been used to study the oxidative stress on the skin following UVA exposure and the effects of antioxidants.

***13.2.3 Macroscopic and Microscopic Imaging Methods:*** The principles of spectroscopy can be applied during macroscopic or microscopic imaging. An added advantage of imaging is that many instances it may be without contact. Using polarization filters in front of the source and the camera lens in an orthogonal orientation, one can exclude the specular reflections and collect purely color information of the imaged object . In the case of skin imaging, color is the result of light absorption by the skin chromophores, primarily melanin in the epidermis and hemoglobins in the vascular network. By filtering the source or the detector with a sequence of narrow bands, one can collect a stack of images that can be used to contract maps of chromophore concentrations of a skin area. The source light and the detected light can be independently filtered to spectral bands specific to skin molecules that can fluoresce. As with fluorescence spectroscopy, fluorescence imaging can be used to monitor the extent of collagen crosslinking, but it can also be used to accentuate pigmentation issues.

**13.3 Direct Detection of Nutrients in Skin**

***13.3.1 Carotenoids:*** They cannot be synthesized in the body and can only be introduced to it in the diet. They are thought to be potent antioxidants and free radical quenchers, and they therefore play a protective role for the tissues where they are found. Carotenoids have been shown to inhibit carcinoma formation in animal models. Owing to their lipophilic nature they naturally accumulate to relatively high concentrations in the SC, where they are thought to play a protective role from sun-induced oxidative stress. Their accumulation in the SC also makes them suitable targets for noninvasive detection. Lycopene and b-carotene absorb light in a broad, short visible range that is demonstrated by an orange color. Although reflectance spectroscopy has been used to estimate the carotenoid level in the skin of fruit  This could is due to its low specificity for carotenoid detection in the presence of much higher concentrations of melanin and hemoglobins. Therefore high concentrations are required for detection. Some carotenoids also exhibit fluorescence, although with low quantum yields  These signals can be followed by noninvasive Raman microspectroscopy and can even be enhanced when the excitation source is in an area where the molecule of interest absorbs light strongly.
In fact, using the appropriate wavelengths, we can enrich the contribution to the measured Raman signal of a particular carotenoid of interest. Comparing a small number of patients, they showed that healthy volunteers (*n* = 6) had higher carotenoid levels than patients with either actinic keratosis (*n* = 14) or basal cell carcinoma (*n* = 14). It was not clear. if there was a casual relation.  Apparently a diet rich in “ecological” eggs was able to increase the skin carotenoids detected with this method by 20% although this study was small (*n* = 6) and with no control group. In another study, 25 volunteers following a 4-week lycopenedeprived diet received oral lactolycopene or placebo for 12 weeks. Apparent concentrations of lycopene on skin were sensitive to lycopene deprivation and supplementation. The amount of skin carotenoids using light-emitting diodes (LEDs) and multichannel detection using four photomultipliers with single-photon sensitivity. The resonance Raman method was criticized for lack of good calibration as a comparison of the Raman data with traditional high-performance liquid chromatography (HPLC) of the same samples was Inconclusive. Using the dual wavelength approached used resonance Raman to detect carotenoid signals in an attempt to separately detect lycopene and b-carotene in skin. It was reported that the relative concentration ratios of b-carotene to lycopene varied significantly among subjects.

***13.3.2 Nutrient Lipids:*** Owing to metabolic processes, most injected molecules undergo significant changes in their structure before they can reach skin tissue. A recent report by Yoshida and coworkers claimed direct measurement of dietary fatty acids on human lips using ATR-FTIR. The lips were chosen as an area unaffected by sebaceous activity that could mask changes in lipid composition following ingestion of fatty acids.

**13.4 Detection and Monitoring Nutrient Effects on Skin:** What we eat can have measurable effects on skin properties. These effects can be immediate, such as in the case of food allergies, such as in the case of antioxidant activity following ingestion of carotenoids or as the chronic accumulation of advanced glycation end-products (AGEs) in the extracellular matrix of the dermis.

***13.4.1 Immediate Effects of Nutrition on Skin: Food Allergies:*** One of the most common immediate symptoms of food allergy are skin erythema (redness), angioedema (swelling), and utricaria (itchy skin). The most common delayed symptoms involving skin include exacerbation or worsening of eczema It has been shown that the concentration of

oxygenated hemoglobin, determined by analysis of diffusely reflected spectra from the skin, is a single parameter that can be used to quantify skin erythema, as in the case of irritant contact dermatitis or UV-induced inflammation. Whereas the DRS method is based on point measurements with a probe that needs to come in contact with the skin, spectral imaging is a noncontact method that can be used to generate concentration maps of the skin chromophores, including oxygenated hemoglobin. This is achieved by generating a water concentration map based on the light-absorbing properties of water in the near-IR. Note that local accumulation of fluid (mostly water) is what causes skin swelling in edema. Orthogonal polarization imaging has been used to document erythema in acne lesions. Compared to spectral imaging, orthogonal polarization is more qualitative than quantitative.

***13.4.2 Midterm Effects of Nutrition on Skin: Antioxidant Activity and Photoprotection***
Measuring spontaneously emitted photons from the skin surface in a completely dark room can provide an evaluation of the oxidative status of the skin. Following UVA exposure, for example, the CL counts initially increase and then decay in an exponential-like fashion. We have reported that the initial burst of the CL signal depends on the UVA fluence rate, whereas the decay of the signal following exposure can be related to the UVA dose. Using clinical evaluation and DRS for assessing UVB-induced skin erythema, it has been demonstrated that oral administration of tomato paste that is rich in lycopene may provide protection against acute and potentially longer-term aspects of photodamage.

***13.4.3 Long-Term Effects of Nutrition on Skin: Glycation:*** Long-lived proteins, such as collagen and elastin molecules in the dermis, in the presence of sugars undergo a series of nonenzymatic glycation and oxidation reactions that result in intra- and intermolecular crosslinks. It has been shown that such AGEs accumulate in the dermis as a function of chronological aging. A convenient way to measure certain dermal AGEs, in particular pentosidine and *N*, elysine, is in vivo fluorescence spectroscopy. The fluorescence excitation/emission maxima are 335/385 nm for pentosidine and 370/440 nm for *N*, e-(carboxymethyl) lysine. Moreover, this method has been used to demonstrate that increased levels of dermal AGEs correlate with increasing risk of developing diabetes related health complications and can even be a strong predictor of cardiac mortality in diabetic patients. Interestingly, AGE-related skin fluorescence has been shown to increase transiently postprandially which opens the possibility of its potential use in metabolic monitoring.

**13.5 Conclusion:** Technological advancements in electronics and photonics catalyzed the development of noninvasive methods for in vivo analysis of skin composition.

# Chapter 14. Nutritional Clinical Studies in Dermatology

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**Core Messages**

• Nutrition has long been associated with playing a principal and multifaceted role in skin health, beauty, integrity, and aging, both directly or indirectly, through multiple pathways and cofactors implicated in skin biology.
• The onset and the clinical courses of various common skin diseases, including acne, psoriasis, atopic dermatitis, and hair loss, have been shown to be critically affected by nutritional patterns and habits.
• Abnormal nutritional conditions, such as obesity anorexia nervosa, manifest in specific cutaneous appearance features and altered function of the skin.
• Skin photoprotection, rendered by various nutrients, has been well documented.
• Appropriate nutritional supplementation has been shown to exert beneficial effects on impaired skin's structural and functional integrity, restore its appearance, and promote skin health.

**14.1 Introduction:** The structural integrity, functional capacity, and regenerative potential of human skin are known to be influenced, to a variable extent, by a plethora of factors that greatly affect our appreciation of its overall appearance and our perception of its health and beauty. Heredity, sunlight, environmental or occupational exposure, chronic disease, medications, drug abuse, hormonal supplementation, psychosomatic stress, and poor socioeconomic conditions have all been implicated in the pathophysiology of skin abnormalities and aging. Nutrition is one of the earliest and most important factors strongly associated with skin health. The degree of its impact on skin physiology and the mechanisms involved in nutrition-dependent alterations in skin structure and function remain highly controversial.

**14.2 Effects of Nutrition on Specific Diseases in Dermatology**

***14.2.1 Nutrition and Acne:*** Acne has traditionally been a skin disease considered to be strongly associated with nutritional behavioral patterns and habits. Within the dermatology medical community, a consensus seems to have emerged that diet is indeed unrelated to the etiology or progression of acne. It is linked to milk consumption. Most of the milk and dairy products consumed in the United States came from pregnant cows. Researchers concluded that there was an association between drinking milk and acne. Furthermore there was a 20% increase in the prevalence of acne in milk drinkers in that study, based solely on memory. A reverse association was reported between the consumption of milk fat and acne. If milk is responsible for elevated insulin levels, is noteworthy that higher dairy intake, especially low-fat dairy intake, may lower the risk of type 2 diabetes in men and women. Insulin and a high glycemic index are perhaps the two factors most scientifically and clinically associated with acne. Foods with a high glycemic index may contribute to acne by elevating serum insulin concentrations, suppressing SHBG and raising androgen levels.

[1] Both authors contributed equally to the manuscript.

***14.2.2 Nutrition and Atopic Disease:*** The beneficial effect of docohexaenoic acid supplementation in atopic eczema. In a similar fashion, oral evening primerose oil was reportedly of benefit to patients with moderate to severe eczema whereas a dietary supplement with fish oil has been shown to improve clinical management of psoriasis and eczema.

***14.2.3 Nutrition and Psoriasis:*** Diet has been suggested to participate in the etiology and pathogenesis of psoriasis, a T-cell-mediated inflammatory disease characterized by hyperproliferation and poor differentiation of epidermal keratinocytes. Protracted fasting and low energy and vegetarian diets were shown to improve psoriasis symptoms in some studies. It has also been established that severe psoriasis may lead to nutrient depletion, especially of protein, folate, and iron. Nutrient deficit events have been attributed mainly to accelerated loss from the hyperproliferation and desquamation of the epidermal layer of skin in psoriasis. Dietary fish oil has been found to have beneficial effect on psoriasis, but dietary supplementation with very-long-chain w-3 fatty acids was no better than corn-oil supplementation in treating psoriasis.

***14.2.4 Nutrition and Hair Loss:*** Hair loss is a common problem for both sexes, affecting up to 80% of men and 50% of women during their lifetime. Nutrition and caloric restriction malnutrition syndromes or anorexia nervosa, are well known to affect hair health and loss.

***14.2.5 Nutrition and Other Skin Diseases:*** Reactive skin is identified by marked sensitivity of the skin to physical or chemical stimuli and occasionally by impaired ability for the skin barrier function to recover.

***14.2.6 Nutrition and Photoprotection:*** The most damage is induced by ultraviolet (UV) exposure is sunburn, and there has been evidence of its prevention by nutritional supplementation. b-Carotene (15–180 mg/day) and lycopene (up to 10 mg/day), two efficient oxygen quenchers, have been shown to prevent sunburn in humans. Photoprotection by nutrients is well documented. Skin exposure to UV radiation leads directly or indirectly, through the generation of reactive oxygen species, to a large range of photodamage affecting cellular lipids, proteins, and DNA. It is involved in erythema appearance, premature skin aging, photoimmunosuppression, and skin cancer. The UV-induced depletion of Langerhans cells, the major antigen-presenting cells in the skin. Oral supplementation with the probiotic strain *Lactobacillus johnsonii* has been shown to accelerate the recovery of human skin immune homeostasis after UV-induced damage. This could enable the design of novel nutrition-based compounds and interventions for preventing UV-induced damaging effects.

**14.3 Nutritional Abnormalities and the Skin**

***14.3.1 Malnutrition:*** Primary nutritional shortfall are rare. Most malnutrition syndromes in current medical practice are related to secondary elementary or macronutrient deficits due to prematurity (infants) or are seen in patients with long-term total parenteral nutrition, gastrointestinal pathology such as Crohn's disease, neoplasias, cystic fibrosis, or intestinal bypass procedures.

***14.3.2 Eating Disorders: Anorexia Nervosa and Bulimia Nervosa:*** Cutaneous display look somatic expression of underlying disorders, vomiting, abuse of drugs such as laxatives and diuretics, and psychiatric morbidity. Skin manifestations of eating disorders have four groups: (1) starvation and/or malnutrition; (2) self-vomiting; (3) drug consumption (4) concomitant psychiatric illness. Further two main groups of signs: (1) frequent signs (2) guiding signs.

Body mass index (BMI) of £16 should be considered a threshold value at or beyond which skin changes are more frequent. Symptoms due to starvation include, in order of frequency: xerosis, lanugo-like body hair, telogen effluvium, carotenoderma, acne, hyperpigmentation, seborrheic dermatitis, acrocyanosis, perniosis, petechiae, livedo reticularis, interdigital intertrigo, paronychia, generalized pruritus, acquired striae distensae, slow wound healing, prurigo, edema, linear eczema craquelé, acral coldness, pellagra and scurvy, acrodermatitis entheropathica.

Lanugo-like body hair is a frequent sign of AN, especially in young patients. It presents as fine, downy, pigmented hairs on the back, abdomen, and forearms. Acne might be referred to starvation but could itself be a risk factor for AN. Carotenoderma is due to marked ingestion of carotenoid-rich vegetables low in calories. Acrocyanosis could represent a more extreme form of a heat-conserving mechanism not uncommon in anorectics. Life-threatening episodes of thrombocytopenia are reported in the typical restricting-type of AN with purpura, gingival, nasal and gastrointestinal bleeding, and apparent bone marrow hypoplasia. Nail fragility, longitudinal ungueal striae, onychocryptosis, periungueal erythema, prurigo pigmentosa, pompholyx, eruptive neurofibromatosis, evident blood vessels due to decreased subcutaneous tissue and acquired pili torti have also been reported. The most characteristic cutaneous sign of purging-type AN is Russell's sign. Self-induced trauma often coexists with AN, varying from unconscious picking at the skin to severe self-destructive actions .

***14.3.3 Obesity***: Obesity is linked with multiple skin disorders having altered skin barrier function, sebaceous gland physiology, sebum production, sweat gland biology, regulation, lymphatic drainage, collagen structure, functional properties, process of wound healing, distribution, pathobiology of subcutaneous adipose tissue and impaired microcirculatory supply.

**14.4 Nutrition, Skin Aging, and Skin Beauty:** Increased life is linked with anticipation to look healthy and handsome. Nutritional factors show potential beneficial actions on the skin. Epidemiological evidence suggested that multivitamin use is associated with longer telomere length, a marker of biological aging, in women. Oral fish polysaccharides improve dermal thickness, skin wrinkling, color, and viscoelasticity after 2 months of supplementation. Silicon enhances skin microrelief and mechanical properties in women with photo-damaged skin.

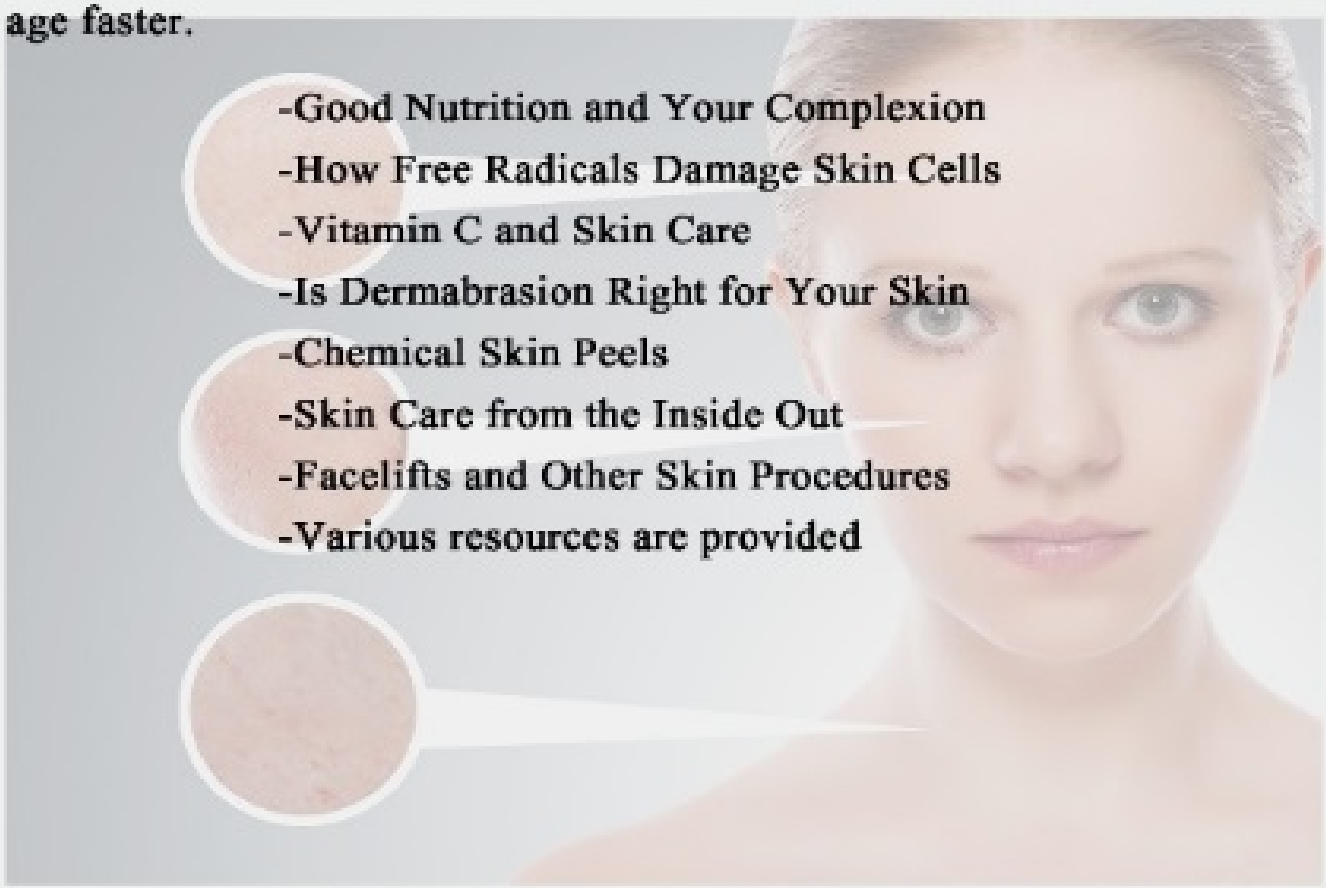
**14.5 Conclusion:** Nutrition is main part effecting "well-being" and regard of "health" in humans. The skin is largest and heaviest endocrine organ in the body provides a first impression about one's biological condition, age, and beauty. It reflects the psychosomatic balance and stress status. It also constitutes a prime target for ingested nutrients, either directly or indirectly. Increasing clinical appreciation of the growing significance of nutritional composition, patterns, habitual exposure and its interplay with multiple hormonal mediators, pathway cofactors, structural elements, and functional parameters of

the skin have been underscored by emerging evidence from the literature. On the other hand, accurate knowledge and appropriate handling of our potential to manipulate individual nutritional aspects as treatment modalities in skin pathologies could provide a powerful, patient-friendly tool to prevent, alleviate, or even cure common diseases in dermatology.

• Proper nutrition in both caloric content and composition is showed in healthy skin.
• Nutritional imbalances revolved around characteristic skin pathologies.
• Hyperinsulinemia and ingestion of high-glycemic-index foods have two factors most linked with acne.
• Cutaneous manifestations of anorexia nervosa are xerosis, alopecia, caries, opaque, fragile hair and nail fragility.
• Probiotics protect skin metabolism having good effect in atopic disease and psoriasis.
• Antioxidants with polyunsaturated fatty acids, zinc, taurine, and plant polyphenols imposed a stable hair cycle leading to decreased hair loss.
• b-Carotene and lycopene prevent sunburn in humans.
• Vitamins, carotenoids, and fatty acid supplementation have optimize skin physiology and prevent some skin diseases.
• Oral fish polysaccharides associated with an antioxidant increase dermal thickness, skin wrinkling, color and viscoelasticity.
• Lycopene, vitamin C, and soy isoflavones retain skin density, revamp skin firmness, provide microrelief, hydration and tone in menopausal women.

# Path To Beauty

When it comes to skin care, there is an overwhelming amount of choices that are available on the market. Unfortunately, most of them are not the best choices for your skin or for your general health. With all the toxic chemicals and cheap fillers that manufacturers put into many cosmetics, it is not surprising that the effect on our skin is only temporary at best. In the long run most products will actually make your skin look older and age faster.



-Good Nutrition and Your Complexion
-How Free Radicals Damage Skin Cells
-Vitamin C and Skin Care
-Is Dermabrasion Right for Your Skin
-Chemical Skin Peels
-Skin Care from the Inside Out
-Facelifts and Other Skin Procedures
-Various resources are provided