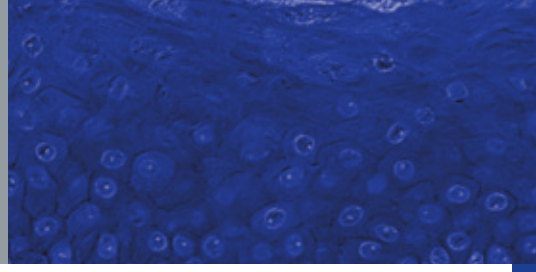
RETAMAX™

Active Vitamin A Micro-Emulsion



Multi-Vectored Mechanisms of Action for Skin Restoration and Rejuvenation

Synergistic Effects of Retinol, Botanical Agents, and Bioengineered Complexes

Summary

Skin aging is characterized by changes in skin structures and biochemistry. Retamax™ is scientifically formulated with components that have multiple mechanisms of action to improve skin structure and function.

Retamax™ includes retinol, which improves skin firmness, texture, complexion and reduces fine lines and wrinkles in general populations and in the elderly with an acceptable safety profile.

Also included is an antioxidant derived from bioengineered stem cells from the butterfly bush (*Buddleja davidii*). *Buddleja* extracts include the phenylpropanoid derivative verbascoside, which has antioxidant, anti-inflammatory, and photoprotective properties.

Ascorbyl palmitate (vitamin C) and alpha-tocopherol (vitamin E) provide antioxidant and anti-inflammatory properties.

Biomimetic hydrolyzed silk peptides (sericin) supports the skin's natural rebuilding process by stimulating the biosynthesis of collagen and hyaluronic acid naturally produced in the skin. Sericin has antioxidant properties and inhibits lipid peroxidation and tyrosinase activity, which may help even skin tone and reduce hyperpigmentation.

Retamax™ minimizes skin sensitivity with broccoli and sunflower seed oil extracts. Broccoli extracts inhibit cellular oxidative stress. Sunflower oil reduces transepidermal water loss and speeds barrier recovery after epidermal injury. It also reduces the formation of glycation end products.

Beta-glucan increases skin firmness and stratum corneum hydration, has anti-inflammatory properties, and provides additional humectants and emollient properties.

Retamax™ represents the ultimate utilization of science in the service of skin care by maximizing multiple mechanisms of action by combining retinol with clinically relevant botanical ingredients and bioengineered components to improve skin function and appearance.

EDITORS

Zein Obagi, MD
Board-Certified Dermatologist
Obagi Skin Health Institute
Beverly Hills, California

Cheryl M. Burgess, MD, FAAD
Center for Dermatology and
Dermatologic Surgery
Washington, D.C.

Introduction

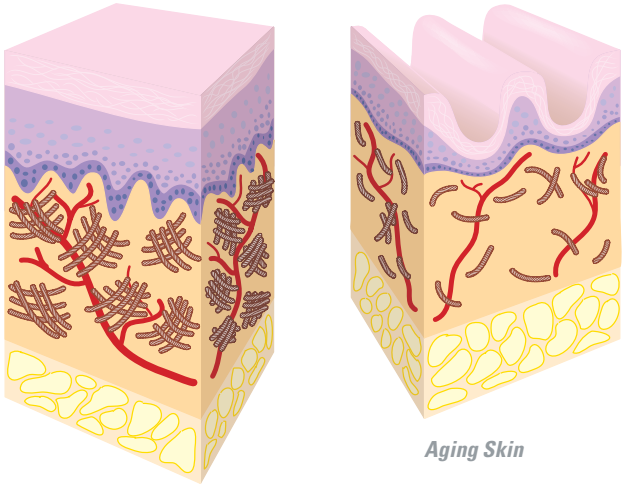
The myth of the Fountain of Youth has appealed to people for thousands of years. Part of that appeal is because the mythical water source promises to restore smoothness to the skin, eliminating the wrinkles that remind everyone who looks in the mirror that time has passed.

Now, in the 21st century, cosmetic products developed with scientific principles as their foundation provide us our Fountain of Youth. Retamax™ is a product that brings together components acknowledged for their proven effect on skin appearance and function with botanical ingredients derived from shrubs, flowering plants, and edibles, recently recognized as providing benefits for the skin as well. This document will outline the multiple mechanisms of action for Retamax™, and provide supportive evidence for its components.

Biomechanics of Skin Aging

Changes in Skin Structures

The causes of skin wrinkles are multifactorial. Some contributors are loss of elasticity from reduced water content of the stratum corneum, thickening of the stratum corneum, atrophy of the epidermis, decline in the amount and quality of dermal collagen and elastic fibers, and the weakening of the muscles supporting the skin.[1] The epidermis becomes visibly thinner, drier, and less elastic as the dermo-epidermal junction flattens.[2]



Young Skin

Aging Skin

Changes in Skin Biochemistry

Both chronologically aged and photo-aged skin share important molecular features including increased production of matrix metalloproteinases and diminished pro-collagen synthesis.[3] UV radiation or the aging process alone increases hydrogen peroxides and other reactive oxygen species and decreases endogenous antioxidant enzymes.[2] UV radiation particularly impairs the synthesis of new type I collagen and alters the organization of collagen fibrils within the skin.[2]

In addition, as an individual ages, the concentration of glycosoaminoglycans (such as hyaluronic acid, a major component of the extracellular matrix [ECM]) in the skin declines.[2] During youth, the ECM binds to large amounts of water, keeping collagen and elastin hydrated. As the individual ages, the ECM's ability to bind water diminishes. Collagen and elastin detach from the ECM and become dry and brittle, leading to fine lines and wrinkles.

Additionally, an excess of reactive oxygen species (ROS) causes oxidative damage to cellular lipids, proteins, and DNA.[4] When young, the body produces endogenous enzymes known as catalase, superoxide dismutase, and peroxidase, each of which minimizes oxidative damage. The concentration of these enzymes also declines as individuals age.[5]

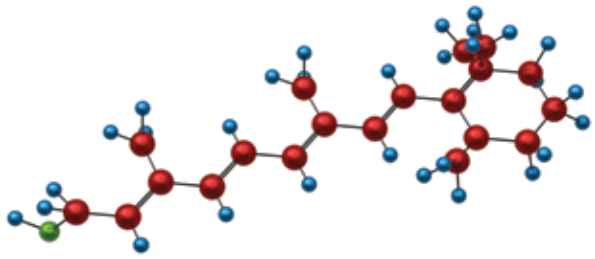
Retamax™

Multiple Mechanisms of Action

Retamax™ was formulated from the following active agents: retinol 0.5%, plant stem cells derived from a Chinese shrub, *Buddleja davidii*; ascorbic acid (vitamin C), alpha-tocopherol (vitamin E, as tocopheryl acetate), hydrolyzed sericin (a proprietary upregulator of collagen and hyaluronic acid), broccoli and sunflower seed extracts (a proprietary skin desensitizer), and a scientifically formulated mix of other humectants, emollients, emulsifiers, and other components. Details of how these ingredients work to enhance the appearance and function of the skin appear below.

Retinol

Retamax™ contains retinol at a concentration of 0.5%, higher than the concentration of most retinol-based products that are currently available over the counter. The retinol is structured as a micronized emulsion to maximize and accelerate the bio-delivery of retinol for rapid onset of results.[2]



Clinical studies have demonstrated retinol's effects on the skin. Visibly, fine lines and wrinkles significantly improved after 12 weeks of retinol treatment among participants in a controlled clinical trial.[6,7]

Invisibly, retinol is converted into retinoic acid, which is the biologically active form of vitamin A.[8] Beneath the surface of the skin, retinol stimulates the skin's natural rebuilding process by supporting the expression of glycosaminoglycans, chemicals that bind to water molecules.[8] This binding of water results in desirable, visible changes, such as firmer skin with a more even tone and texture, and fewer lines and wrinkles. In addition, retinol enhances the proliferation of epidermal cells, thus thickening the epidermis, and upregulates the expression of cellular retinoic acid binding protein (CRABP) II and CRABP mRNAs and proteins.[2] Retinol also inhibits accelerated induction of matrix metalloproteinases by exposure to ultraviolet light.[2]

Safety and Efficacy

Although initial clinical studies of retinol involved participants of middle age, retinol has also been effective and safe in the elderly. Kafi et al conducted a controlled clinical trial in 18 human volunteers (mean age 87 years) exposed to 0.4% retinol lotion on one arm compared to 18 who received vehicle 3 times a week for 24 weeks.[9] This trial showed a statistically significant induction of type I procollagen protein from baseline to week 24 ($P = 0.049$). The study also showed that retinol stimulated a statistically significant difference in glycosaminoglycan expression compared to control ($P = .02$).[9]

Irritation

Importantly, retinol is also less irritating than retinoic acid, leading to less erythema and scaling.[2,6] Individuals treated with topical retinol may experience photosensitivity, so patients are advised to avoid excessive sun exposure and to apply a broad-spectrum sunscreen.[2] Retinols are generally safe, but there is a slight potential for a retinoid reaction, although that is more common with the stronger retinoids like tretinoin and tazarotene. This reaction generally occurs within the first few days of beginning treatment, and is characterized by itching, a burning sensation where the product was applied, erythema, and peeling.[2] Adding natural agents like sitosterol, a recognized component of the *Buddleja* plant family, concomitantly with retinol may counteract some of these irritant effects.[2] Importantly, during 30 years of retinoid availability, young adults treated with topical retinoids over the long term have not experienced systemic side effects.[2]

Antioxidants

Buddleja Davidii

The importance of antioxidants for skin care is well established. The primary antioxidant in Retamax™ is derived from bioengineered stem cells from the native Chinese shrub butterfly bush (*Buddleja davidii*). This shrub has been used in traditional Chinese medicine for its wound healing, anti-inflammatory, antiviral, and antibacterial properties.[10,11] The components of this botanical source have been well characterized. Plants in the *Buddlejaceae* family have been shown to contain components that have antioxidant and anti-inflammatory properties.[10-13] In particular, *Buddleja* is rich in phytosterols and phenylpropanoids.[10]

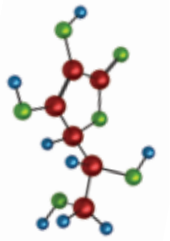
In addition, this botanical contains triterpenes and carotenoids (which have anti-inflammatory properties), and flavonoids (which are antioxidants).[11] In particular, the flavone linarin is present in *Buddleja*, which has been shown to have anti-inflammatory properties.[14] *Buddleja* plant extracts also contain polysaccharide components.[14]

The advantage to using *Buddleja* stem cells, rather than extracts from whole plants, is ecological. Culturing plant stem cells is sustainable, as opposed to field harvesting huge numbers of mature plants. In addition, stem cell cultures contain no contaminants.

Buddleja davidii extracts include phenylpropanoid derivatives, particularly verbascoside.[10] Verbascoside has antioxidant, anti-inflammatory and photoprotective properties. In particular, in cell cultures of human keratinocytes, verbascoside from a non-*Buddleja* source dose-dependently reduced the release of pro-inflammatory chemokines.[10,12] Verbascoside also accelerated wound healing in vivo in full-thickness excision wounds.[12]

Vitamin C and Vitamin E

In addition to antioxidant properties from *Buddleja*, Retamax™ contains two other antioxidants – ascorbic acid (vitamin C) in the form of ascorbyl palmitate, and alpha-tocopherol (vitamin E) in the form of tocopheryl acetate. Not only do these vitamins have antioxidant and anti-inflammatory properties, but data suggests that the two scavenge free radicals in a synergistic manner by recycling each other.[8,15]

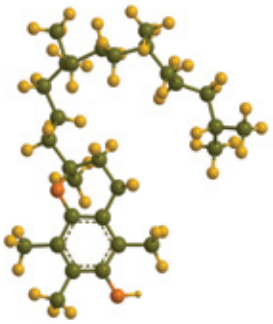


Vitamin C

Ascorbyl palmitate is a compound derived from of ascorbic acid (vitamin C) and palmitic acid, a naturally occurring fatty acid. Ascorbyl palmitate is the ester formulation of ascorbic acid, which has better stability when used in topically applied products.[15] Ascorbic acid has the capability to scavenge superoxide radicals.[16]

Alpha-tocopherol is a lipophilic (oil-soluble antioxidant and free-radical scavenger) which is naturally found in cell membranes.[17] Alpha-tocopherol also acts as a humectant, improving the skin's ability to bind water and reducing transepidermal water loss.[18] Participants in a 4-week study of a 5% RRR alpha-tocopherol oil-in-water cream applied to crow's feet experienced diminished skin roughness, length of facial lines, and depth of wrinkles compared to placebo, as measured by optical profilometry. In another study by the same investigators, participants who applied topical vitamin E for 10 days experienced improved skin smoothness.[19]

Fifteen animal studies have demonstrated the photo-damage prevention properties of topical pretreatment with vitamin E, which lowers erythema, wrinkling (visibly), sunburn cell formation and lipid peroxidation on the molecular level.[20] Photo-damage prevention properties have been confirmed in clinical trials as well. For example, Chung et al. applied 5% vitamin E topical (24 hours prior to UV exposure) and found that it inhibited UV-induced matrix metalloproteinase-12 mRNA by 47%.[20]



Vitamin E

Because vitamin C regenerates oxidized vitamin E, the combination of vitamin C and vitamin E in Retamax™ provides synergistic benefits, especially in terms of UV protection.[8,16,21]

Hydrolyzed Sericin

Hydrolyzed sericin consists of biomimetic hydrolyzed silk peptides (sericin), phospholipids, and butylene glycol. It supports the skin's natural rebuilding process by stimulating the bio-synthesis of collagen and hyaluronic acid naturally produced in the skin. Human dermal fibroblasts treated with hydrolyzed sericin protein express hyaluronic acid within 48 hours of initial exposure. Simultaneously, hyaluronidase (the enzyme that destroys hyaluronic acid) concentrations decline.

In addition, sericin is associated with antioxidant properties and inhibits lipid peroxidation and tyrosinase activity, which may help even skin tone and reduce hyperpigmentation.[22]

Sericin may also confer a photo-damage prevention effect against UVB-induced acute damage based on suppression of cyclooxygenase-2 (COX-2) protein and other markers of sun damage in epidermis.[23]

Anti-Irritants

Retamax™ minimizes skin sensitivity with a proprietary standardized extract derived from two botanical sources: *Brassica oleracea italica* (broccoli) and *Helianthus annuus* (sunflower seed) oil. Other components include glucosinolates, di-indolylmethane, sulforaphane, and glutathione conjugates.

Brassica Oleracea Italica

Broccoli extracts inhibit cellular oxidative stress, as demonstrated by a decrease of reactive oxygen species-induced oxidation of dichlorofluorescein, visible as a change in fluorescence.[4] Broccoli extracts contain recognized antioxidants such as tocopherols and polyphenols (including flavonoids), as well as carotenoids such as beta carotene and lutein.[4]

Helianthus Annuus

Sunflower seed contains a high concentration of linoleic acid, an essential fatty acid that the body does not synthesize endogenously.[24] Investigators measured the rate of transepidermal water loss (TEWL) after tape-stripping the *stratum corneum* of hairless mouse epidermis and applying sunflower seed oil. A single application significantly speeded barrier recovery within 1 hour of application, an effect that was sustained for at least 5 hours (both $P<.01$). The skin barrier in both mice and humans is essentially identical structurally and biochemically, so the tape-stripped hairless mouse model is widely accepted as a model for preclinical studies of dermatological products in humans. The presence of a fatty acid transporter on keratinocytes also enables epidermis to metabolize lipids from topically applied sunflower oil and use them as nutritional building blocks for the epidermal barrier.[18] Sunflower seed oil may also confer photo-damage prevention properties because the absorbance spectrum of sunflower seed oil shows considerable absorbance of ultraviolet radiation in the 300 to 360 nm range.[24]

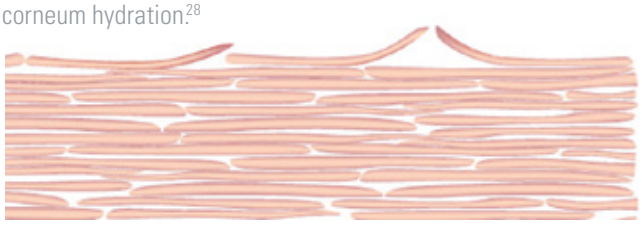
Glycation is a non-enzymatically driven reaction (known as the Maillard reaction) between free amine groups like those of amino acids in proteins and reducing sugars like glucose. This reaction leads to the formation of advanced glycation end products such as carboxymethyl lysine, pentosidine and others, which can form cross-links between macromolecules by covalent bonding.[25] Epidermal markers in particular are affected by glycation. For example, the function of integrins, which are epidermal markers related to epidermal stratification and differentiation, change after glycation of collagen. In particular, b1 integrin is involved in a wide variety of important biological functions such as epithelial cytoskeletal organization, basement membrane biosynthesis, adhesion and migration of fibroblasts and keratinocytes, and collagen synthesis.[25]

Brassica oleracea italica (broccoli) and *Helianthus annuus* (sunflower seed) oil extract reduces the formation of glycation end products, probably because of the sunflower component.[26] Sunflower sprouts strongly inhibited the formation of advanced glycation end products when administered orally to rats.[26] Sunflower also increases the production of the protective enzymes, catalase and glutathione, in rat liver when ingested.[27]

Barrier Restoration

Beta-Glucan

Beta-glucan helps with water retention in the *stratum corneum* (in combination with chitin) in humans.[28] In a 16-week randomized, placebo-controlled study, 20 men showing signs of aging skin applied 1.5% chitin-glucan formulation twice daily. Participants experienced a progressive increase in skin firmness and stratum corneum hydration.[28]



Beta-glucan is also associated with anti-inflammatory functions. The expected acute inflammatory response to xylene treatment declined in a statistically significant and dose-dependent manner after oral administration of beta-glucan to mice.[29]

Humectants

Humectants reinforce the skin's barrier function by replenishing the moisture level, thereby hydrating the skin. The skin produces its own exogenous humectants like glycosaminoglycans.[17] In the *stratum corneum*, the endogenous humectant is called natural moisturizing factor.[19] Exogenous humectants penetrate into the skin and increase the degree of hydration of the *stratum corneum*.[19] Humectants also stabilize the cosmetic itself, and have bacteriostatic properties.[1]

Retamax™ contains the following humectants:

Glycerin (also called glycerol) exists in the *stratum corneum* as a natural endogenous humectant. Exogenous glycerin increases the ability of the *stratum corneum* to bind water, and many cosmetics include this ingredient because it moisturizes the skin in a way that mimics the glycerin made by the body.[19,30]

Pentylene Glycol is included in Retamax™ for its humectant properties, but it also has antibacterial properties.[18]

Conclusions

Retamax™ represents the ultimate utilization of science in the service of skin health restoration and rejuvenation. By incorporating retinol with its recognized, multi-vectored mechanisms of action with botanical ingredients and bioengineered components, Retamax™ comes as close as we are likely to get to the mythical Fountain of Youth.

Major Components of Retamax™

Table 1. Evidence base for components of Retamax™

Table 1 summarizes the evidence that supports the benefits of the major components of Retamax.™

INGREDIENT	COMPONENTS	STUDY TYPE	FINDINGS	REFERENCE
Retinol		Clinical 12 weeks	Fine lines, wrinkles significantly improved	6, 7
***Buddleja davidii* stem cells**		Pre-clinical	Contains verbascoside and other substances with antioxidant and anti-inflammatory properties	10-13
Biomimetic sericin protein	biomimetic hydrolyzed silk peptides (sericin), phospholipids, and butylene glycol	Pre-clinical	Sericin treatment upregulates hyaluronic acid within 48 hours of initial exposure	31
		Pre-clinical	Sericin has antioxidant properties and inhibits lipid peroxidation and tyrosinase activity on suppression of cyclooxygenase-2	22
Broccoli extract	*Brassica oleracea italica* (broccoli)	Pre-clinical	Inhibits cellular oxidative stress and contains recognized antioxidants such as tocopherols and polyphenols (including flavonoids)	4
Sunflower seed oil extract	*Helianthus annuus* (sunflower seed) oil	Pre-clinical	Reduces transepidermal water loss to restore epidermal barrier	24
Ascorbyl palmitate (vitamin C)		Pre-clinical	Scavenges superoxide and peroxyl radical	15, 16
Alpha-tocopherol (vitamin E)		Clinical 10 days	Improved skin smoothness	19
			Inhibits UV-induced matrix metalloproteinase-12 mRNA by 47%	20
Beta-glucan		Clinical 16 weeks	Increases skin firmness and stratum corneum hydration	28

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BY ZEIN OBAGI, MD

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