Photoaging: Mechanisms and repair

Jessica H. Rabe, MD, Adam J. Mamelak, MD, Patrick J. S. McElgunn, MD,
Warwick L. Morison, MD, and Daniel N. Sauder, MD
Baltimore, Maryland

Aging is a complex, multifactorial process resulting in several functional and esthetic changes in the skin. These changes result from intrinsic as well as extrinsic processes, such as ultraviolet radiation. Recent advances in skin biology have increased our understanding of skin homeostasis and the aging process, as well as the mechanisms by which ultraviolet radiation contributes to photoaging and cutaneous disease. These advances in skin biology have led to the development of a diversity of treatments aimed at preventing aging and rejuvenating the skin. The focus of this review is the mechanism of photoaging and the pathophysiology underlying the treatments specifically designed for its prevention and treatment. (J Am Acad Dermatol 2006;55:1-19.)

Learning objectives: At the conclusion of this learning activity, participants should be familiar with the mechanism of photoaging, the treatments for photoaging, and the data that supports the use of these treatments.

The aging process encompasses progressive physiological changes in an organism that lead to senescence; it refers to the decline of biological functions and the organism's ability to adapt to metabolic stress with time.[1] Many cultures revere the elderly as a source of wisdom. However, in western societies there is a stigma associated with aging.[2] The search for rejuvenation is as old as humankind and is reflected in ancient stories, including the Greek Argonauts and the Fountain of Youth, where the extensive efforts taken to restore youth are illustrated.[3]

More and more, individuals are seeking treatment for reversal of age-associated changes in skin. Patients are benefiting from recent advances by the medical and cosmeceutical industries that increase our understanding of skin homeostasis and the aging process. Our perception of age as well as beauty is largely dependent on the appearance of exposed skin,[4] and its condition is dictated in part by environmental effects, especially ultraviolet (UV) light.[5] This article reviews the mechanisms of UV-induced skin aging and discusses the compounds that have been shown, or have potential, to improve the appearance of photoaged skin.

Abbreviations used:

AHA:	α-hydroxy acid
AP:	activator protein
CoQ_{10}:	coenzyme Q_{10}
CO_2:	carbon dioxide
GA:	glycolic acid
GTP:	green tea polyphenols
HA:	hyaluronic acid
IL:	interleukin
MMP:	matrix metalloproteinase
NF:	nuclear factor
RA:	retinoic acid
RAR:	retinoic acid receptor
ROS:	reactive oxygen species
RXR:	retinoid x receptor
SNAP:	synaptosomal associated membrane protein
SPF:	sun protection factor
TGF:	transforming growth factor
TIMP:	tissue inhibitors of matrix metalloproteinases
TNF:	tumor necrosis factor
UPF:	ultraviolet protection factor
UV:	ultraviolet radiation
YAG:	yttrium-aluminum-garnet

THE AGING PROCESS

The basic biologic processes involved in aging lead to reductions in function and ability to tolerate

From the Department of Dermatology, Johns Hopkins University.
Supported by an educational grant from Connetics Corporation (to A. J. M).
Disclosure: Dr Sauder is a consultant for Amgen, Biogen, Centecor, Genentech, and Abbott Laboratories. Dr. McElgunn has received research funding from Syneron Medical Ltd and is a consultant for Advanced Aesthetic Institute Inc, Allergan, and Medicis.
Reprint requests: Warwick L. Morison, MD, Johns Hopkins at Green Spring, 10753 Falls Road, Suite 355, Lutherville, MD 21093. E-mail: wmorison@jhmi.edu.
0190-9622/$32.00
© 2006 by the American Academy of Dermatology, Inc.
doi:10.1016/j.jaad.2005.05.010

injury. There are two general theories of aging.[6] The first states that aging is a preordained process that is genetically determined. Support for this theory comes from telomere lengths, the terminal portions of chromosomes that shorten at every cell cycle. Once the telomeres reach a critical length, cell cycle arrest or apoptosis occurs.[7] Furthermore, primary cell cultures cannot continue to replicate indefinitely, thought by some to be a cancer-prevention strategy.[8]

Another theory suggests that aging is largely due to cumulative environmental damage.[6,9] For example, free radicals can be generated from oxygen during normal metabolism and likely contribute to this process.[10] Organisms have evolved cellular defense systems against the toxicity of free radicals, particularly oxygen-based free radicals, or reactive oxygen species (ROS). Longer lived species have higher degrees of enzymatic protection against ROS.[11] The activity of antioxidant enzymes[9] and the levels of nonenzymatic antioxidants decline with age,[12] allowing oxidative damage to occur.

In the skin, both genetic and environmental mechanisms likely contribute to the aging process. For instance, environmental factors such as UV radiation can damage telomeres and induce ROS, thereby inducing cellular senescence. Thus genetic processes and environmental effects may share a common final pathway.[13]

STRUCTURE, FUNCTION, AND AGE-RELATED CHANGES IN THE SKIN

A major function of the skin is to protect the organism from physical and environmental assaults. These stressors come in many forms; solar radiation, infection, temperature extremes, dehydration, and mechanical trauma are but a few. The skin also possesses and mediates immune, endocrine, and neural functions. All of these functions can decline with age (Table I).[3,6,14-39]

PHOTOAGING OF SKIN

Beyond the intrinsic aging process, sun-exposed areas such as the face, neck, and dorsum of hands and forearms encounter additional damaging effects, largely due to exposure to UV. Photoaging refers to the effects of long-term UV exposure and sun damage superimposed on intrinsically aged skin. It affects lighter skinned individuals most severely.[40] Many of the functions of skin that decline with age show an accelerated decline in photoaged skin.[6]

Clinical alterations

The clinical signs associated with photoaging are dyspigmentation, laxity, a yellow hue, wrinkles, telangiectasia, a leathery appearance, and cutaneous malignancies.[27,41-43] Old, photoprotected skin may have increased laxity and fold accentuation, but it is thin and lacks signs of actinic damage.[44] Seborrheic keratoses are common benign proliferative growths characteristic of aged skin[6] and may be related to sun exposure.[45] Specific phenotypes resulting from sun exposure, such as actinic elastosis and Favré-Racouchot syndrome (nodular elastosis with cysts and comedones), are also well described.[27]

Histopathologic alterations

Histopathologically, photoaged skin may show a loss of epidermal polarity or orderly maturation of keratinocytes. Individual keratinocytes are characterized by atypia, especially in the lower epidermal layers.[4,46] The thickness of sun-protected epidermis may decrease with age,[4] although it has been reported that it remains fairly constant.[47] However, epidermal thickness is greater in sun-exposed skin.[47] There is a flattening of the dermoepidermal junction that can lead to the appearance of atrophy, such as that seen in poikiloderma.[6,47]

Overall, the cell population of the photoaged dermis increases; fibroblasts are numerous and hyperplastic, and inflammatory infiltrates abound.[5] This chronic inflammation in photoaged skin is termed *heliodermatitis*.[48] The microvasculature is also altered[44] and vessel walls are thickened with deposition of a basement membrane–like material.[48] Fibroblasts in photoaged skin are elongated and collapsed.[49] Decreases in type I and III collagen are seen in intrinsically aged skin; however, these decreases are accelerated in sun-exposed regions.[47] Interestingly, fibroblasts from photodamaged skin are able to produce collagen in culture similar to cells from sun-protected aged skin.[50] Thus an intrinsic difference in synthesis is not thought to be responsible for decreases in collagen seen with photodamaged skin.

Elastin quantity decreases with age, yet in sun-exposed skin the quantity of elastin increases in proportion to the amount of sun exposure.[27] Elastin has been shown to be induced in vitro by UV radiation.[51] The accumulated elastin in the skin appears abnormal and seems to occupy the areas previously held by collagen.[47] It has been suggested that the increase in abnormal elastin results from a biphasic process beginning with hyperplasia of normal elastic tissue. The elastin becomes abnormal in appearance because of the effects of chronic inflammation.[27,48]

Photodamage is manifested primarily as the disorganization of collagen fibrils that constitute the bulk of the connective tissue and the accumulation of abnormal, amorphous, elastin-containing material.[44]

Table I. Skin components and systems: functions and changes with age

Cell type/component/system	Function	Change with age
Keratinocytes	Numerous eg, barrier function, mechanical protection, cytokine production, cell signaling[14]	↓ Proliferation and differentiation[6] ↓ Cell signaling and growth factor response[15,16] ↓ Barrier function with injury[17]
Melanocytes	Synthesize pigment for protection from UV radiation[18,19]	↓ Melanocyte number[6] ↓ Life span and growth factor response[20]
Langerhans cells	Antigen presentation[14]	↓ In number by 20%-50%; morphologic abnormalities[6] ↓ Cutaneous immune function[21,22]
Fibroblasts	Synthesis and degradation of ECM	↓ In number[6] ↓ Growth factor response[23]
Collagen	ECM component	↓ Biosynthesis[24] ↑ Stability and resistance to enzymatic degradation[25]
Elastin	ECM component	↓ Microfibril content[26] Porous, indistinct, and fragmented appearance[27]
Tissue inhibitors of matrix metalloproteinases	Protect collagen and elastin from endogenous breakdown systems	↓ Function[28]
Dermal vascular bed	Thermoregulation	Structural loss[6]
Subcutaneous fat	Thermoregulation, energy storage	Structural loss[3,29]
Endocrine system—vitamin D	UV protection,[30] calcium homeostasis	↓ Production[31]
Endocrine system—estrogen	Improves collagen content and quality, increase skin thickness, enhance vascularization[32]	↓ Production[33,34]
Nervous system	Sensation, thermoregulation	↓ Facial innervation, ↑ truncal innervation[35] ↓ Tolerance to cold exposure[36]
Miscellaneous		Delayed wound healing[37] ↓ Ability to repair DNA damage[38] ↓ Function of early population doubling level of cDNA-1, an inhibitor of angiogenesis[39]

ECM, extracellular matrix; *UV*, ultraviolet.

Solar elastosis is used to describe the accumulation of elastin material associated with prolonged sun exposure (Fig 1).[27] Fine wrinkles are a prominent feature of both intrinsically aged and photoaged skin; a precise histological correlate has not been identified.[52]

UV RADIATION AND SKIN BIOLOGY

UV radiation has numerous direct and indirect effects on the skin. It is estimated that approximately 50% of UV-induced damage is from the formation of free radicals, whereas direct cellular injury and other mechanisms account for the remainder of UV effects.[53]

Molecular and genetic changes

The molecular changes in DNA induced by UV radiation have been studied extensively in relation to photocarcinogenesis. Chromophores in tissue absorb energy and reach "excited states." They then either undergo chemical changes, transfer their energy to other molecules, or give off the extra energy as light or heat.[54]

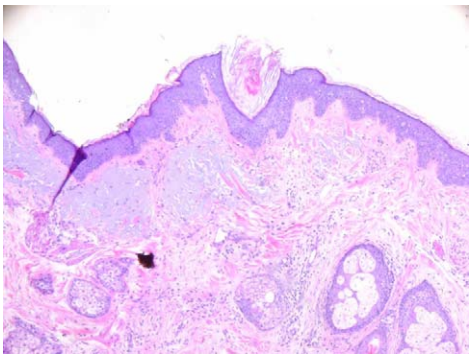

Fig 1. Section of photoaged skin with prominent dermal solar elastosis and mild perivascular and periadnexal inflammatory infiltrate. (Hematoxylin-eosin stain; original magnification: ×40.)

UV radiation from 245 to 290 nm is absorbed maximally by DNA,[55] thus implicating UVB as a primary mutagen.[56] UVB-induced DNA mutations occur by chemical change and include cyclobutane pyrimidine dimers and (6-4) photoproducts formed between adjacent pyrimidine bases (Fig 2).[57] Other DNA photoproducts have been reported, such

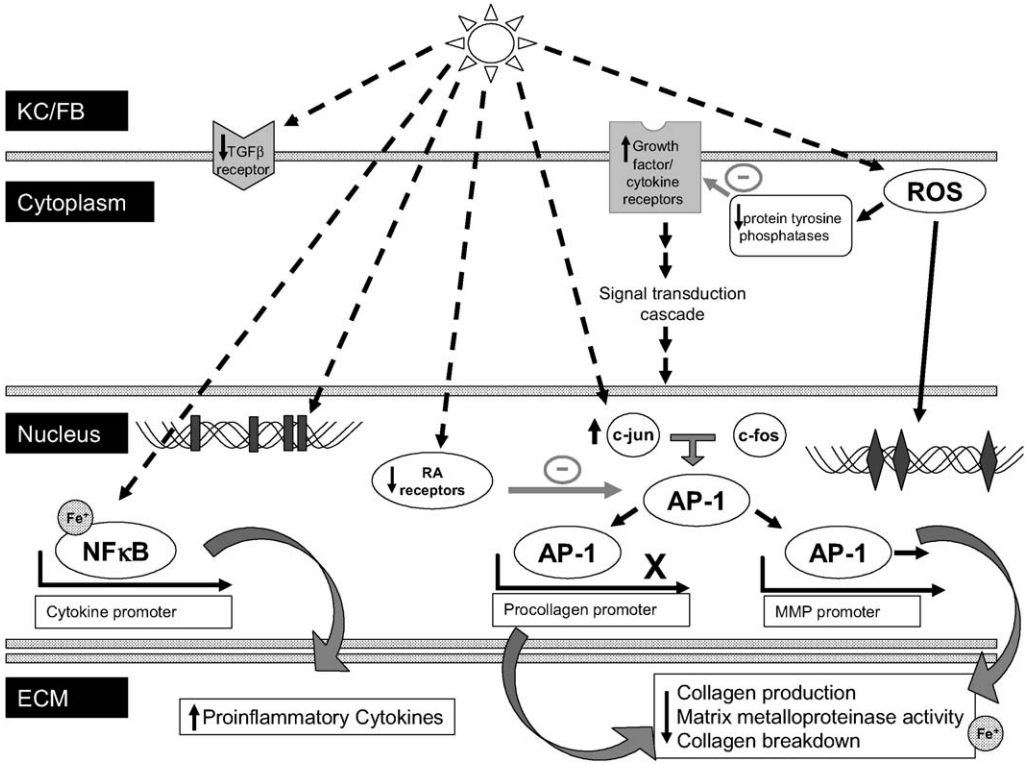

Fig 2. Effects of UV light on the keratinocyte *(KC)* and fibroblast *(FB)*. UV induces reactive oxygen species (ROS) which can damage DNA (◆) or can inhibit tyrosine phosphatases, leading to increased signal transduction and ultimately up-regulation of AP-1 transcription factor. UV can also directly up-regulate c-Jun, a component of AP-1, and can down-regulate retinoic acid *(RA)* receptors, which decreases RA inhibition of AP-1. Further effects of UV include direct DNA mutagenesis (▮), up-regulation of nuclear factor-κB *(NF-κB)*, and down-regulation of TGF-β signaling. These effects have been related to collagen production and breakdown, as well as to inflammatory cytokine production. *AP-1*, Activator protein 1; *ECM*, extracellular matrix; *MMP*, matrix metalloproteinase; *ROS*, reactive oxygen species.

as cytosine photohydrates, that are produced at low efficiency in experimental systems.[58] These DNA mutations may be clinically related to specific signs of photoaging as wrinkling, increases in elastin, and collagen damage are observed in animals exposed to UVB.[54,59,60] However, specific mechanisms by which DNA photoproducts lead to the photodamaged phenotype have not been elucidated.

UVA, and to a lesser extent UVB, can damage DNA indirectly through the generation of ROS (Fig 2).[61] These include superoxide anion, peroxide, and singlet oxygen.[62] ROS damage cellular DNA as well as lipids and proteins.[63,64] Mutagenesis by UVA may involve *trans*-urocanic acid and results in the production of singlet oxygen and DNA nicks.[65,66] 8-Hydroxy guanine is also a product of UVA mutagenesis via ROS induction.[54]

A recently recognized 4977 base-pair deletion of mitochondrial DNA (the "common deletion") is found in fibroblasts in the dermal compartment of photoaged skin. It is induced by UVA via ROS in vitro[67] and in vivo[68] and is considered a marker for UVA damage.[69] The mitochondrion, responsible for aerobic energy production, has the highest ROS turnover in the cell. Many of the genes involved in this process are encoded in the mitochondrial DNA, and mutations in the mitochondrial genome may be associated with the functional changes seen with aging.[70,71]

Effects on pigmentation

Sun exposure contributes to pigmentary changes. Suntan occurs in two steps: immediate pigment darkening, occurring in individuals with Fitzpatrick skin types III-IV, and delayed formation of new melanin. Immediate pigment darkening reaches its maximal state seconds after UV exposure and results from redistribution of melanin.[72] Delayed tanning is associated with an increase in the activity and

number of melanocytes.[73] Its function is photoprotection.[72]

Lentigines and guttate hypomelanosis are typically found on sun-exposed skin and are considered a consequence of UV exposure.[6] However, the mechanisms of their induction by UV radiation are not clear. Histologically, lentigines show a considerable increase in melanosome content of the basal keratinocytes.[74] Melanocytes have an increased capacity for melanin production,[75] and in some samples the number of melanocytes is increased.[76] In guttate hypomelanosis, decreases in melanin content[77] as well as melanocyte number are seen. Remaining melanocytes display abnormal morphology.[78]

Inflammation and vasodilation result from UV exposure, and this is clinically manifested as a sunburn.[72] The transcription factor nuclear factor (NF)-κB is activated by UV radiation; this is thought to be the initial step in the inflammation of sunburn reactions (Fig 2). NF-κB activation leads to increases in the proinflammatory cytokines interleukin 1 (IL-1), IL-6, vascular endothelial growth factor, and tumor necrosis factor (TNF)-α,[79] attracting neutrophils which increase oxidative damage through their production of free radicals.[53]

Vascular alterations

UV radiation has been shown to create a favorable environment for angiogenesis, mediated in part through increases in vascular endothelial growth factor.[80,81] Additionally, thrombospondin-1, an angiogenesis inhibitor, was down-regulated and platelet-derived endothelial cell growth factor, an angiogenesis activator, was up-regulated in keratinocytes exposed to UVB.[82] These changes in gene expression may contribute to the telangiectases seen in sun-exposed areas as well as facilitate the growth of UV-induced neoplasms.

Immunosuppression

UV irradiation has also been implicated in local and systemic immunosuppression,[83] which may have implications in cutaneous tumor surveillance.[84] Langerhans cells undergo numeric, functional, and morphologic changes after UV exposure, resulting in their depletion from the skin.[85] Decreases in contact hypersensitivity responses[86] as well as delayed-type hypersensitivity[87] occurring after UV exposure have been noted. This immunosuppression is partially mediated by DNA damage[88] as well as by altered cytokine expression. Increased production of the immunosuppressive cytokine IL-10 has been noted in the cutaneous inflammatory infiltrates after exposure to UV.[89,90] UV-induced immunosuppression may have arisen to prevent an autoimmune response to inflammatory products resulting from UV-mediated damage (eg, UV-damaged DNA).

UV effects on the extracellular matrix

Accumulating evidence from in vitro studies suggests that UV radiation mimics the actions of receptor ligands via the generation of ROS.[62,91] Within 15 minutes after UV exposure, receptors for epidermal growth factor, IL-1, and TNF-α are activated in keratinocytes and fibroblasts.[62] It is postulated that ROS oxidize and thereby inhibit protein-tyrosine phosphatases which function to down-regulate these receptors,[92] thereby resulting in receptor up-regulation (Fig 2).[93]

This increased receptor activation is thought to lead to activation of signaling kinases throughout the skin,[94] although the precise mechanism is unknown. The nuclear transcription factor activator protein 1 (AP-1) is ultimately expressed and activated. AP-1 controls transcription of matrix metalloproteinases (MMPs), enzymes responsible for degradation of the extracellular matrix. The MMPs include metalloproteinase-1 (a collagenase), MMP-3 (stromelysin), and MMP-9 (92-kd gelatinase). MMP expression is localized in both epidermal keratinocytes as well as in dermal fibroblasts.[95] Iron is required for the activation of MMP-1,[96] the major metalloproteinase responsible for collagen degradation (Fig 2).[97] ROS therefore directly contribute to tissue oxidation and degradation as well as interfere with signal transduction pathways involved in the expression of genes that are important regulators of collagen metabolism.[41]

Like AP-1, the transcription factor NF-κB is also activated by UV light via an iron-dependent mechanism.[98] It amplifies the UV response by stimulating the transcription of inflammatory cytokines and attracting neutrophils that contain preformed neutrophil collagenase (MMP-8) (Fig 2). NF-κB is also able to increase expression of MMP-9.[99]

MMP up-regulation can occur after only a minimal dose of UV, well below that required to produce erythema.[100] Furthermore, there is a dose-response relationship between UV exposure and MMP induction.[41] Exposure to UV light that is insufficient to cause sunburn can therefore facilitate the degradation of skin collagen and, presumably, eventual photoaging.[99] Minimal repetitive exposures to UV light at a dose equivalent to 5 to 15 minutes of exposure to midday sun on an every-other-day basis is sufficient to maintain these elevated levels of MMP.[95]

Collagen production is reduced in photoaged skin.[101] After UV irradiation, the procollagen pool

is markedly decreased and notably absent by 24 hours after exposure in vivo.[102] AP-1 and transforming growth factor (TGF)-β are involved in this UV-mediated down-regulation of collagen synthesis (Fig 2). AP-1 is composed of two subunits, the constitutively expressed c-Fos and the UV-inducible c-Jun.[94,100] Overexpression of the c-Jun component of AP-1 in cultured fibroblasts can decrease expression of type I collagen (Fig 2).[102] In addition, decreased expression of TGF-β2 and its receptor is noted throughout the epidermis and dermis after UV irradiation.[103] TGF-β is an important promoter of collagen synthesis,[104,105] and its predominant subtype in human skin is thought to be TGF-β2.[103]

Finally, damaged collagen itself may also down-regulate new collagen synthesis. When dermal fibroblasts are incubated in contact with type I collagen that has been degraded by MMP in vitro, synthesis of type I procollagen is decreased.[106] Similar effects are seen in vivo.[107] Mechanical effects are thought to contribute to this decreased collagen synthesis in the photoaged dermis. Collagen production occurs most efficiently in cells that maintain a high mechanical tension. Impaired spreading and attachment of fibroblasts onto degraded collagen may contribute to inhibition of collagen synthesis. A cycle is formed by which decreased production of new collagen due to poor adhesion of fibroblasts to damaged collagen leads to progressively worse photodamage.[49]

The sequence of events observed in photoaging can be compared with that in wound healing. Tissue inhibitors of metalloproteinases are part of this response, but like all wound healing, the process is not perfect. The result is a minute defect referred to as a solar scar.[95] The accumulation of these over many years via multiple exposures to UV light is thought to contribute to the photoaged phenotype.[99]

Retinoic acids and photodamage

The retinoic acid (RA) family constitutes several compounds including vitamin A (all-*trans*-retinol) and its natural and synthetic derivatives, known as retinoids. RA is important for normal epithelial growth and differentiation as well as for maintenance of normal skin homeostasis.[108] RA compounds have been shown to negatively regulate AP-1.[109]

RA compounds exert their effects through two families of nuclear receptors, namely, retinoic acid receptors (RARs) and retinoid X receptors (RXRs).[108] UV radiation rapidly decreases the expression of the two predominant retinoid receptors in human skin, RAR-γ and RXR-α, in vivo. This is associated with a near complete loss of the induction of RA-responsive genes.[110] In UVB-irradiated cultured keratinocytes and melanocytes, these receptors are also decreased. This is normalized in melanocytes within 2 to 3 days, but not in keratinocytes. A decrease in the receptors for the RAs may allow an increase in activity of the AP-1 pathway, further increasing MMP activity (Fig 2).[111] As such, UV irradiation results in a functional deficiency of vitamin A in the skin.[112]

INHERENT DEFENSE MECHANISMS AGAINST UV RADIATION

Numerous endogenous mechanisms protect the skin from UV-induced damage. These include increased epidermal thickness,[72] pigment,[113] DNA repair mechanisms,[57] apoptosis,[114] tissue inhibitors of metalloproteinase,[95] and antioxidants.[115] Apoptotic mechanisms and endogenous antioxidants are thought to decline with age.[9,72] Thus, over a lifetime, these protective mechanisms may be overwhelmed, allowing the skin to succumb to the hazards of UV exposure, leading to photoaging and other conditions, such as skin cancer.

Epidermal thickness

An increase in epidermal thickness occurs after UV exposure and helps protect from further UV damage.[72] Increased epidermal and dermal mitotic activity has been reported about 24 to 48 hours after acute UV exposure.[116] The importance of stratum corneum thickening in photoprotection has been demonstrated in patients with vitiligo who lack melanin in specific areas.[117]

Pigment

The protective role of melanin pigment should not be underestimated. Black skin differs from white skin with respect to the size and number of melanosomes as well as aggregation pattern within melanocytes and keratinocytes.[118] Compared with black skin, white skin shows more dermal DNA photodamage, infiltrating neutrophils, keratinocyte activation, and IL-10 expression after UV exposure. Levels of MMPs are also increased.[113] Thus the distribution of melanin is thought to provide protection from sunburn, photoaging, and carcinogenesis by absorbing and scattering detrimental UV rays.[119]

Repair of DNA mutations and apoptosis

With UV-induced DNA damage, p53 transcription is activated and the cell is arrested in G1 phase to allow for DNA repair.[120] UV-induced mutations such as cyclobutane pyrimidine dimers and (6-4) photoproducts are repaired by endogenous mechanisms such as the nucleotide excision repair system.[121] If the damage is too severe, apoptosis may occur.[114] "Sunburn cells" describe keratinocytes undergoing apoptosis and therefore serve as a histologic marker

of UV damage. They can be found as early as 30 minutes after exposure to UV irradiation. An age-associated decrease in sunburn cell induction by UV irradiation is noted,[72] which suggests that apoptotic mechanisms decline with age. If DNA repair mechanisms or apoptosis should fail, cutaneous tumorigenesis may result.[122]

Tissue inhibitors of MMPs

Tissue inhibitors of MMPs (TIMPs) regulate the actions of MMP. Conflicting results have been found regarding the responses of TIMPs to UV irradiation. In a fibroblast culture, both TIMP-1 and TIMP-2 levels were decreased in a dose-dependent fashion after UV exposure.[123] However, UV has been shown to induce TIMP-1 in vivo.[95]

Antioxidants

The skin is equipped with enzymatic as well as nonenzymatic cutaneous antioxidants. Endogenous antioxidants include vitamin E, coenzyme Q_{10} (CoQ_{10}), ascorbate, and carotenoids,[115,124] whereas enzymatic antioxidants include superoxide dismutase, catalase, and glutathione peroxidase.[125] These provide protection from ROS produced during normal cellular metabolism. Excessive exposure to UV radiation is thought to overwhelm and deplete this antioxidant supply, thereby leading to a state of oxidative stress.[126] Concentrations of carotenoids are lower in human cutaneous malignancies, such as basal cell carcinoma, suggesting that these antioxidants are important in the skin's defense against UV radiation and photocarcinogenesis.[124]

UV radiation can influence endogenous antioxidant enzyme levels. After a single low or moderate dose of UV radiation, there is an initial decrease in antioxidant enzyme transcript levels in cultured fibroblasts. This is followed by an up-regulation of superoxide dismutase and glutathione peroxidase above baseline levels by day 5.[125] In a separate experiment examining irradiated fibroblasts, catalase and superoxide dismutase both decreased and recovered only to baseline levels at 5 days after a single exposure to UV radiation.[127] Both studies clearly demonstrated an initial decrease in antioxidant enzyme activity, which lasted for days. Repeated UV exposures before enzyme activity returns have the potential to lead to increased tissue damage.[127]

TREATMENT OF PHOTOAGING

Strategies for medical treatment and intervention for photoaging can be categorized into a unique paradigm based on disease prevention. Primary prevention refers to the reduction of risk factors before a disease or condition has occurred. The goal of secondary prevention is early detection of disease, potentially while still asymptomatic, to allow positive interference to prevent, postpone, or attenuate the symptomatic clinical condition. Tertiary prevention is the treatment of an existing symptomatic disease process to ameliorate its effects or delay its progress (Fig 3).[128] These treatments can further be classified in a manner similar to other medical interventions based on the type of clinical evidence used to demonstrate their efficacy (Table II).[12,129-155] At present, too many of the available therapies for photoaging have not been subjected to large, randomized, placebo-controlled, double-blind clinical trials.

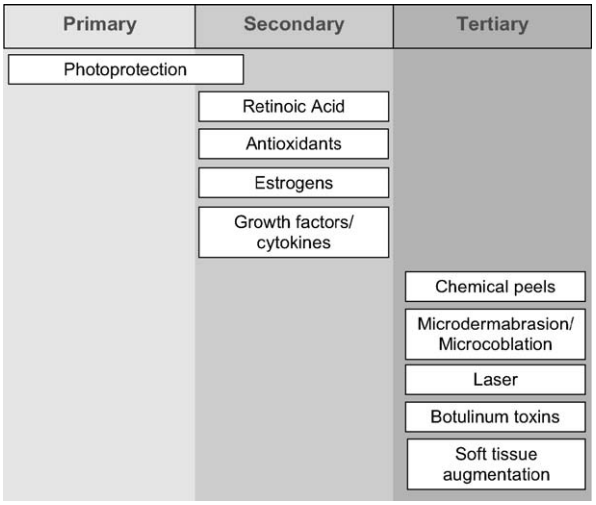

Fig 3. Photoaging treatments categorized by prevention strategy. Primary prevention reduces risk factors before disease occurs. Secondary prevention postpones or attenuates the condition. Tertiary prevention treats an existing symptomatic disease process to ameliorate its affects or delay its progress.[128]

Primary prevention

Sun protection. Perhaps the single most cost effective therapy that can be offered to patients is sun protection[156] in the form of sun avoidance, sun-protective clothing, and sunscreens. Peak times for UV exposure are between 10 am and 4 pm, and sun avoidance should be encouraged during this time.

Clothing, hats, and sunglasses that protect from sun exposure should be part of a package of protection. Photoprotective clothing is rated using the UV protection factor (UPF), which utilizes a defined source of UV and a photodetector to measure the amount of radiation transmitted through a sample of fabric. The UPF is calculated as a ratio of these two measurements with an allowance for the differing biologic effectiveness of the various wavelengths in UV radiation. A UPF of 40 to 50 provides excellent UV protection transmitting less than 2.6%

Table II. Evidence for antiaging therapies

Type of intervention	Drug/Process	Type of evidence*
Retinoids	Tretinoin	A2[131]
	Tretinoin	A1[132]
	Tazarotene, tretinoin	A1[133]
	Tazarotene	A1[134]
Anti-oxidants	Vitamin C	A2[135]
	Oral supplement (antioxidants, glucosamine, amino acids, and minerals)	B[136]
	Oral antioxidant supplement (vitamin E, vitamin C, carotenoid, selenium, and proanthocyanidin)	A2[137]
	Coenzyme Q_{10}	C[12]
	α-Lipoic acid	A2[138]
Hormonal	Estrogen, systemic	A2[139]
	Estrogen, topical	D2[140]
	Estrogen, topical	D2[141]
Growth factors and cytokines	—	D2[142]
New compounds	FROP-3	D2[143]
	Date palm kernel extract	A2[144]
Chemical peels	Glycolic acid (50%) peel	B[145]
Resurfacing techniques	Microdermabrasion	D2[146]
	Microcoblation	D2[147]
Laser systems	Erbium:YAG laser	D2[148]
	1450 nm diode laser (nonablative)	C[149]
Radiofrequency technology	—	D2[150]
Botulinum toxins	Botulinum toxin A	A1[151]
	Botulinum toxin B	A2[152]
Soft tissue augmentation	Bovine collagen	D1[153]
	Acellular dermal graft	D2[154,155]
	Hyaluronic acid derivative	D1[153]

Evidence categories: *A1*, Randomized, controlled, double-blind trial, N > 100; *A2*, randomized, controlled, double-blind trial, N < 100; *B*, randomized, controlled, single-blind trial ***or*** controlled, double-blind, nonrandomized trial; *C*, controlled trial (treatment vs placebo); *D1*, observational study, >1 treatment group, double blind; D2, observational study, no blinding.

*Evidence categories modified from Guyatt GH, Sackett DL, Sinclair JC, Hayward R, Cook D, Cook RJ. JAMA 1995;274:1880-4[129]; Cochrane reviewers' handbook 2.2.2 (updated March 2004). In: The Cochrane Library, issue 1, 2004. Chichester (UK): John Wiley and Sons, Ltd; 2004.[130]

of effective UV radiation.[157] Regular summer clothing typically has a UPF of 10 or higher and therefore provides protection equivalent to that of an SPF 30 sunscreen in normal use.[158]

Sunscreens have traditionally been divided into chemical agents, which absorb specific photons of UV light, and physical agents (sunblock), which reflect or scatter radiation. UVB-absorbing sunscreens include *p*-aminobenzoic acid and its esters (padimate A and O), the cinnamates, and salicylates. UVA sunblocks contain titanium dioxide or zinc oxide, whereas UVA-absorbing sunscreens include avobenzone (Parsol 1789) and terephthalylidene dicamphor sulfonic acid.[159] Parsol 1789 can degrade quickly[160] unless it is stabilized. Zinc oxide has been shown to provide better protection against UVA than titanium dioxide.[161]

The sun protection factor (SPF) is an internationally accepted standard by which the efficacy of sunscreens is assessed. The determination of the SPF is based on the minimal erythema dose of solar-simulated radiation and hence the prevention of mainly UVB-mediated erythema.[162] Since UVA also has a role in photoaging, SPF may be a poor guide to the ability of a sunscreen to protect against photoaging. Sunscreens containing both UVA and UVB protection may permit as much as 50% of UVA-induced free radical production even if they have a high SPF (>20) and are applied at the recommended dose (2 mg/cm^2).[163] Sunscreens with greater UVA blocking or absorbing ability may better protect against photodamage.[164] Some sunscreens are not effective in their protection from UV-induced photodamage even when properly applied and should therefore be regarded as an important but adjuvant therapy.

In animal studies, sunscreens have been shown to prevent photodamage and allow for its repair.[165,166] Although direct clinical evidence is lacking, indirect evidence that sunscreens allow for repair of photodamage comes from numerous clinical trials in which sunscreens are used in both control and treatment arms. For example, in one study use of sunscreen with an SPF of at least 15 produced an improvement in photodamage compared with baseline after 24 weeks.[134]

Secondary prevention

Retinoids. For years, retinoids have been the mainstay of topical therapy for the prevention and treatment of photoaging.[156] Tretinoin (all-*trans*-retinoic acid), a nonselective agent that activates all RARs directly and RXRs indirectly,[167] has been shown to improve the clinical signs of photoaging in controlled clinical trials.[131,132] Several weeks of

treatment are required before clinical improvement is appreciated.[168] The greatest obstacle to tretinoin use is irritation in the form of erythema, peeling, and stinging,[132,156] which decline with continued use.[169]

The benefits of retinoids are thought to be mediated at least in part by their effects on collagenase induction. Pretreatment with all-*trans*-retinoic acid inhibits UV-mediated induction of c-Jun protein, AP-1, and MMP.[94,100] Pretreatment also reduces loss and accelerates recovery of RAR-γ and RXR—α following UV exposure.[110] Partial restoration of markedly reduced collagen appears to be responsible for the observed clinical improvement.[170]

Tazarotene is a second-generation retinoid that selectively binds RAR-γ and RAR-β.[108] Like tretinoin, tazarotene is effective in the treatment of photodamage. Reduced atypia and restoration of keratinocyte polarity have also been noted after tazarotene therapy.[171] In a 24-week randomized, controlled, double-blind study, treatment with 0.1% tazarotene resulted in significant improvement in numerous clinical assessments of photodamage. Additional clinical improvement occurred during an open-label extension and had not reached a plateau by week 52.[134] When compared with a standard dose of tretinoin, a high-dose tazarotene regimen produced faster improvements in fine wrinkling and mottled pigmentation.[133] Tazarotene is also a strong irritant and, like tretinoin, is thought to inhibit AP-1—dependent gene expression.[109]

An active area of research is the development of receptor-selective retinoids to optimize therapy and minimize side effects.[108] Up-regulation of RA-response elements and the antagonizing actions of AP-1 are not linked,[109] which suggests that receptor-selective retinoids hold promise.

Antioxidants. Numerous antioxidants have been analyzed for their ability to prevent or reverse clinical signs associated with photoaging secondary to ROS. Strategies utilizing endogenous skin antioxidants as well as plant-derived or chemical compounds have been examined.

Topical vitamin C. Vitamin C, a potent antioxidant,[172] has been shown to prevent erythema and sunburn cell formation after UV exposure.[173] Vitamin C can also up-regulate collagen and TIMP synthesis in human skin.[174] Because of the short half-life of vitamin C,[175] skin care formulations commonly include its derivatives, which do not penetrate the skin as readily.[156] However, in a 6-month, double-blind, placebo-controlled, randomized trial examining the use of a topical vitamin C compound, a significant decrease in wrinkles was found when measured by optical profilometry.[135] Profilometry is a technique that utilizes skin replicas measured with special image processing software (optical profilometry) or laser (laser profilometry) to obtain an objective quantification of the skin surface topography.[176]

Oral supplements. Oral supplements offer a systemic method to treat photoaging. An oral supplement containing a combination of l-proline, l-lysine, manganese, copper, zinc, quercetin, grape seed extract, N-acetyl D-glucosamine, and glucosamine sulfate was shown to improve wrinkles by 34% in a pilot study when measured by optical profilometry.[136] Another oral supplement composed of a combination of vitamin E, vitamin C, carotenoid, selenium, and proanthocyanidin led to a significant decrease in induction of MMP after UV exposure. A reduction in UV-induced erythema was also noted, but it did not reach statistical significance.[137]

CoQ_{10}. CoQ_{10} is a component of the mitochondrial electron transport chain; it also acts as an antioxidant in the skin, with 10-fold higher levels in the epidermis than the dermis.[115] Topical CoQ_{10} use led to significant reductions in wrinkle measurements assessed by optical profilometry in a vehicle-controlled 6-month pilot study.[12]

α-Lipoic acid. α-Lipoic acid is an antioxidant and anti-inflammatory agent that has been previously shown to reduce the production of transcription factors such as NF-κB and indirectly affect the gene expression of inflammatory cytokines.[177] Treatment with α-lipoic acid has led to significant improvements in clinical and objective measurements of photoaging, including laser profilometry.[138]

Estrogens. In a cross-sectional analysis, oral estrogen use was associated with a statistically significant decrease in the risk for dry skin and wrinkling, but not atrophy.[33] These clinical changes may be due to an increase in collagen content.[139] Topical estrogen therapy can also lead to significant increases in collagen,[140] firmness, and elasticity, as well as wrinkle depth measured by optical profilometry.[141]

Growth factors and cytokines. Topical application of a combination of growth factors and cytokines has been evaluated in a pilot study for its effect on photoaged skin. A majority of patients showed clinical improvement in at least one facial area and a significant change in objective measurements by optical profilometry. In addition, new collagen formation was observed in biopsy specimens.[142]

New compounds. The fucose-rich polysaccharide "FROP-3" increases glycosaminoglycan biosynthesis in fibroblast cultures.[178] In a pilot study examining skin-surface microrelief, the pattern of fine wrinkling found in skin of any age, a cream containing FROP-3 showed a 10- to 15-year decrease in apparent age after 4 weeks of treatment in a

majority of patients.[143] Skin-surface microrelief changes predictably with age, with younger persons having a regular pattern of fine, thin lines. Wrinkles become deeper and thicker with increasing age.[179]

In a 5-week pilot study, an extract of date palm kernel was shown to reduce wrinkles by optical profilometry and visual assessment compared with placebo.[144]

Tertiary therapies

Tertiary therapies have been popularized because they not only target the clinical characteristics of photoaged skin, but can also be used in intrinsic aging as well as cosmetic augmentation. There are very few well-designed published studies that have specifically examined the effect of these therapies on photoaging and its clinical phenotype. For that reason, these therapies are briefly reviewed with an emphasis on their role in photoaging.

Chemical peels. A variety of chemical peels, including α-hydroxy acids (AHAs), salicylic acid, trichloroacetic acid, and phenol, are used to treat acne, acne scars, photodamage, and mottled hyperpigmentation.[180,181] They are classified as superficial, medium, and deep, which correlate with the depth of injury induced.[181] Portions of the epidermis and dermis are damaged with subsequent regeneration, resulting in a controlled wound and re-epithelialization with rejuvenation of skin.[182]

Glycolic acid (GA) is an AHA superficial peel that improves skin texture and reduces fine wrinkling and the number of actinic keratoses. It can also thin the stratum corneum and epidermis, as well as increase dermal collagen thickness.[145] GA is found in many skin creams and has been shown to modestly improve photodamage when used in this fashion.[183] GA can also increase sunburn cell formation and sensitivity to UV-induced erythema. Therefore it may paradoxically enhance short-term sensitivity to the damaging effects of UV light.[184]

Resurfacing techniques. Microdermabrasion exfoliates and ablates the superficial epidermis. Microcoblation uses low-frequency radiofrequency energy delivered via a recessed electrode bathed in saline solution on the skin. Both are hypothesized to create superficial epidermal injury and trigger a healing response.[147]

Microdermabrasion activates a dermal wound healing cascade and increases cytokines, MMPs, and type I procollagen mRNA with treatment.[185] Significant increases in the thickness of papillary dermis and improved organization in elastin and collagen have been observed[146] as well as improvements in hyperchromatic pigmentation.[186] Microcoblation acutely gives rise to a zone of vasculopathy in the mid-epidermal layers. Both processes give similar results for texture, appearance, clarity, and oiliness when evaluated subjectively.[147]

Laser systems. There are numerous applications for cutaneous laser surgery, including destruction of vascular and pigmented lesions, striae, verrucae, as well as dermal remodeling for treatment of photodamage.[187] Some lasers work through selective photothermolysis where controlled destruction of a chromophore occurs without damage to surrounding normal tissue.[188] Ablative and nonablative laser systems have been successfully used in the treatment of photodamage and wrinkles. Both methods increase collagen production; however, the exact mechanism by which this occurs is unknown.[187]

Ablative laser systems. Ablative systems include the carbon dioxide (CO_2) and erbium:yttrium-aluminum-garnet (YAG) lasers. The CO_2 laser is considered the "gold standard." Facial resurfacing with the CO_2 laser typically produces at least a 50% improvement in overall skin tone, wrinkle severity, and atrophic scar depth.[187,189,190] The erbium:YAG laser, developed to reduce the morbidity associated with CO_2 laser resurfacing,[187] has demonstrated comparable results with fewer side effects in some studies.[148] The biochemical changes seen after CO_2 laser resurfacing include increased mRNA of several cytokines (IL-1β, TNF-α, and TGF-β1), type I and type III procollagen, and MMPs.[191] Undesired effects of ablative systems include hypertrophic scar formation and pigmentary alterations. In addition, they induce significant morbidity until re-epithelialization occurs, which requires at least 1 week, and the full recovery period can be a month or more.

Nonablative laser systems. Nonablative systems are thought to induce collagen remodeling by creation of a dermal wound without disruption of the epidermis. They are popular among patients who are unwilling or unable to undergo the postoperative recovery associated with ablative procedures.[149] They are much less effective than ablative systems in the treatment of photoaging[187] but can reduce hyperpigmentation and telangiectases. The clinical efficacy of nonablative systems continues to be debated.[192] For example, a controlled half-face study of the 1450-nm diode laser demonstrated significant clinical improvement in periorbital rhytides as well as increases in dermal collagen assessed histologically.[149] However, in a separate study, 25 dermatologists clinically evaluated patients after 1450-nm diode treatment. Although all patients reported mild to moderate improvement, only 2 of the 25 dermatologists recorded a significant

positive treatment effect,[192] suggesting that modest changes induced by the laser may not be clinically meaningful.

Radiofrequency technology

Radiofrequency devices produce an electric current that generates heat through resistance in the dermis and subcutaneous tissue. They have been shown to improve cheek and neck laxity in a pilot study.[150] These clinical changes are thought to reflect collagen contraction followed by secondary collagen synthesis and remodeling.[193] Adverse events such as erythema, soreness, and second-degree burning have been reported.[150,194]

Botulinum toxins

Botulinum toxin A is a naturally occurring exotoxin produced by *Clostridium botulinum* that prevents local neuromuscular transmission. It was approved for cosmetic use by the Food and Drug Administration for glabellar lines in 2002. The toxin facilitates cleavage of synaptosomal associated membrane protein (SNAP)-25, which is required for exocytosis of acetylcholine,[195] thereby inhibiting muscle contraction. Although botulinum toxin A does not directly reverse changes in the extracellular matrix caused by photodamage, it gives the appearance of rejuvenation by relaxation of the underlying musculature. In a large placebo-controlled trial, it significantly reduced glabellar line severity.[151] The effects typically last 3 months.[196]

Botulinum toxin B targets the synaptobrevin protein, ultimately inhibiting acetylcholine release.[197] In a pilot study it was shown to be effective in the correction of crow's feet and was well tolerated.[152] However, its use in the treatment of facial wrinkles has not yet been approved by the Food and Drug Administration.[152]

Soft tissue augmentation

Soft tissue augmentation, or "fillers," are designed to address the subcutaneous atrophy that accompanies senescence. Fillers have been used to treat fine lines and sallowness in photoaging, but have a greater market in intrinsically aged skin and for other cosmetic purposes. Some approaches are mentioned briefly herein.

In autologous lipoaugmentation, fat is typically harvested from one region and can then be frozen and used in staged injections. The results are thought to last longer than other methods, although no objective studies have been completed.[198]

Bovine collagen has been regarded as the "gold standard" of injectable fillers. There are two commercially available products: an original bovine collagen preparation and a glutaraldehyde cross-linked bovine collagen that is more stable and longer lasting.[153] Maintenance injections are required approximately every 4 to 6 months.[199] Although these are effective fillers, drawbacks include immunogenicity and potential hypersensitivity reactions.[200]

An acellular dermal graft derived from human cadavers contains collagen, elastin, and glycosaminoglycans. It is available in sheets, requiring an incision for placement, as well as in a micronized injectable form. Its advantage is that it is human in origin. When compared with bovine collagen, this human cadaver–derived dermal graft retains a higher percentage of the original implant volume.[154,155] The usage of these products has decreased with the introduction of new fillers.

The hyaluronic acid (HA) derivatives, derived from rooster combs or through bacterial fermentation,[201] are less immunogenic compared with bovine collagen preparations because HA is chemically identical across species.[202] When tested in an animal system, a reduced inflammatory response and no signs of incompatibility were noted in the HA group compared with bovine collagen.[203] In a 6-month randomized study the HA product was judged superior to bovine collagen by a wrinkle-severity rating score and global aesthetic improvement scale, with longer lasting effects.[153]

A mixture of microspheres of polymethylmethacrylate (20%) and bovine collagen (80%) has been developed and is available outside the United States. One of its strengths is that the microspheres are too large to be phagocytosed, and therefore it is considered to last longer than other methods of augmentation.[204]

EMERGING THERAPIES

Numerous compounds have demonstrated efficacy in experimental systems and may prove to have a clinical benefit for the treatment of photoaging (Table III).[53,79,205-216]

Antioxidants

Oral soy isoflavones. Soy isoflavones can enhance the activity of endogenous antioxidant enzymes[217] and protect against UV-induced aging. Mice fed a solution containing isoflavones (primarily genistein and daidzein) and chronically exposed to UV for 4 weeks exhibited significant decreases in skin roughness measured by optical profilometry. In addition, epidermal thickness was significantly lower and the level of procollagen higher in the isoflavone-treated group. Dose-dependent decreases in MMP induction by UV radiation were also noted in an in vitro study of human fibroblasts treated with isoflavones.[205]

Table III. Compounds demonstrating efficacy in photoaging treatment in laboratory or animal systems

Type of intervention	Compound
Antioxidants	Soy-isoflavones[205]
	Genistein[206]
	N-Acetyl cysteine[206]
	Gluconolactone[53]
	Green tea polyphenols[207,208]
	N-Furfuryladenine (kinetin)[209,210]
	Dietary lutein[211]
	Pine tree extract[212]
Iron chelators	Kojic acid[213]
Anti-inflammatory agents	Hydrocortisone, naproxen, and ibuprofen[214]
	Celecoxib[215]
Novel compounds	Lipospondin[216]
	Oligodeoxynucleotides[79]

Topical genistein and N-acetyl cysteines. Topical genistein was shown to prevent c-Jun and collagenase up-regulation after UV exposure in human skin in vivo.[206] Beyond its antioxidant activity, genistein is an inhibitor of tyrosine kinase activity[218] and may inhibit signal transduction induced by UV light. Similar effects were found with the antioxidant N-acetyl cysteine, a precursor to glutathione.[206,219]

Gluconolactone. Gluconolactone is a polyhydroxy acid, related to AHAs such as glycolic acid. Gluconolactone has antioxidant properties while sharing in some of the effects of AHAs. Pretreatment with gluconolactone was shown to reduce UV induction of elastin by 50% in murine fibroblast cultures, potentially through its free-radical scavenger activity. Gluconolactone has already been incorporated into numerous cosmetic preparations,[53] apparently serving as a preventive treatment for solar elastosis.

Green tea polyphenols. Green tea polyphenols (GTPs) are potent antioxidants found in numerous skin care products.[220] Oral administration of GTPs markedly inhibited UV-induced expression of MMP in mouse skin, which suggests that GTP has a potential antiphotoaging effect.[207] Even in the absence of UV light, (−)-epigallocatechin-3-gallate, a component of green tea, was shown to inhibit the expression of various MMPs.[208]

N-Furfuryladenine. N-Furfuryladenine (kinetin) is a synthetic plant growth hormone with antioxidant properties. It has been shown to decrease or delay some of the age-related changes that occur in human fibroblasts during serial passage in cell culture.[209] It can also reduce ROS-mediated damage to DNA.[210] Currently, there are no published clinical studies of this compound available for review; however, it has been introduced into cosmeceuticals and may be useful in patients who are unable to tolerate retinoids.[156]

Other antioxidants. Dietary supplementation with lutein, a carotenoid, was shown to decrease UV-mediated inflammation and immunosuppression in a murine system.[211] An antioxidant extract from pine trees was shown to protect mice from inflammation, immunosuppression, and carcinogenesis induced by UV light when applied immediately after UV exposure.[212]

Iron chelators

Because MMP activation is dependent on iron,[96] the iron chelator kojic acid was investigated to determine its potential preventive effects on photoaging. Kojic acid is produced by the fungus *Aspergillus oryzae* and is found in Japanese soy-based products.[221] It has antioxidant properties[221] and is a tyrosinase inhibitor that has been used in the treatment of hyperpigmentation disorders, such as melasma.[222]

Pretreatment of mice with kojic acid before long-term UV exposure was found to reduce clinical assessments of wrinkling. Furthermore, UV-induced increases in dermal dermatan sulfate, chondroitin, epidermal hyperplasia, and dermal fibrosis were all reduced in the kojic acid–treated group when evaluated histologically. Kojic acid is currently incorporated into many Japanese cosmetic products.[213]

Anti-inflammatory agents

The protective effects of topical hydrocortisone, naproxen, and ibuprofen were examined in the hairless mouse. All 3 compounds significantly prevented wrinkling and increases in collagen damage, elastosis, and dermal cellularity in hairless mice exposed to UV over a long period.[214] Recently, celecoxib has been shown to reduce inflammation caused by short- and long-term UV exposure. Hairless mice treated with topical celecoxib have significant decreases in p53 activation and DNA damage 24-hours after UV exposure. When exposed to radiation over a long period, skin from celecoxib-treated mice demonstrated significant decreases in inflammatory markers, such as numbers of neutrophils, myeloperoxidase levels, and prostaglandin E_2.[215]

Novel compounds

Lipospondin. "Lipospondin" is a tripeptide linked to elaidic acid. It was designed to simultaneously activate latent TGF-β (through its peptide domain) and inhibit MMPs (through its lipophilic moiety, elaidic acid). It was able to up-regulate collagen and TIMP production and down-regulate MMP in fibroblast cultures; therefore "lipospondin" may show potential as a therapy for photoaging.[216]

Oligodeoxynucleotide technology. Oligodeoxynucleotide technology uses synthetic decoy *cis* elements to block the binding of transcription factors to promoter regions of target genes.[223] An NF-κB oligodeoxynucleotide has been developed and was shown to reduce UV-induced inflammatory changes (eg, swelling, leukocyte infiltration, epidermal hyperplasia, and accumulation of proinflammatory cytokines) when topically applied to mice.[79] This experiment focused on the role of NF-κB in sunburn. As NF-κB also has a role in MMP induction and photoaging, modification of this pathway may prove to have a future preventive role in photoaging.

SUMMARY

Like all organs, skin undergoes characteristic changes with age. In addition, photoaging due to UV radiation causes undesirable changes in skin appearance. Recent advances in skin biology have elucidated mechanisms by which photoaging occurs and have given rise to new treatments to prevent and reverse this process. There is currently a wide array of options available for those persons seeking to improve the appearance of their skin, with even more exciting treatments, including novel antioxidants, new compounds, and receptor-selective retinoids, on the horizon.

Many thanks to Dr Lauren Hammock for supplying the photomicrograph.

REFERENCES

1. Encyclopedia Britannica Premium Service. Aging. Encyclopedia Britannica. Accessed August 10, 2004.
2. Kligman AM, Graham JA. The psychology of appearance in the elderly. Clin Geriatr Med 1989;5:213-22.
3. Gonzalez-Ulloa M, Flores ES. Senility of the face—basic study to understand its causes and effects. Plast Reconstr Surg 1965;36:239-46.
4. Lavker RM, Zheng P, Dong G. Morphology of aged skin. Clin Geriatr Med 1989;5:53-67.
5. Kligman L. Photoaging: manifestations, prevention, and treatment. Clin Geriatr Med 1989;5:235-51.
6. Yaar M, Gilchrest BA. Aging of skin. In: Freedberg IM, Eisen AZ, Wolff K, Austen KF, Goldsmith LA, Katz S, editors. Fitzpatrick's dermatology in general medicine. New York: McGraw-Hill; 2003. pp. 1386-98.
7. Vaziri H, Benchimol S. From telomere loss to p53 induction and activation of a DNA-damage pathway at senescence: the telomere loss/DNA damage model of cell aging. Exp Gerontol 1996;31:295-301.
8. Campisi J. Replicative senescence: an old lives' tale? Cell 1996;84:497-500.
9. Yasui H, Sakurai H. Age-dependent generation of reactive oxygen species in the skin of live hairless rats exposed to UVA light. Exp Dermatol 2003;12:655-61.
10. Harman D. Aging: a theory based on free radical and radiation chemistry. J Gerontol 1956;11:298-300.
11. Tolmasoff JM, Ono T, Cutler RG. Superoxide dismutase: correlation with life-span and specific metabolic rate in primate species. Proc Natl Acad Sci U S A 1980;77:2777-81.
12. Hoppe U, Bergemann J, Diembeck W, Ennen J, Gohla S, Harris I, et al. Coenzyme Q10, a cutaneous antioxidant and energizer. BioFactors 1999;9:371-8.
13. Kosmadaki MG, Gilchrest BA. The role of telomeres in skin aging/photoaging. Micron 2004;35:155-9.
14. Chu DH, Haake AR, Holbrook K, Loomis CA. The structure and development of skin. In: Freedberg IM, Eisen AZ, Wolff K, Austen KF, Goldsmith LA, Katz S, editors. Fitzpatrick's dermatology in general medicine. New York: McGraw-Hill; 2003. pp. 58-88.
15. Stanulis-Praeger BM, Gilchrest BA. Growth factor responsiveness declines during adulthood for human skin-derived cells. Mech Ageing Dev 1986;35:185-98.
16. Reenstra WR, Yaar M, Gilchrest BA. Aging affects epidermal growth factor receptor phosphorylation and traffic kinetics. Exp Cell Res 1996;227:252-5.
17. Elias PM, Ghadially R. The aged epidermal permeability barrier: basis for functional abnormalities. Clin Geriatr Med 2002;18:103-20.
18. Yaar M, Gilchrest BA. Ageing and photoageing of keratinocytes and melanocytes. Clin Exp Dermatol 2001;26:583-91.
19. Bergstresser PR, Costner MI. Anatomy and physiology. In: Bolognia J, Jorizzo J, Rapini R, editors. Dermatology. St Louis: Mosby; 2003. pp. 25-38.
20. Gilchrest BA, Blog FB, Szabo G. Effects of aging and chronic sun exposure on melanocytes in human skin. J Invest Dermatol 1979;73:141-3.
21. Sunderkotter C, Kalden H, Luger T. Aging and the skin immune system. Arch Dermatol 1997;133:1256-62.
22. Sauder DN. Effect of age on epidermal immune function. Clin Geriatr Med 1989;5:149-60.
23. Edwards DR, Leco KJ, Beaudry PP, Atadja PW, Veillette C, Riabowol KT. Differential effects of transforming growth factor-beta 1 on the expression of matrix metalloproteinases and tissue inhibitors of metalloproteinases in young and old human fibroblasts. Exp Gerontol 1996;31:207-23.
24. Uitto J, Fazio MJ, Olsen DR. Molecular mechanisms of cutaneous aging: age associated connective tissue alterations in the dermis. J Am Acad Dermatol 1989;21:614-22.
25. Bentley JP. Aging of collagen. J Invest Dermatol 1979;73:80-3.
26. Varadi DP. Studies on the chemistry and fine structure of elastic fibers from normal adult skin. J Invest Dermatol 1972; 59:238-46.
27. Lewis KG, Bercovitch L, Dill SW, Robinson-Bostom L. Acquired disorders of elastic tissue: part I. Increased elastic tissue and solar elastotic syndromes. J Am Acad Dermatol 2004;51:1-21.
28. Hornebeck W. Down-regulation of tissue inhibitor of matrix metalloprotease-1 (TIMP-1) in aged human skin contributes to matrix degradation and impaired cell growth and survival. Pathol Biol 2003;51:569-73.
29. Hughes VA, Roubenoff R, Wood M, Frontera WR, Evans WJ, Fiatarone Singh MA. Anthropometric assessment of 10-y changes in body composition in the elderly. Am J Clin Nutr 2004;80:475-82.
30. Wong G, Gupta R, Dixon KM, Deo SS, Choong SM, Halliday GM, et al. 1,25-Dihydroxyvitamin D and three low-calcemic analogs decrease UV-induced DNA damage via the rapid response pathway. J Steroid Biochem Mol Biol 2004;89-90: 567-70.
31. MacLaughlin J, Holick MF. Aging decreases the capacity of human skin to produce vitamin D3. J Clin Invest 1985;76: 1536-8.

32. Brincat MP. Hormone replacement therapy and the skin. Maturitas 2000;29:107-17.
33. Dunn LB, Damesyn M, Moore A, Reuben DB, Greendale GA. Does estrogen prevent skin aging? Arch Dermatol 1997;133: 339-42.
34. Ashcroft GS, Dodsworth J, Van Boxtel E, Tarnuzzer RW, Horan MA, Schultz GS, et al. Estrogen accelerates cutaneous wound healing associated with an increase in TGF-beta1 levels. Nat Med 1997;3:1209-15.
35. Besne I, Descombes C, Breton L. Effect of age and anatomical site on density of sensory innervation in human epidermis. Arch Dermatol 2002;138:1445-50.
36. Young AJ. Effects of aging on human cold tolerance. Exp Aging Res 1991;17:205-13.
37. Gosain A, DiPietro LA. Aging and wound healing. World J Surg 2004;28:321-6.
38. Goukassian D, Gad F, Yaar M, Eller M, Nehal U, Gilchrest B. Mechanisms and implications of the age-associated decrease in DNA repair capacity. FASEB J 2000;14:1325-34.
39. Francis MK, Appel S, Meyer C, Balin SJ, Balin AK, Cristofalo VJ. Loss of EPC-1/PEDF expression during skin aging in vivo. J Invest Dermatol 2004;122:1096-105.
40. Kligman A, Kligman L. Photoaging. In: Freedberg IM, Eisen AZ, Wolff K, Austen F, Goldsmith LA, Katz SI, et al, editors. Fitzpatrick's dermatology in general medicine. New York: McGraw-Hill; 1999. pp. 1717-23.
41. Wlaschek M, Tantcheva-Poor I, Naderi L, Ma W, Schneider LA, Razi-Wolf Z, et al. Solar UV irradiation and dermal photoaging. J Photochem Photobiol B 2001;63:41-51.
42. Gilchrest BA, Yaar M. Ageing and photoageing of the skin: observations at the cellular and molecular level. Br J Dermatol 1992;127:25-30.
43. Leyden JJ. Clinical features of aging skin. Br J Dermatol 1990;122:1-3.
44. Lavker RM. Cutaneous aging: chronologic versus photoaging. In: Gilchrest BA, editor. Photodamage. Cambridge: Blackwell Science; 1995. pp. 123-35.
45. Yeatman JM, Kilkenny M, Marks R. The prevalence of seborrhoeic keratoses in an Australian population: does exposure to sunlight play a part in their frequency? Br J Dermatol 1997;137:411-4.
46. Gilchrest BA. A review of skin ageing and its medical therapy. Br J Dermatol 1996;135:867-75.
47. El Domyati M, Attia S, Saleh F, Brown D, Birk DE, Gasparro F, et al. Intrinsic aging vs. photoaging: a comparative histopathological, immunohistochemical, and ultrastructural study of skin. Exp Dermatol 2002;11:398-405.
48. Lavker RM, Kligman AM. Chronic heliodermatitis: a morphologic evaluation of chronic actinic damage with emphasis on the role of mast cells. J Invest Dermatol 1988;90: 325-30.
49. Varani J, Schuger L, Dame MK, Leonard C, Fligiel SEG, Kang S, et al. Reduced fibroblast interaction with intact collagen as a mechanism for depressed collagen synthesis in photodamaged skin. J Invest Dermatol 2004;122:1471-9.
50. Varani J, Spearman D, Perone P, Fligiel SEG, Datta SC, Wang ZQ, et al. Inhibition of type I procollagen synthesis by damaged collagen in photoaged skin and by collagenase-degraded collagen in vitro. Am J Pathol 2001;158: 931-42.
51. Bernstein EF, Brown D, Urbach F, Forbes D, DelMonaco M, Wu M, et al. Ultraviolet radiation activates the human elastin promoter in transgenic mice: a novel in vivo and in vitro model of cutaneous photoaging. J Invest Dermatol 1995; 105:269-73.
52. Kligman AM, Zheng P, Lavker RM. The anatomy and pathogenesis of wrinkles. Br J Dermatol 1985;113:37-42.
53. Bernstein EF, Brown DB, Schwartz MD, Kaidbey K, Ksenzenko SM. The polyhydroxy acid gluconolactone protects against ultraviolet radiation in an in vitro model of cutaneous photoaging. Dermatol Surg 2004;30:189-96.
54. Kochevar IE. Molecular and cellular effects of UV radiation relevant to chronic photodamage. In: Gilchrest BA, editor. Photodamage. Cambridge: Blackwell Science; 1995. pp. 51-67.
55. Tornaletti S, Pfeifer GP. UV damage and repair mechanisms in mammalian cells. BioEssays 1996;18:221-8.
56. Linge C. Relevance of in vitro melanocytic cell studies to the understanding of melanoma. Cancer Surv 1996;26:71-87.
57. Ichihashi M, Ueda M, Budiyanto A, Bito T, Oka M, Fukunaga M, et al. UV-induced skin damage. Toxicology 2003;189: 21-39.
58. Mitchell DL, Jen J, Cleaver JE. Relative induction of cyclobutane dimers and cytosine photohydrates in DNA irradiated in vitro and in vivo with ultraviolet-C and ultraviolet-B light. Photochem Photobiol 1991;54:741-6.
59. Bissett DL, Hannon DP, Orr TV. Wavelength dependence of histological, physical, and visible changes in chronically UV-irradiated hairless mouse skin. Photochem Photobiol 1989;50:763-9.
60. Kligman L, Sayre RM. An action spectrum for ultraviolet induced elastosis in hairless mice: quantification of elastosis by image analysis. Photochem Photobiol 1991;53:237-42.
61. Cadet J, Douki T, Pouget J-P, Ravanat J-L. Singlet oxygen DNA damage products: formation and measurement. Methods Enzymol 2000;319:143-53.
62. Fisher GJ, Kang S, Varani J, Bata-Csorgo Z, Wan Y, Datta S, et al. Mechanisms of photoaging and chronological skin aging. Arch Dermatol 2002;138:1462-70.
63. Berlett BS, Stadtman ER. Protein oxidation in aging, disease, and oxidative stress. J Biol Chem 1997;272:20313-6.
64. de Gruijl FR. Photocarcinogenesis: UVA vs UVB. Methods Enzymol 2000;319:359-66.
65. Hanson K, Simon J. Epidermal trans-urocanic acid and the UV-A-induced photoaging of the skin. Proc Natl Acad Sci U S A 1998;96:10576-8.
66. Menon EL, Morrison H. Formation of singlet oxygen by urocanic acid by UVA irradiation and some consequences thereof. Photochem Photobiol 2002;75:565-9.
67. Berneburg M, Grether-Beck S, Kurten V, Ruzicka T, Briviba K, Sies H, et al. Singlet oxygen mediates the UVA-induced generation of the photoaging-associated mitochondrial common deletion. J Biol Chem 1999;274:15345-9.
68. Berneburg M, Plettenberg H, Medve-Konig K, Pfahlberg A, Gers-Barlag H, Gefeller O, et al. Induction of the photoaging-associated mitochondrial common deletion in vivo in normal human skin. J Invest Dermatol 2004;122:1277-83.
69. Birch-Machin MA, Tindall M, Turner R, Haldane F, Rees JL. Mitochondrial DNA deletions in human skin reflect photo- rather than chronologic aging. J Invest Dermatol 1998;110: 149-52.
70. Krutmann J. Ultraviolet A radiation-induced biological effects in human skin: relevance for photoaging and photodermatosis. J Dermatol Sci 2000;23(Suppl):S22-6.
71. Mamelak AJ, Kowalski J, Murphy K, Yadava N, Zahurak M, Kouba DJ, et al. Downregulation of NDUFA1 and other oxidative phosphorylation related genes is a consistent feature of basal cell carcinoma. Exp Dermatol 2005;14:336-48.
72. Soter NA. Sunburn and suntan: immediate manifestations of photodamage. In: Gilchrest BA, editor. Photodamage. Cambridge (MA): Blackwell Science; 1995. pp. 12-25.

73. Halaban R, Hebert DN, Fisher GJ. Biology of melanocytes. In: Freedberg IM, Eisen AZ, Wolff K, Austen F, Goldsmith LA, Katz SI, editors. Fitzpatrick's dermatology in general medicine. New York: McGraw-Hill; 2003. pp. 127-48.
74. Braun-Falco O, Schoefinius HH. Lentigo senilis. Hautarzt 1971; 22:277-83.
75. Hodgson C. Lentigo senilis. Arch Dermatol 1963;87:197-207.
76. Montagna W, Hu F, Carlisle K. A reinvestigation of solar lentigines. Arch Dermatol 1980;116:1151-4.
77. Ortonne J-P, Perrot H. Idiopathic guttate hypomelanosis. Ultrastructural study. Arch Dermatol 1980;116:664-8.
78. Wilson PD, Lavker RM, Kligman AM. On the nature of idiopathic guttate hypomelanosis. Acta Derm Venerol 1982;62:301-6.
79. Abeyama K, Eng W, Jester JV, Vink AA, Edelbaum D, Cockerell CJ, et al. A role for NF-κB-dependent gene transactivation in sunburn. J Clin Invest 2000;105:1751-9.
80. Kosmadaki MG, Yaar M, Arble BL, Gilchrest BA. UV induces VEGF through a TNF-alpha independent pathway. FASEB J 2003;17:446-8.
81. Yano K, Kajiya K, Ishiwata M, Hong YK, Miyakawa T, Detmar M. Ultraviolet B-induced skin angiogenesis is associated with a switch in the balance of vascular endothelial growth factor and thrombospondin-1 expression. J Invest Dermatol 2004; 122:201-8.
82. Howell BG, Wang B, Freed I, Mamelak AJ, Watanabe H, Sauder DN. Microarray analysis of UVB-regulated genes in keratinocytes: downregulation of angiogenesis inhibitor thrombospondin-1. J Dermatol Sci 2004;34:185-94.
83. Aubin F. Mechanisms involved in ultraviolet light-induced immunosuppression. Eur J Dermatol 2003;13:515-23.
84. Kripke ML. Immunology and photocarcinogenesis: new light on an old problem. J Am Acad Dermatol 1986;14:149-55.
85. Toews GB, Bergstresser P, Streilein JW. Epidermal Langerhans cell density determines whether contact hypersensitivity or unresponsiveness follows skin painting with DNFB. J Immunol 1980;124:445-53.
86. Noonan FP, Kripke ML, Pedersen GM, Greene MI. Suppression of contact hypersensitivity in mice by ultraviolet irradiation is associated with defective antigen presentation. Immunology 1981;43:527-33.
87. Molendijk A, van Gurp RJ, Donselaar IG, Benner R. Suppression of delayed-type hypersensitivity to histocompatibility antigens by ultraviolet radiation. Immunology 1987;62: 299-305.
88. Vink AA, Moodycliffe AM, Shreedhar V, Ullrich SE, Roza L, Yarosh DB, et al. The inhibition of antigen-presenting activity of dendritic cells resulting from UV irradiation of murine skin is restored by in vitro photorepair of cyclobutane pyrimidine dimers. Proc Natl Acad Sci U S A 1997; 94:5255-60.
89. Kang K, Hammerberg C, Meunier L, Cooper KD. CD11b+ macrophages that infiltrate human epidermis after in vivo ultraviolet exposure potently produce IL-10 and represent the major secretory source of epidermal IL-10 protein. J Immunol 1994;153:5256-64.
90. Weiss E, Mamelak AJ, La Morgia S, Wang B, Feliciani C, Tulli A, et al. The role of interleukin 10 in the pathogenesis and potential treatment of skin diseases. J Am Acad Dermatol 2004;50:657-75.
91. Heck DE, Gerecke DR, Vetrano AM, Laskin JD. Solar ultraviolet radiation as a trigger of cell signal transduction. Toxicol Appl Pharmacol 2004;195:288-97.
92. Gross S, Knebel A, Tenev T, Neininger A, Gaestel M, Herrlich P, et al. Inactivation of protein-tyrosine phosphatases as mechanism of UV-induced signal transduction. J Biol Chem 1999;274:26378-86.
93. Barford D, Jia Z, Tonks NK. Protein tyrosine phosphatases take off. Nat Struct Biol 1995;2:1043-53.
94. Fisher GJ, Talwar HS, Lin J, Lin P, McPhillips F, Wang Z, et al. Retinoic acid inhibits induction of c-jun protein by ultraviolet radiation that occurs subsequent to activation of mitogen-activated protein kinase pathways in human skin in vivo. J Clin Invest 1998;101:1432-40.
95. Fisher GJ, Wang Z, Datta SC, Varani J, Kang S, Voorhees JJ. Pathophysiology of premature skin aging induced by ultraviolet light. N Engl J Med 1997;337:1419-29.
96. Polte T, Tyrrell RM. Involvement of lipid peroxidation and organic peroxides in UVA-induced matrix metalloproteinase-1 expression. Free Radic Biol Med 2004;36:1566-74.
97. Brennan M, Bhatti H, Nerusu KC, Bhagavathula N, Kang S, Fisher GJ, et al. Matrix metalloproteinase-1 is the major collagenolytic enzyme responsible for collagen damage in UV-irradiated human skin. Photochem Photobiol 2003;78: 43-8.
98. Reelfs O, Tyrrell RM, Pourzand C. Ultraviolet A radiation-induced immediate iron release is a key modulator of the activation of NF-κB in human skin fibroblasts. J Invest Dermatol 2004;122:1440-7.
99. Kang S, Fisher GJ, Voorhees JJ. Photoaging: pathogenesis, prevention, and treatment. Clin Geriatr Med 2001;17: 643-59.
100. Fisher GJ, Datta S, Talwar HS, Wang Z, Varani J, Voorhees JJ. Molecular basis of sun-induced premature skin ageing and retinoid antagonism. Nature 1996;379:335-9.
101. Talwar HS, Griffiths CEM, Fisher GJ, Hamilton TA, Voorhees JJ. Reduced type I and type III procollagens in photodamaged adult human skin. J Invest Dermatol 1995;105:285-90.
102. Fisher GJ, Datta S, Wang Z, Li XY, Quan T, Chung JH, et al. c-Jun-dependent inhibition of cutaneous procollagen transcription following ultraviolet irradiation is reversed by all-trans retinoic acid. J Clin Invest 2000;106:663-70.
103. Quan T, He T, Kang S, Voorhees JJ, Fisher GJ. Ultraviolet irradiation alters transforming growth factor beta/smad pathway in human skin in vivo. J Invest Dermatol 2002;119: 499-506.
104. Penttinen RP, Kobayashi S, Bornstein P. Transforming growth factor beta increases mRNA for matrix proteins both in the presence and in the absence of changes in mRNA stability. Proc Natl Acad Sci U S A 1988;85:1105-8.
105. Chung KY, Agarwal A, Uitto J, Mauviel A. An AP-1 binding sequence is essential for regulation of the human alpha2(I) collagen (COL1A2) promoter activity by transforming growth factor-beta. J Biol Chem 1996;271:3272-8.
106. Varani J, Perone P, Fligiel SEG, Fisher GJ, Voorhees JJ. Inhibition of type I procollagen production in photodamage: correlation between presence of high molecular weight collagen fragments and reduced procollagen synthesis. J Invest Dermatol 2002;119:122-9.
107. Fligiel SEG, Varani J, Datta SC, Kang S, Fisher GJ, Voorhees JJ. Collagen degradation in aged/photodamaged skin in vivo and after exposure to matrix metalloproteinase-1 in vitro. J Invest Dermatol 2003;120:842-8.
108. Nagpal S, Chandraratna RAS. Recent developments in receptor-selective retinoids. Curr Pharm Des 2000;6:919-31.
109. Nagpal S, Athanikar J, Chandraratna RAS. Separation of transactivation and AP1 antagonism functions of retinoic acid receptor alpha. J Biol Chem 1995;270:923-7.
110. Wang Z, Boudjelal M, Kang S, Voorhees JJ, Fisher GJ. Ultraviolet irradiation of human skin causes functional

vitamin A deficiency, preventable by all-trans retinoic acid pre-treatment. Nat Med 1999;5:418-22.
111. Andersson E, Rosdahl I, Torma H, Vahlquist A. Differential effects of UV irradiation on nuclear retinoid receptor levels in cultured keratinocytes and melanocytes. Exp Dermatol 2003;12:563-71.
112. Berne B, Nilsson M, Vahlquist A. UV irradiation and cutaneous vitamin A. An experimental study in rabbit and human skin. J Invest Dermatol 1984;83:401-4.
113. Rijken F, Bruijnzeel PLB, van Weelden H, Kiekens RCM. Responses of black and white skin to solar-simulating radiation: differences in DNA photodamage, infiltrating neutrophils, proteolytic enzymes induced, keratinocyte activation, and IL-10 expression. J Invest Dermatol 2004;122: 1448-55.
114. Matsumura Y, Ananthaswamy HN. Toxic effects of ultraviolet radiation on the skin. Toxicol Appl Pharmacol 2004;195: 298-308.
115. Shindo Y, Witt E, Han D, Epstein W, Packer L. Enzymic and non-enzymic antioxidants in epidermis and dermis of human skin. J Invest Dermatol 1994;102:122-4.
116. Epstein JH. Effects of ultraviolet radiation on the mitotic cycle and DNA, RNA and protein synthesis in mammalian epidermis in vivo. Photochem Photobiol 1970;12:57-65.
117. Everett MA. Protection from sunlight in vitiligo. Arch Dermatol 1961;84:997-8.
118. Szabo G. Pigment cell biology. In: Gordon M, editor. Mitochondria and other cytoplasmic inclusions. New York: Academic Press; 1959.
119. Kaidbey K, Agin PP, Sayre RM, Kligman AM. Photoprotection by melanin—a comparison of black and Caucasian skin. J Am Acad Dermatol 1979;1:249-60.
120. Huang LC, Clarkin KC, Wahl GM. Sensitivity and selectivity of the DNA damage sensor responsible for activating p53-dependent G1 arrest. Proc Natl Acad Sci U S A 1996;93: 4827-32.
121. Cleaver JE, Cortes F, Karentz D, Lutze L, Morgan WF, Player AN, et al. The relative importance of cyclobutane and (6-4) pyrimidine-pyrimidone dimer photoproducts in human cells: evidence from a xeroderma pigmentosum revertant. Photochem Photobiol 1988;48:41-9.
122. Young LC, Hays JB, Tron VA, Andrew SE. DNA mismatch repair proteins: potential guardians against genomic instability and tumorigenesis induced by ultraviolet photoproducts. J Invest Dermatol 2003;121:435-40.
123. Oh JH, Chung AS, Steinbrenner H, Sies H, Brenneisen P. Thioredoxin secreted upon ultraviolet A irradiation modulates activities of matrix metalloproteinase-2 and tissue inhibitor of metalloproteinase-2 in human dermal fibroblasts. Arch Biochem Biophys 2004;423:218-26.
124. Hata TR, Scholz TA, Ermakov IV, McClane RW, Khachik F, Gellermann W, et al. Non-invasive raman spectroscopic detection of carotenoids in human skin. J Invest Dermatol 2000;115:441-8.
125. Leccia MT, Yaar M, Allen N, Gleason M, Gilchrest BA. Solar simulated irradiation modulates gene expression and activity of antioxidant enzymes in cultured human dermal fibroblasts. Exp Dermatol 2001;10:272-9.
126. Fuchs J. Potentials and limitations of the natural antioxidants RRR-alpha-tocopherol, L-ascorbic acid and beta-carotene in cutaneous photoprotection. Free Radic Biol Med 1998;25: 848-73.
127. Shindo Y, Hashimoto T. Time course of changes in antioxidant enzymes in human skin fibroblasts after UVA irradiation. J Dermatol Sci 1997;14:225-32.
128. Matzen RN. Preventive medicine: definition and application. In: Lang RS, Hensrud DD, editors. Clinical preventive medicine. AMA Press; 2004. pp. 3-9.
129. Guyatt GH, Sackett DL, Sinclair JC, Hayward R, Cook D, Cook RJ. User's guide to the medical literature IX. A method for grading health care recommendations. JAMA 1995;274:1800-4.
130. Cochrane Reviewers' Handbook 2.2.2 [updated March 2004]. In: The Cochrane Library, Issue 1, 2004. Chichester (UK): John Wiley and Sons Ltd; 2004.
131. Weiss JS, Ellis CN, Headington JT, Tincoff T, Hamilton TA, Voorhees JJ. Topical tretinoin improves photodamaged skin: a double-blind, vehicle-controlled study. JAMA 1988;259: 527-32.
132. Weinstein GD, Nigra TP, Pochi PE, Savin RC, Allan A, Benik K, et al. Topical tretinoin for treatment of photodamaged skin. A multicenter study. Arch Dermatol 1991;127:659-65.
133. Kang S, Leyden JJ, Lowe NJ, Ortonne JP, Phillips TJ, Weinstein GD, et al. Tazarotene cream for the treatment of facial photodamage: a multicenter, investigator-masked, randomized, vehicle-controlled, parallel comparison of 0.01%, 0.025%, 0.05%, and 0.1% tazarotene creams with 0.05% tretinoin emollient cream applied once daily for 24 weeks. Arch Dermatol 2001;137:1597-604.
134. Phillips TJ, Gottlieb AB, Leyden JJ, Lowe NJ, Lew-Kaya DA, Sefton J, et al. Efficacy of 0.1% tazarotene cream for the treatment of photodamage: a 12-month multicenter, randomized trial. Arch Dermatol 2002;138:1486-93.
135. Humbert PG, Haftek M, Creidi P, Lapiere C, Nusgens B, Richard A, et al. Topical ascorbic acid on photoaged skin. Clinical, topographical and ultrastructural evaluation: double-blind study vs. placebo. Exp Dermatol 2003;12:237-44.
136. Murad H, Tabibian MP. The effect of an oral supplement containing glucosamine, amino acids, minerals, and antioxidants on cutaneous aging: a preliminary study. J Dermatolog Treat 2001;12:47-51.
137. Greul AK, Grundmann JU, Heinrich F, Pfitzner I, Bernhardt J, Ambach A, et al. Photoprotection of UV-irradiated human skin: an antioxidative combination of vitamins E and C, carotenoids, selenium and proanthocyanidins. Skin Pharmacol Appl Skin Physiol 2002;15:307-15.
138. Beitner H. Randomized, placebo-controlled, double blind study on the clinical efficacy of a cream containing 5% alpha-lipoic acid related to photoageing of facial skin. Br J Dermatol 2003;149:841-9.
139. Sauerbronn AVD, Fonseca AM, Bagnoli VR, Saldiva PH, Pinotti JA. The effects of systemic hormonal replacement therapy on the skin of postmenopausal women. Int J Gynaecol Obstet 2000;68:35-41.
140. Brincat MP, Versi E, O'Dowd T, Moniz CF, Magos A, Kabalan S, et al. Skin collagen changes in post menopausal women receiving oestradiol gel. Maturitas 1987;9:1-5.
141. Schmidt JB, Binder M, Demschik G, Bieglmayer C, Reiner A. Treatment of skin aging with topical estrogens. Int J Dermatol 1996;35:669-74.
142. Fitzpatrick RE, Rostan EF. Reversal of photodamage with topical growth factors: a pilot study. J Cosmet Laser Ther 2003;5:25-34.
143. Robert C, Robert AM, Robert L. Effect of a fucose-rich polysaccharide preparation on the age-dependent evolution of the skin surface micro-relief. Pathol Biol 2003;51:586-90.
144. Bauza E, Dal Farra C, Berghi A, Oberto G, Peyronel D, Domloge N. Date palm kernel extract exhibits antiaging properties and significantly reduces skin wrinkles. Int J Tissue React 2002;24:131-6.

145. Newman N, Newman A, Moy LS, Babapour R, Harris AG, Moy RL. Clinical improvement of photoaged skin with 50% glycolic acid: a double-blind vehicle-controlled study. Dermatol Surg 1996;22:455-60.
146. Freedman BM, Rueda-Pedraza E, Waddell SP. The epidermal and dermal changes associated with microdermabrasion. Dermatol Surg 2001;27:1031-4.
147. Abraham MT, Keller GS, Pinkosky G, Feibleman CE, Kelly J, Man D, et al. Microcoblation: nonablative skin rejuvenation. Facial Plast Surg 2004;20:51-6.
148. Weiss RA, Harrington AC, Pfau RC, Weiss MA, Marwaha S. Periorbital skin resurfacing using high energy erbium:YAG laser: results in 50 patients. Lasers Surg Med 1999;24: 81-6.
149. Tanzi EL, Williams CM, Alster TS. Treatment of facial rhytides with a nonablative 1,450-nm diode laser: a controlled clinical and histologic study. Dermatol Surg 2003;29:124-8.
150. Alster TS, Tanzi E. Improvement of neck and cheek laxity with a nonablative radiofrequency device: a lifting experience. Dermatol Surg 2004;30:503-7.
151. Carruthers JA, Lowe NJ, Menter MA, Gibson J, Nordquist M, Mordaunt J, et al. A multicenter, double-blind, randomized, placebo-controlled study of the efficacy and safety of botulinum toxin type A in the treatment of glabellar lines. J Am Acad Dermatol 2002;46:840-9.
152. Baumann L, Slezinger A, Vujevich J, Halem M, Bryde J, Black L, et al. A double-blinded, randomized, placebo-controlled pilot study of the safety and efficacy of Myobloc (botulinum toxin type B)-purified neurotoxin complex for the treatment of crow's feet: a double-blinded, placebo-controlled trial. Dermatol Surg 2003;29:508-15.
153. Narins RS, Brandt F, Leyden J, Lorenc ZP, Rubin M, Smith S. A randomized, double-blind, multicenter comparison of the efficacy and tolerability of Restylane versus Zyplast for the correction of nasolabial folds. Dermatol Surg 2003;29: 588-95.
154. Sclafani AP, Romo T 3rd, Jacono AA, McCormick S, Cocker R, Parker A. Evaluation of acellular dermal graft in sheet (AlloDerm) and injectable (Micronized AlloDerm) forms for soft tissue augmentation: clinical observations and histological analysis. Arch Facial Plast Surg 2000;2:130-6.
155. Sclafani AP, Romo T 3rd, Jacono AA, McCormick SA, Cocker R, Parker A. Evaluation of acellular dermal graft (AlloDerm) sheet for soft tissue augmentation: a 1-year follow-up of clinical observations and histological findings. Arch Facial Plast Surg 2001;3:101-3.
156. Glaser DA. Anti-aging products and cosmeceuticals. Facial Plast Surg Clin North Am 2003;11:219-27.
157. Morison WL. Photoprotection by clothing. Dermatol Ther 2003; 16:16-22.
158. Diffey BL. Sun protection with clothing. Br J Dermatol 2001; 144:449-51.
159. Seite S, Colige A, Piquemal-Vivenot P, Montastier C, Fourtanier A, Lapiere C, et al. A full-UV spectrum absorbing daily use cream protects human skin against biological changes occurring in photoaging. Photodermatol Photoimmunol Photomed 2000;16:147-55.
160. Naylor M, Gasparro F. What you and your patients should know about sunscreens. Skin Aging 1998;6:44-50.
161. Pinnell SR, Fairhurst D, Gillies R, Mitchnick MA, Kollias N. Microfine zinc oxide is a superior sunscreen ingredient to microfine titanium dioxide. Dermatol Surg 2000;26: 309-14.
162. Food and Drug Administration Final Sunscreen Monograph. 64 Federal Register (98):27691-2. May 21, 1999.
163. Haywood R, Wardman P, Sanders R, Linge C. Sunscreens inadequately protect against ultraviolet-A-induced free radicals in skin: implications for skin aging and melanoma? J Invest Dermatol 2003;121:862-8.
164. Takeuchi T, Uitto J, Bernstein EF. A novel in vivo model for evaluating agents that protect against ultraviolet A-induced photoaging. J Invest Dermatol 1998;110:343-7.
165. Kligman L, Akin FJ, Kligman AM. Prevention of ultraviolet damage to the dermis of hairless mice by sunscreens. J Invest Dermatol 1982;78:181-9.
166. Kligman L, Akin FJ, Kligman AM. Sunscreens promote repair of ultraviolet radiation-induced dermal damage. J Invest Dermatol 1983;81:98-102.
167. Levin AA, Sturzenbecker LJ, Kazmer S, Bosakowski T, Huselton C, Allenby G, et al. 9-cis Retinoic acid stereoisomer binds and activates the nuclear receptor RXR alpha. Nature 1992; 355:359-61.
168. Kang S, Voorhees JJ. Topical retinoids. In: Freedberg IM, Eisen AZ, Wolff K, Austen F, Goldsmith LA, Katz SI, editors. Fitzpatrick's dermatology in general medicine. New York: McGraw-Hill; 2003. pp. 2328-34.
169. Olsen EA, Katz HI, Levine N, Nigra TP, Pochi PE, Savin RC, et al. Tretinoin emollient cream for photodamaged skin: results of 48-week, multicenter, double-blind studies. J Am Acad Dermatol 1997;37:217-26.
170. Griffiths C, Russman AN, Majmudar G, Singer RS, Hamilton TA, Voorhees JJ. Restoration of collagen formation in photodamaged human skin by tretinoin (retinoic acid). N Engl J Med 1993;329:530-5.
171. Sefton J, Kligman AM, Kopper SC, Lue JC, Gibson JR. Photodamage pilot study: a double-blind, vehicle-controlled study to assess the efficacy and safety of tazarotene 0.1% gel. J Am Acad Dermatol 2000;43:656-63.
172. Colven RM, Pinnell SR. Topical vitamin C in aging. Clin Dermatol 1996;14:227-34.
173. Lin JY, Selim MA, Shea CR, Grichnik JM, Omar MM, Monteiro-Riviere NA, et al. UV photoprotection by combination topical antioxidants vitamin C and vitamin E. J Am Acad Dermatol 2003;48:866-74.
174. Nusgens BV, Humbert P, Rougier A, Colige AC, Haftek M, Lambert CA, et al. Topically applied vitamin C enhances the mRNA level of collagens I and III, their processing enzymes and tissue inhibitor of matrix metalloproteinase 1 in the human dermis. J Invest Dermatol 2001;116:853-9.
175. Greco RJ. Topical vitamin C. Plast Reconstr Surg 2000;105: 464-5.
176. Fischer TW, Wigger-Alberti W, Elsner P. Direct and non-direct measurement techniques for analysis of skin surface topography. Skin Pharmacol Appl Skin Physiol 1999;12:1-11.
177. Suzuki YJ, Aggarwal BB, Packer L. Alpha-lipoic acid is a potent inhibitor of NF-kappa B activation in human T cells. Biochem Biophys Res Commun 1992;189:1709-15.
178. Isnard N, Fodil-Bourahla I, Robert AM, Robert L. Pharmacology of skin aging. Stimulation of glycosaminoglycan biosynthesis by L-fucose and fucose-rich polysaccharides, effect of in vitro aging of fibroblasts. Biomed Pharmacother 2004;58: 202-4.
179. Voros E, Robert C, Robert AM. Age-related changes of the human skin surface microrelief. Gerontology 1990;36:276-85.
180. Ghersetich I, Brazzini B, Peris K, Cotellessa C, Manunta T, Lotti T. Pyruvic acid peels for the treatment of photoaging. Dermatol Surg 2004;30:32-6.
181. Kauvar ANB, Dover JS. Facial skin rejuvenation: laser resurfacing or chemical peel: choose your weapon. Dermatol Surg 2001;27:209-12.

182. Brody HJ. Chemical peeling: an updated review. J Cutan Med Surg 1999;3:14-20.
183. Stiller MJ, Bartolone J, Stern R, Smith S, Kollias N, Gillies R, et al. Topical 8% glycolic acid and 8% L-lactic acid creams for the treatment of photodamaged skin. A double-blind vehicle-controlled clinical trial. Arch Dermatol 1996;132:631-6.
184. Kaidbey K, Sutherland B, Bennett P, Wamer WG, Barton C, Dennis D, et al. Topical glycolic acid enhances photodamage by ultraviolet light. Photodermatol Photoimmunol Photomed 2003;19:21-7.
185. Karimipour DJ, Kang S, Johnson TM, Orringer JS, Hamilton T, Hammerberg C, et al. Microdermabrasion: a molecular analysis following a single treatment. J Am Acad Dermatol 2005; 52:215-23.
186. Coimbra M, Rohrich RJ, Chao J, Brown SA. A prospective controlled assessment of microdermabrasion for damaged skin and fine rhytides. Plast Reconstr Surg 2004;113:1438-43.
187. Tanzi EL, Lupton JR, Alster TS. Lasers in dermatology: four decades of progress. J Am Acad Dermatol 2003;49:1-31.
188. Anderson RR, Parrish JA. Selective photothermolysis: precise microsurgery by selective absorption of pulsed radiation. Science 1983;220:524-7.
189. Apfelberg DB. Ultrapulse carbon dioxide laser with CPG scanner for full-face resurfacing for rhytids, photoaging, and acne scars. Plast Reconstr Surg 1997;99:1817-25.
190. West TB, Alster TS. Improvement of infraorbital hyperpigmentation following carbon dioxide laser resurfacing. Dermatol Surg 1998;24:615-6.
191. Orringer JS, Kang S, Johnson TM, Karimipour DJ, Hamilton T, Hammerberg C, et al. Connective tissue remodeling induced by carbon dioxide laser resurfacing of photodamaged human skin. Arch Dermatol 2004;140:1326-32.
192. Kopera D, Smolle J, Kaddu S, Kerl H. Nonablative laser treatment of wrinkles: meeting the objective? Assessment by 25 dermatologists. Br J Dermatol 2004;150:936-9.
193. Zelickson BD, Kist D, Bernstein E, Brown DB, Ksenzenko S, Burns J, et al. Histological and ultrastructural evaluation of the effects of a radiofrequency-based nonablative dermal remodeling device: a pilot study. Arch Dermatol 2004;140: 204-9.
194. Fitzpatrick RE, Geronemus R, Goldberg D, Kaminer MS, Kilmer S, Ruiz-Esparza J. Multicenter study of noninvasive radiofrequency for periorbital tissue tightening. Laser Surg Med 2003;33:232-42.
195. Montecucco C, Schiavo G. Tetanus and botulinum neurotoxins: a new group of zinc proteases. Trends Biochem Sci 1993;18:324-7.
196. Vartanian AJ, Dayan SH. Facial rejuvenation using botulinum toxin A: review and updates. Facial Plast Surg 2004;20:11-9.
197. Setler P. The biochemistry of botulinum toxin type B. Neurology 2000;55(Suppl):S23-8.
198. Donofrio LM. Structural autologous lipoaugmentation: a panfacial technique. Dermatol Surg 2000;26:1129-34.
199. Matti BA, Nicolle FV. Clinical use of Zyplast in correction of age- and disease-related contour deficiencies of the face. Aesthetic Plast Surg 1990;14:227-34.
200. Stegman SJ, Chu S, Armstrong R. Adverse reactions to bovine collagen implant: clinical and histologic features. J Dermatol Surg Oncol 1988;14:39-48.
201. Manna F, Dentini M, Desideri P, De Pita O, Mortilla E, Maras B. Comparative chemical evaluation of two commercially available derivatives of hyaluronic acid (hylaform from rooster combs and restylane from streptococcus) used for soft tissue augmentation. J Eur Acad Dermatol Venereol 1999;13: 183-92.
202. Larsen NE, Pollak CT, Reiner K, Leshchiner E, Balazas EA. Hylan gel biomaterial: dermal and immunologic compatibility. J Biomed Mater Res 1993;27:1129-34.
203. Piacquadio D, Jarcho M, Goltz R. Evaluation of hylan b gel as a soft-tissue augmentation implant material. J Am Acad Dermatol 1997;36:544-9.
204. Lemperle G, Romano JJ, Busso M. Soft tissue augmentation with artecoll: 10-year history, indications, techniques, and complications. Dermatol Surg 2003;29:573-87.
205. Kim SY, Kim SJ, Lee JY, Kim WG, Park WS, Sim YC, et al. Protective effects of dietary soy isoflavones against UV-induced skin-aging in hairless mouse model. J Am Coll Nutr 2004;23:157-62.
206. Kang S, Chung JH, Lee JH, Fisher GJ, Wan YS, Duell EA, et al. Topical N-acetyl cysteine and genistein prevent ultraviolet-light-induced signaling that leads to photoaging in human skin in vivo. J Invest Dermatol 2003;120:835-41.
207. Vayalil PK, Mittal A, Hara Y, Elmets CA, Katiyar SK. Green tea polyphenols prevent ultraviolet light-induced oxidative damage and matrix metalloproteinases expression in mouse skin. J Invest Dermatol 2004;122:1480-7.
208. Dell'Aica I, Dona M, Sartor L, Pezzato E, Garbisa S. (-)Epigallocatechin-3-gallate directly inhibits MT1-MMP activity, leading to accumulation of nonactivated MMP-2 at the cell surface. Lab Invest 2002;82:1685-93.
209. Rattan S, Clark BFC. Kinetin delays the onset of aging characteristics in human fibroblasts. Biochem Biophys Res Commun 1994;201:665-72.
210. Olsen A, Siboska GE, Clark BFC, Rattan SIS. N(6)-Furfuryladenine, kinetin, protects against Fenton reaction-mediated oxidative damage to DNA. Biochem Biophys Res Commun 1999;265:499-502.
211. Lee EH, Faulhaber D, Hanson KM, Ding W, Peters S, Kodali S, et al. Dietary lutein reduces ultraviolet radiation-induced inflammation and immunosuppression. J Invest Dermatol 2004;122:510-7.
212. Sime S, Reeve VE. Protection from inflammation, immunosuppression and carcinogenesis induced by UV radiation in mice by topical Pycnogenol. Photochem Photobiol 2004;79: 193-8.
213. Mitani H, Koshiishi I, Sumita T, Imanari T. Prevention of the photodamage in the hairless mouse dorsal skin by kojic acid as an iron chelator. Eur J Pharmacol 2001;411: 169-74.
214. Bissett DL, Chatterjee R, Hannon DP. Photoprotective effect of topical anti-inflammatory agents against ultraviolet radiation-induced chronic skin damage in the hairless mouse. Photodermatol Photoimmunol Photomed 1990;7:153-8.
215. Wilgus TA, Koki AT, Zweifel BS, Kusewitt DF, Rubal PA, Oberyszn TM. Inhibition of cutaneous ultraviolet light B-mediated inflammation and tumor formation with topical celecoxib treatment. Mol Carcinog 2003;38:49-58.
216. Cauchard JH, Berton A, Godeau G, Hornebeck W, Bellon G. Activation of latent transforming growth factor beta 1 and inhibition of matrix metalloprotease activity by a thrombospondin-like tripeptide linked to elaidic acid. Biochem Pharmacol 2004;67:2013-22.
217. Cai Q, Wei H. Effect of dietary genistein on antioxidant enzyme activities in SENCAR mice. Nutr Cancer 1996;25: 1-7.
218. Wei H, Cai Q, Rahn RO. Inhibition of UV light- and Fenton reaction-induced oxidative DNA damage by the soybean isoflavone genistein. Carcinogenesis 1996;17:73-7.
219. De Vries N, De Flora S. N-acetyl l-cysteine. J Cell Biochem Suppl 1993;(Suppl 17F);270-8.

220. Katiyar SK, Elmets CA. Green tea polyphenolic antioxidants and skin photoprotection. Int J Oncol 2001;18: 1307-13.
221. Niwa Y, Akamatsu H. Kojic acid scavenges free radicals while potentiating leukocyte functions including free radical generation. Inflammation 1991;15:303-15.
222. Garcia A, Fulton JE. The combination of glycolic acid and hydroquinone or kojic acid for the treatment of melasma and related conditions. Dermatol Surg 1996;22:443-7.
223. Bielinska A, Shivdasani RA, Zhang L, Nabel GJ. Regulation of gene expression with double-stranded phosphorothioate. Science 1990;250:997-1000.