

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization

International Bureau



(10) International Publication Number

WO 2013/022788 A1

(43) International Publication Date
14 February 2013 (14.02.2013)

WIPO | PCT

(51) International Patent Classification:

A01N 65/00 (2009.01)

(21) International Application Number:

PCT/US2012/049619

(22) International Filing Date:

3 August 2012 (03.08.2012)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

61/515,754 5 August 2011 (05.08.2011) US

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(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

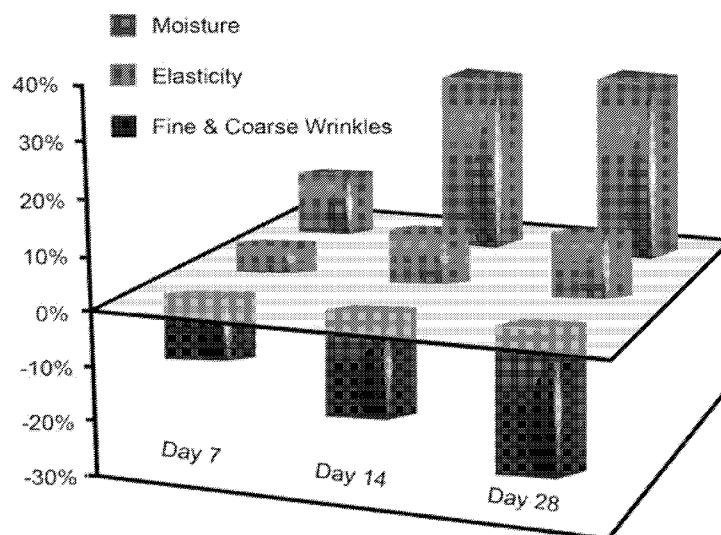
(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

— with international search report (Art. 21(3))

(54) Title: SKIN CARE COMPOSITIONS CONTAINING COMBINATIONS OF NATURAL INGREDIENTS

Figure 11



(57) Abstract: Described herein are various natural ingredients for skin-related applications. Individual compounds provide beneficial effects for improved collagen production, fibroblast proliferation, antioxidant protection and free radical inhibition. Combinations of these individual compounds exhibit synergistic effects, leading to dramatic improvements in skin moisture levels, wrinkle reduction, and elasticity. Various examples of combined ingredients are provided, which can be applied in methods related to conditioning the skin, and in cosmetic formulations for improved aesthetic appearance.

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SKIN CARE COMPOSITIONS CONTAINING COMBINATIONS OF NATURAL INGREDIENTS

FIELD OF THE INVENTION

5 This invention relates to the use of combinations of natural ingredients in skin-related applications.

BACKGROUND

All publications herein are incorporated by reference to the same extent as if each individual publication or patent application was specifically and individually indicated to be 10 incorporated by reference. The following description includes information that may be useful in understanding the present invention. It is not an admission that any of the information provided herein is prior art or relevant to the presently claimed invention, or that any publication specifically or implicitly referenced is prior art.

15 Although compositions for skin care products operate through a variety of mechanisms, most provide only temporary moisture retention for improved hydration, transitory improvement in appearance and/or superficial relief of skin integrity.

Human skin is composed of three primary layers: the epidermis, dermis, and 20 hypodermis. Each of these layers contains specific cell populations embedded within a complex physical and biochemical matrix, thereby providing the mechanical and functional organization necessary for the skin to maintain barrier function. It is well-known that the epidermis sloughs millions of cells every day, creating an enormous demand for replacement 25 cells. Central to this process, which operates continuously throughout an organism's lifetime, are the various populations of progenitors and stem cells and the associated fibroblasts which provide biomechanical and trophic support. Progenitors and stem cells proliferate and differentiate as part of the natural repair and regeneration mechanism in the body, in order to meet this continual demand for new cells in the skin. Fibroblasts, which populate the dermis, provide vital trophic support, while producing important protein components of the skin.

There is a clear need in the art for compositions for skin care products, and methods 30 of preparing skin care products, that have a proven capacity to directly act upon skin cells. This includes enhancement of proliferation and maintenance of skin cell populations for the purpose of promoting the skin repair and regenerative mechanisms in the body.

As described herein, the inventor has discovered new and useful compositions derived from natural ingredients in many of these plants act by supporting the proliferation, migration and differentiation of skin progenitor and stem cells and/or fibroblasts. These natural

ingredients can be applied topically in a blended mixture (Coconut oil, Nilotica butter, Rosa mosqueta, Olivem 1000, Cocoa butter, Soy lecithin, Chilean Soapnut, NovHyal, Fucoidan Maritech, Moringa, Aloe vera, Genistein, Black mamaku, Cehami, Amla, AFA, Pomegranate, Sangre de drago, Guar, Vanilla, Colostrum, Cytokines, Maqui, Synergy berries, 5 Green Tea Extract, Vanilla 20 fold extract, Cacao, Olive extract (Hydroxytyrolosol), Vitamin E (Sunflower), Samambaia, Bulgarian Rose, Jasmine, Sweet Orange, Ylang-ylang, Tangerine Essential Oil, Bitter Orange extract, and Honeysuckle), proved to have a significant effect on moisture retention in the skin, skin elasticity, as well as wrinkle reduction.

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SUMMARY OF THE INVENTION

The present invention provides, in one embodiment, a purified skin care composition including one or more of the following components selected from the group including: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or 15 extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, 20 berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* 25 or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof, and a cosmetically acceptable carrier. In another embodiment, the purified skin care composition is an emulsion substantially free of artificial ingredients.

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Another aspect of the present invention provides, in one embodiment, a method of conditioning the skin of a subject including providing a quantity of a purified composition, wherein the composition including one or more of the following components selected from the group including: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda*

cunninghamii or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof, and administering a quantity of the purified composition to the subject in an amount sufficient to condition the skin of the subject. In another embodiment, conditioning the skin of the subject results in an increase in progenitor cell, stem cell, and/or dermal fibroblast proliferation. In another embodiment, conditioning the skin of the subject results in an increase in progenitor cell, stem cell, and/or dermal fibroblast mobilization. In another embodiment, conditioning the skin of the subject results in an increase in progenitor cell, stem cell, and/or dermal fibroblast migration. In another embodiment, conditioning the skin of the subject results in an increase in antioxidant protection. In another embodiment, conditioning the skin of the subject results in inhibition of free radical formation.

Another aspect of the present invention provides, in one embodiment, a method of improving skin appearance in a subject, including providing a quantity of a purified composition, wherein the composition includes one or more of the following components selected from the group including: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa*

rubiginosa or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof, and administering a quantity of the purified composition to the subject in an amount sufficient to improve skin appearance in a subject. In another embodiment, the improved skin appearance includes improved skin tone. In another embodiment, the improved skin appearance includes improved skin elasticity. In another embodiment, the improved skin appearance includes enhanced skin thickness. In another embodiment, the improved skin appearance includes improved skin hydration.

Another aspect of the present invention provides, in one embodiment, A method of reducing age-related features in the skin of a subject including providing a quantity of a purified composition, wherein the composition includes one or more of the following components selected from the group including: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof, and administering a quantity of the purified composition to the subject in an amount sufficient to reducing age-related features in the skin of a subject. In another embodiment, age-related features include wrinkles. In another embodiment, the age-related features include fine lines. In another embodiment, the age-related features include dark patches of skin or age spots.

BRIEF DESCRIPTION OF THE DRAWINGS

Exemplary embodiments are illustrated in referenced figures. It is intended that the embodiments and figures disclosed herein are to be considered illustrative rather than restrictive.

5 **Figure 1** depicts effects of individual compounds on cell proliferation. In neonatal dermal fibroblasts, a clear increase in proliferation of fibroblasts was observed for the following individual compound as described: *Aphanizomenon flos-aquae* (AFA) (96%), aloe (*Aloe Barbadensis*) (87%), fucoidan from *Undaria pinnatifida* (55%), cehami (*Centipeda cunninghamii*) (30%), cacao (*Theobroma cacao*) (54%), maqui berry (*Aristotelia chilensis*)
10 (58%) and vanilla (*Vanilla planifolia*) (25%).

Figure 2 depicts effects of individual compounds on cell proliferation. In adult human dermal fibroblasts, *Aphanizomenon flos-aquae* (AFA), aloe (*Aloe Barbadensis*), fucoidan from *Undaria pinnatifida*, black mamaku (*Cyathea medullaris*), cehami (*Centipeda cunninghamii*), cytokine blend, cacao (*Theobroma cacao*), and maqui berry (*Aristotelia chilensis*) significantly increase dermal fibroblast proliferation, ranging from an increase of over 10% up to nearly 70% across various compounds. Vanilla (*Vanilla planifolia*) and colostrum did not significantly increase adult dermal fibroblast proliferation on their own but synergistically potentiated the effect of AFA. A blend of cytokines, including epidermal growth factor, fibroblast growth factors, keratinocytes growth factor, hepatocyte growth factors, and stem cell factor, led to substantial increase in dermal fibroblast proliferation above baseline.
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Figure 3 depicts the effect of combinations of compounds. In adult dermal fibroblasts, individual compounds clearly demonstrated synergistic effects when blended together in enhancing the proliferation. AFA enhanced the proliferation of adult dermal fibroblasts by 26%. However, aloe (*Aloe Barbadensis*), fucoidan from *Undaria pinnatifida* and cehami (*Centipeda cunninghamii*) individually reduced proliferation by 37%, 22% and 30%, respectively. When combined, blends of these various ingredients enhanced proliferation by 42% to 86%. Most significant effects were observed for blends of AFA, fucoidan and cehami (86%), aloe and fucoidan (74%), aloe, fucoidan, and cehami (71%),
25
30 fucoidan and cehami (70%), AFA, aloe and fucoidan (70%), and a blend of all four ingredients (63%). Blending of AFA with cacao (*Theobroma cacao*) and vanilla (*Vanilla planifolia*) also created a synergy that increased the proliferation adult dermal fibroblasts by 30% and 15%.

Figure 4 depicts effects of individual compounds on collagen production. In adult human dermal fibroblasts, collagen production by primary adult human dermal fibroblasts in culture was increased by fucoidan from *Undaria pinnatifida*, bovine colostrum extract, cytokines, cehami (*Centipeda cunninghamii*), and amla (*Phyllanthus emblica*). Whereas 5 control untreated fibroblasts secreted approximately 0.3 µg/mL of collagen, treated samples secreted amounts ranging from 0.8 to over 1.2 µg/mL of collagen.

Figure 5 depicts a wound healing model scratch test. **(A)**: Diagram of an *in vitro* cellular bioassay model used for wound healing, wherein adult human dermal fibroblasts were cultured until formation of a dense monolayer. A scratch was created through the 10 monolayer, and following treatment with individual compounds, recover was observed after 36 hours. Phenotypic observations with exemplary scoring system ranging from 0-10 is shown. **(B)**: Effect of ingredients on *in vitro* scratch recovery. A cytokine blend, including epidermal growth factor, fibroblast growth factors, keratinocytes growth factor, hepatocyte growth factors, and stem cell factor, led to a substantial increase in dermal fibroblast 15 migration and accelerated recovery in the *in vitro* scratch assay. In addition, aloe (*Aloe Barbadensis*), *Aphanizomenon flos-aquae* (AFA), Sangre de drago (*Croton lechleri*), maqui berry (*Aristotelia chilensis*), and bovine colostrum extract supported accelerated recovery.

Figure 6. depicts effect of stem cell proliferation and migration. Neonatal and adult skin fibroblasts grown to confluence in wells were subjected to the scratch test prior to 20 exposure to extracts from various plants in order to study the effect of herbal extracts on stem cell proliferation and migration. Representative results are shown as follows: **(A)**: Control **(B)**: aloe (*Aloe Barbadensis*) **(C)**: black mamaku (*Cyathea medullaris*) **(D)**: cacao (*Theobroma cacao*) **(E)**: maqui berry (*Aristotelia chilensis*) **(F)**: berry extracts blend.

Figure 7. depicts wrinkle reduction following short-term application. **(A)**: Representative result of applying combination of natural ingredient over 7-day period. Measurements indicated a 74% reduction in wrinkles. **(B)**: Representative result of applying combination of natural ingredient over 7-day period. Measurements indicated a 81% reduction in wrinkles.

Figure 8. depicts examples of improvements in skin appearance. Skin appearance is 30 improved following 28 day application of combined ingredients, as shown by wrinkle depth, darkness under eyes, and skin hydration, indicated by red arrows. In open-label study, 10 individuals (5 males and 5 females) used combination of ingredients twice daily for 28 days. Measurements of skin elasticity and moisture, along with surface skin analysis, were performed at 0, 7, 14, and 28 days. A first representative patient example is shown in **(A)**: 0

days, **(B)**: 7 days, and **(C)**: 28 days of treatment. A second representative example is shown in **(D)**: 0 days, **(E)**: 7 days, and **(F)**: 28 days of treatment.

Figure 9. depicts examples of wrinkle reduction following prolonged application.

(A): Application of combination of ingredients over a 28 day period resulted in an 82% wrinkle reduction at the base of the eyelid. **(B):** Representative result of wrinkle reduction on the forehead as a result of topical application of the serum containing a combination of natural ingredients.

Figure 10. depicts measurements of skin properties. **(A):** Visioscan image analysis

system was deployed to image wrinkle appearance as associated with surface roughness from fine and coarse wrinkle depth. Combinations of ingredients applied over a 28-day period demonstrated dramatic decreases in surface roughness, with highly significant decreases appearing at 7 days (** indicating statistical significance, $p<0.001$), and remained highly significant at 14 and 28 days of product use (** indicating statistical significance, $p<0.02$).

Average reductions were 9.8%, 17.4%, and 25.3% after 7, 14, and 28 days of use, respectively, with maximum % improvement reaching 39.3%. **(B):** Evaluation of skin

elasticity/flexibility via cutometer indicated an increase in biological elasticity on the test sites treated with the test product. The increases were statistically significant from baseline after 14 and 28 days of use (as indicated by *), and averaged a 10% increase in elasticity, with maximum % improvement reaching 31.9%. **(C):** Nova Dermal Phase meter was deployed for skin hydration measurement. Measurements demonstrated that the test product dramatically increase the skin moisture content. The increases were statistically significant from baseline after 14 and 28 days of use (as indicated by *) with average increases of 29.7% and 30.5%.

Figure 11. depicts improvements in moisture retention, elasticity and wrinkle

removal. Topical application of a serum containing cold infused extracts of *Aphanizomenon flos-aquae* (AFA), fucoidan from *Undaria pinnatifida*, aloe (*Aloe Barbadensis*), cehami (*Centipeda cunninghamii*), black mamaku (*Cyathea medullaris*), cacao (*Theobroma cacao*), berry extracts blend, maqui berry (*Aristotelia chilensis*), colostrum, vanilla (*Vanilla planifolia*), pomegranate (*Punica granatum*), Sangre de drago (*Croton lechleri*), and green tea (*Camellia sinensis*) led to a 25% reduction in wrinkle, 30% increase in moisture retention and 10% increase in elasticity. Results across 7-day, 14-day and 28-day time periods clearly demonstrate an improvement over time.

Figure 12. depicts effects of individual compounds on cellular antioxidant protection.

Cellular antioxidant protection capacity of each ingredient was tested in the CAP-e bioassay.

The *in vitro* data demonstrated potent antioxidant bioavailability by amla (*Phyllanthus emblica*), pomegranate (*Punica granatum*), Sangre de drago (*Croton lechleri*), and green tea (*Camellia sinensis*).

Figure 13. depicts effects of individual compounds on inhibiting free radical formation. The inhibitory effect of each ingredient on free radical formation by inflammatory cells placed under oxidative stress conditions was tested. The *in vitro* data demonstrated reduced free radical production by individual compounds, including fucoidan from *Undaria pinnatifida*, Shea butter, rosa mosqueta (*Rosa rubiginosa*), maqui berry (*Aristotelia chilensis*), vanilla (*Vanilla planifolia*), and green tea (*Camellia sinensis*).

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DETAILED DESCRIPTION

All references cited herein are incorporated by reference in their entirety as though fully set forth. Unless defined otherwise, technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Singleton *et al.*, *Dictionary of Microbiology and Molecular Biology* 3rd ed., J. Wiley & Sons (New York, NY 2001); March, *Advanced Organic Chemistry Reactions, Mechanisms and Structure* 5th ed., J. Wiley & Sons (New York, NY 2001); and Sambrook and Russel, *Molecular Cloning: A Laboratory Manual* 3rd ed., Cold Spring Harbor Laboratory Press (Cold Spring Harbor, NY 2001); Dixon, *Plant Cell Culture: A Practical Approach*, Oxford University Press (U.S.A. 1994) provide one skilled in the art with a general guide to many of the terms used in the present application.

One skilled in the art will recognize many methods and materials similar or equivalent to those described herein, which could be used in the practice of the present invention. Indeed, the present invention is in no way limited to the methods and materials described.

25 For purposes of the present invention, the following terms are defined below.

As used in the description herein and throughout the claims that follow, the meaning of “a,” “an,” and “the” includes plural reference unless the context clearly dictates otherwise. Also, as used in the description herein, the meaning of “in” includes “in” and “on” unless the context clearly dictates otherwise.

30 “Artificial ingredients” as used herein refers to ingredients that are not found in nature and therefore must be synthetically produced as artificial ingredients. Examples includes additives commonly produced for the purpose of adding color, flavoring, taste, surfactants and emollients desired physical properties or preservatives for extended storage.

“Colostrum” as used herein refers to a fluid secreted by the mammary glands of

female mammals during the first few days of lactation, containing various nutrients and protease inhibitors that keep it from being destroyed by the processes of digestion. Humans produce relatively small amounts of colostrum in the first two days after giving birth, but cows produce about nine gallons of colostrum.

5 “Cosmetically effective amount” as used herein is the quantity of a composition provided for administration and at a particular dosing regimen that is sufficient to achieve a desired appearance, feel, and/or protective effect. For example, an amount that results in the prevention of or a decrease in the appearance and/or symptoms associated with an undesirable condition, such as wrinkles, fine lines, skin thinness, loss of skin elasticity or 10 suppleness, or other characteristics of skin associated with aging, UV, chemical exposure, adverse climate (e.g., temperature, humidity), dietary intake, biological agents, environmental oxidants, among others.

“Fucoidan” as used herein, describes sulfated fucans obtained from algae. Fucoidan has been obtained from a broad range Algae species as provided in the following non- 15 exhaustive list: *Cladosiphon okamuranus*, *Chordaria flagelliformis*, *Ch. Gracilis*, *Saundersella simplex*, *Desmaestia intermedia*, *Dictyosiphon foeniculaceus*, *Dictyota dichotoma*, *Padina pavonica*, *Spatoglossum schroederi*, *Adernocystis utricularis*, *Pylayella littoralis*, *Ascophyllum nodosum*, *Bifurcaria bifurcata*, *Fucus. Visculosus*, *F. spiralis*, *F. serratus*, *F. evaescens*, *Himanthalia lorea*, *Hizikia fusiforme*, *Pelvetia canaliculata*, *P. 20 wrightii*, *Sargassum stenophyllum*, *S. honeri*, *S. Khellmanium*, *S. muticum*, *Alaria fistulosa*, *A. marginata*, *Arthrothammus bifidus*, *Chorda film*, *Ecklonia kurome*, *E. cava*, *Eisenia bicyclis*, *Laminaria angustata*, *L. brasiliensis*, *L. cloustoni*, *L. digitata*, *L. japonica*, *L. religiosa*, *L. saccharina*, *Macrocytis integrifolia*, *M. pyrifera*, *Nereocystis luetkeana*, *Undaria pinnatifida*, *Petalonia fascia*, *Scytoniphon lomentaria*.

25 “Plant” and “plants” as used herein, include all species of the Kingdom Plantae, including plants under the Division Chlorophyta, Division Rhodophora, Division Paeophyta, Division Bryophyta and Division Tracheophyta; Subdivision Lycopsida, Subdivision Sphenopsida, Subdivision Pteropsida and Subdivision Spermopsida, Class Gymnospermae, Class Angiospermae, Subclass Dicotyledonidae and Subclass Monocotyledonidae. Plants 30 include herbs, lower plants such as fungi and algae, and any organism colloquially known as a plant, fruit, or vegetable. Plants further include single cells, a plurality of cells, primary cell cultures, cell lines, cellular explants, individual organs, or any other cellular derivatives obtained from all species of Kingdom Plantae described herein. “Plant” and “plants” are

used interchangeably herein with “botanical”, wherein “botanical” includes all species of Kingdom Plantae described herein.

“Progenitor cell” as used herein refers to a cell that gives rise to progeny in a defined cell lineage.

5 “Subject” refers to a mammal, preferably a human, seeking to improve a condition, disorder or disease, including changes in aesthetic appearance, such as reduction/removal of undesirable aesthetic features (e.g., wrinkle reduction, improved hydration and elasticity of skin).

10 “Stem cell” as used herein refers to a cell that has the ability to divide (self replicate) for indefinite periods, often throughout the life of an organism, and which, under certain conditions or given particular signals, can differentiate to many different cell types that make up the organism. That is, stem cells have the potential to develop into mature cells that have characteristic shapes and specialized functions, such as keratinocytes, sebocytes, transit amplifying cells, or melanocytes. Stem cells may reside in the epithelial basal layer, 15 interfollicular epidermis (IFE) niches, subaceous gland and hair follicle bulge.

Progenitor and Stem Cells in the Skin. A classic example of the central role of progenitor and stem cells in skin repair and regeneration is that of keratinocyte stem cells (KSCs) maintaining epidermal barrier function (Mimeault and Batra, 2010). The epidermal barrier is characterized by a continual loss of terminally differentiated keratinocytes in the outermost epidermal layer, stratum corneum shed from the surface of the skin. KSCs that are located near the basement membrane of the innermost epithelial basal layer provide an essential role in replenishing of mature keratinocytes of epidermis, under both normal homeostatic conditions and in response to skin injury/wound healing. This is achieved by a 20 characteristic asymmetric division of KSCs within the basal layer niche, resulting in both propagation of KSCs and more committed transit amplifying (TA)/intermediate cells. These TA cells then migrate outside the niche, giving rise to the terminally differentiated keratinocytes that constitute nearly 95% of cells found in various epidermal layers.

25 Although the skin contains wide potential for repair and regeneration, over an organism’s lifetime, intrinsic and extrinsic factors combine to collectively alter the structural integrity and underlying repair and regenerative capacity of skin (Racila and Bickenbach, 2009). Aging of skin is characterized by loss of tone and elasticity, which may be coupled with increased epidermal dehydration, wrinkles, susceptibility to injury, and slow wound healing. A chief extrinsic factor causing accelerated skin-aging is UV exposure, wherein

advanced exposure to UV light results in the cross-linking between skin proteins. Cross-linking of elastins and collagens, which are extracellular matrix (“ECM”) proteins found in the skin, reduces reactivity with other skin matrix components, thereby reducing the optimal mechanical properties of skin. Simultaneously, more subtle molecular changes limit the self-renewal ability and regenerative capacity. For example, stem cells expressing melanoma chondroitin sulfate proteoglycan (MCSP) and $\beta 1$ integrins possess a high degree of self-renewal capacity, and expression of these markers is found to be drastically reduced in aging skin. In this regard, aging can be visualized as a multi-faceted process of change in both mechanical structure and a progressive reduction in underlying functional capacity.

10 Although keratinocytes are among the best known progenitor and stem cells supporting the populations of mature keratinocytes in the epidermis, a variety of other progenitor and stem cells are known to exist, performing specialized roles in the dynamic environment of skin maintenance, regeneration and repair. Importantly, many of these different types of progenitor and stem cell subpopulations are localized within very specific
15 microenvironments located in the skin structure. For example, consider the different types of progenitor and stem cell subpopulations found within interfollicular epidermis (IFE) niches, subaceous gland and hair follicle bulge region niches. It appears that unipotent CK5/14 and BLIMP1 positive progenitor cells exist near the basement membrane surrounding the cutaneous sebaceous gland in the hair follicle. These cells have a primary role directed at
20 regeneration of the subaceous gland, including terminal differentiation into sebocytes, which are responsible for sebum secretion. In contrast, the hair follicle bulge appears to contain multipotent epithelial stem cells (bESCs) that also give rise to mature keratinocytes, which in turn regenerate epidermal cells or repair epithelial tissue in response to injuries (Fuchs and Horsley, 2008). Further contrasting these two examples, the lower permanent portion of the
25 bulge appears contain a population of melanocyte precursors, which play an important role in hair and skin pigmentation. In short, a complex interplay exists between the progenitor and stem cells found in these structures and their specific contribution to the maintenance of skin integrity and function.

30 **Dermal Fibroblasts.** As the epidermis is avascular (i.e., not associated with or supplied by blood vessels), the primary means of supporting biological function is achieved through diffusion of nutrients by the underlying dermal layer. This layer is characterized by the presence of collagen and elastin fibers which provide mechanical structure of skin tissue, these ECM proteins themselves being produced by dermal fibroblasts. Reduced collagen

production is characteristic of accelerated aging, in addition to post-translational modifications such as cross-linking caused by UV, as described above. While fibroblasts are the principal source of ECM in the skin, they are also a primary source of metalloproteinase proteins (“MMPs”), which are key molecules involved in degradation of ECM proteins. To 5 the extent that MMP expression by fibroblasts is a normal process related to the natural turnover of ECM proteins in the skin, elevated MMP expression level by fibroblasts have been associated with accelerated aging. In turn, these higher levels of protein degrading enzymes lead to depletion and fragmentation of skin collagen, a reduction in collagen synthesis, depletion of growth factors and nutrients within reservoirs providing biotrophic 10 support for skin cells, and diminished support from dermal fibroblasts and skin progenitor and stem cells within the basal lamina and epithelia. These observations affirm the view that aging is a combination of immediate, direct changes to the ECM protein structure and organization via elevated MMP activity, coupled with a gradual progressive decrease in regenerative capacity via diminished progenitor and stem cell activity. This progressive loss 15 being the result of lowered mechanical and trophic support provided by fibroblasts establishing stem cell niche microenvironments.

Natural ingredients. A variety of anti-aging and skin care products exist on the market today. However, most of these compositions merely provide temporary improvement in skin 20 hydration, tonicity and appearance, without tapping into the natural repair and regenerative mechanism in the body. Further, natural ingredients, when derived from animal and/or botanical sources, provides a renewable, non-toxic, and consistent source of materials from deriving skin care components.

A number of natural ingredients have been shown to have a beneficial effect on 25 wound healing and the regeneration of the skin, though the mechanisms of action have often times remained unclear. For example, the capacity of genistein accelerate wound healing is only partially mediated via classical estrogen receptor-dependent signaling pathways, while the other mechanism of action remained unknown. *Phyllanthus emblica* was shown to accelerate wound healing, in part by supporting collagen formation and inhibiting matrix- 30 metalloproteinase. A similar mechanism of action has been postulated for the beneficial effect of cacao on the skin. Support of immune functions was the hypothesis proposed for the mechanism of action underling the beneficial effect of *Undaria pinnatifida* on the healing of wounds associated with herpes. The benefits of green tea were shown to be partially mediated by polyphenols that would bring antioxidant protection and support the secretion of

growth factors. The benefits of *Aloe barbadensis* were shown to be at least partially linked to the modulation of cytokines and growth factors in the skin. On the other hand, many others plants such as cehami (*Centipeda cunninghamii*), vanilla (*Vanilla planifolia*), *Aphanizomenon flos-aquae* (AFA), and black mamaku (*Cyathea medullaris*) have acquired a 5 reputation for being beneficial to the skin without any valid scientific assessment.

As an expanded example demonstrates, fucoidan is a sulfated fucan with a demonstrated role in promoting regeneration and repair mechanism via promotion of blood stem cell activity. Further, it has been shown that fucoidan promotes collagen production in dermal fibroblasts, via elevated integrin. Given that shifting levels of collagen production are 10 important steps in aging and wound healing, it is evident that natural ingredients such as fucoidan can serve as important components in compositions for enhancing skin care repair and regeneration by supporting the activity of progenitors and stem cells, as well as fibroblasts.

These many observations suggest that natural ingredients can serve as effective 15 ingredients for reducing undesirable traits associated with aging and/or compounds for facilitating wound care in different skin care applications. However, the wide ranging types of important cellular actors and complexity of biochemical processes governing mechanisms of skin repair and regeneration, makes it less likely that a single natural ingredient can address the variety of factors causing progressive loss of desirable skin appearance, integrity 20 and function. As such, without being bound by any theory, the inventor believes that combinations of natural ingredients provide important synergistic effects in supporting the many different processes involved in skin repair and regeneration. Whereas single ingredients may touch upon an important biochemical process leading to improved results, applying combination of more than one natural ingredient is more likely to provide 25 synergistic effects to more rapidly and effectively engage with several different processes for improving skin repair and regeneration. A blended mixture (Coconut oil, Nilotica butter, Rosa mosqueta, Olivem 1000, Cocoa butter, Soy lecithin, Chilean Soapnut, NovHyal, Fucoidan Maritech, Moringa, Aloe vera, Genistein, Black mamaku, Cehami, Amla, AFA, Pomegranate, Sangre de drago, Guar, Vanilla, Colostrum, Cytokines, Maqui, Synergy berries, 30 Green Tea Extract, Vanilla 20 fold extract, Cacao, Olive extract (Hydroxytyrolosol), Vitamin E (Sunflower), Samambaia, Bulgarian Rose, Jasmine, Sweet Orange, Ylang-ylang, Tangerine Essential Oil, Bitter Orange extract, and Honeysuckle), proved to have a significant effect on moisture retention in the skin, skin elasticity, as well as wrinkle reduction. This example

demonstrates the effective results that may be achieved using a combination of natural ingredients.

Further, as an additional advantage, the various biochemical properties of natural ingredients allows their desirable features to be combined together at the exclusion of 5 artificial ingredients. For example, pomengranate or extracts thereof can provide a desirable color and/or scent to a composition, without the use of a synthetic dye or flavoring. Likewise, *Croton lechleri* contains a natural elastic polymer component that can provide extensivity (i.e., spreadability), which could otherwise require the addition of a synthetic polymer. Similarly, Aloe extract, bitter orange extract from *Citrus aurantium*, and 10 honeysuckle extract from *Lonicera Japonica* all contain natural bacteriostatic properties, which reduces the need for synthetic preservatives or addition of a synthetic bacteriocide. Together, the different natural ingredients in a combined composition provide dual benefits: effective components for skin care conditioning, without the need for artificial ingredients for coloring, flavoring, taste, extended preservation, among other

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The present invention provides new compositions and methods for providing a wide range of cosmetic and/or therapeutic benefits for conditioning the skin by administering to a subject, compositions containing combinations of natural ingredients. In one embodiment, the topical skin care composition contains one or more of the following: fucoidan or extracts 20 thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, 25 *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof and a cosmetically acceptable 30

carrier. In another embodiment, the topical skin care composition is an emulsion substantially free of artificial ingredients.

In other embodiments, the composition includes one or more of the following natural ingredients in a specific amount (% w/w): Coconut oil purified from *Cocos nucifera* in an amount of about 0 - 10%, Nilotica butter purified from *Vitellaria nilotica* in an amount of about 0 – 3.6%, Rosa mosqueta purified from *Rosa rubiginosa* in an amount of about 0 – 3.6%, Olivem 1000 purified from *Ceteryl olivate* in an amount of about 0 – 6.0%, Cocoa butter purified from *Theobroma cacao* in an amount of about 0 – 3.6%, Soy lecithin purified from *Glycine Max* in an amount of about 0 – 1.6%, Chilean Soapnut purified from *Quillaja saponaria* in an amount of about 0 – 3.0%, NovHyal purified from *NAG6P* in an amount of about 0 - 4.0%, Fucoidan purified from *Laminaria japonica* in an amount of about 0 – 3.0%, Moringa purified from *Moringa oleifera* in an amount of about 0 – 3.0%, Aloe vera purified from *Aloe Barbadensis* in an amount of about 0 – 2.0%, Genistein purified from soy in an amount of about 0 – 1.33%, Black mamaku purified from *Cyathea medularis* in an amount of about 0 – 1.33%, Cehami purified from *Centipeda cunninghamii* in an amount of about 0 – 1.33%, Amla purified from *Phyllanthus emblica* in an amount of about 0 – 1.33%, AFA purified from *Aphanizomenon flos-aquae* in an amount of about 0 - 0.66%, Pomegranate purified from *Punica granatum* in an amount of about 0 - 0.66%, Sangre de drago purified from *Croton lechleri* in an amount of about 0 - 0.66%, Guar purified from *Cyamopsis tetragonolobus* in an amount of about 0 – 1.33%, Vanilla purified from *Vanilla planifolia* in an amount of about 0 – 1.33%, Colostrum purified from first milk in an amount of about 0 - 0.66%, Cytokines purified from *E. coli* in an amount of about 0 - 0.20%, Maqui purified from *Aristotelia chilensis* in an amount of about 0 – 0.50%, berries extract obtained from Wild Bilberry, Wild Ligonberry, Black Currant, Aronia, Pomegranate, Wild Blueberry, Concorde 25 Grape, Sour Cherry, Wild Elderberry, Wild Cranberry, Red Raspberry, and/or Black Raspberry in an amount of about 0 – 0.50%, Green Tea Extract purified from *Camellia sinensis* in an amount of about 0 – 0.50%, Vanilla extract purified from *Vanilla planifolia* in an amount of about 0 – 0.44%, Cacao purified from *Theobroma cacao* in an amount of about 0 - 0.4%, Olive extract (Hydroxytyrolosol) purified from *Olea europaea* in an amount of about 0 – 2.0%, Vitamin E (Sunflower) purified from *Helianthus annuus* in an amount of about 0 – 1.6%, Samambaia purified from *Polypodium leucotomos* in an amount of about 0 – 1.0%, Bulgarian Rose purified from *Rosa damascena* in an amount of about 0 - 0.44%, Jasmine purified from *Jasminum grandiflorum* in an amount of about 0 - 0.48%, Sweet Orange purified from *Citrus sinensis* in an amount of about 0 - 0.80%, Ylang-ylang purified

from *Cananga odorata* in an amount of about 0 - 0.24%, Tangerine Essential Oil purified from *Citrus reticulata* in an amount of about 0 - 0.020%, Bitter Orange extract purified from *Citrus aurantium* in an amount of about 0 - 2.5%, and/or Honeysuckle purified from *Lonicera japonica* in an amount of about 0 - 2.5% and combinations thereof. In different 5 embodiments, the combinations of ingredient should constitute about 12-15% of the final composition volume.

The present invention further provides a method to condition the skin of a subject, including: providing a quantity of a composition, wherein the composition contains one or 10 more of the following components selected from the group including: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or 15 extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts 20 thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof; and administering a quantity 25 of the composition to the subject in an amount sufficient to condition the skin of the subject.

In another embodiment, conditioning the skin of the subject results in an increase in progenitor cell, stem cell and/or dermal fibroblast proliferation. In another embodiment, conditioning the skin of the subject results in an increase in progenitor cell, stem cell and/or dermal fibroblast mobilization. In another embodiment, conditioning the skin of the subject 30 results in an increase in progenitor cell, stem cell and/or dermal fibroblast migration. In another embodiment, conditioning the skin of the subject results in protection from antioxidant damage. In another embodiment, conditioning the skin of the subject results in inhibition of free radical formation. In another embodiment, conditioning the skin of the subject results in enhanced collagen production. In another embodiment, conditioning the

skin of the subject results in enhanced moisture retention, reduced appearance of fine lines, reduced appearance of wrinkles, reduced appearance of dark spots, age spots, improved complexion, improved skin tone, improved skin elasticity, enhanced healing of wounds, or enhanced skin thickness.

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The present invention further provides a method of improving skin tone, improving skin elasticity, or enhanced skin thickness, including: providing a quantity of a composition, wherein the composition contains one or more of the following components selected from the group including: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof; and administering a quantity of the composition to the subject in an amount sufficient to improve the skin tone, improve skin elasticity, or enhance thickness of the skin of the subject.

In another embodiment, improving skin tone, improving skin elasticity, or enhancing thickness of the skin of the subject results in an increase in progenitor cell, stem cell and/or dermal fibroblast proliferation. In another embodiment, improving skin tone, improving skin elasticity, or enhancing thickness of the skin of the subject is characterized by an increase in progenitor cell, stem cell and/or dermal fibroblast mobilization. In another embodiment, improving skin tone, improving skin elasticity, or enhancing thickness of the skin of the subject is characterized by an increase in progenitor cell, stem cell and/or dermal fibroblast migration. In another embodiment, improving skin tone, improving skin elasticity, or enhancing thickness of the skin of the subject is characterized by protection from antioxidant

damage. In another embodiment, improving skin tone, improving skin elasticity, or enhancing thickness of the skin of the subject is characterized by inhibition of free radical formation. In another embodiment, improving skin tone, improving skin elasticity, or enhancing thickness of the skin of the subject is characterized by enhanced collagen 5 production. In another embodiment, improving skin tone, improving skin elasticity, or enhancing thickness is characterized by enhanced moisture retention.

The present invention further provides a method of reducing the appearance of fine lines or reducing the appearance of wrinkles, including: providing a quantity of a 10 composition, wherein the composition contains one or more of the following components selected from the group including: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, 15 *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa 20 rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, 25 and/or *Vanilla* or extracts thereof; and administering a quantity of the composition to the subject in an amount sufficient to reduce the appearance of fine lines or reduce the appearance of wrinkles in the skin of the subject.

In another embodiment, reducing the appearance of fine lines or reducing the 30 appearance of wrinkles in the skin of the subject results in an increase in progenitor cell, stem cell and/or dermal fibroblast proliferation. In another embodiment, reducing the appearance of fine lines or reducing the appearance of wrinkles in the skin of the subject is characterized by an increase in progenitor cell, stem cell and/or dermal fibroblast mobilization. In another embodiment, reducing the appearance of fine lines or reducing the appearance of wrinkles in skin of the subject is characterized by an increase in progenitor cell, stem cell and/or dermal

fibroblast migration. In another embodiment, reducing the appearance of fine lines or reducing the appearance of wrinkles in the skin of the subject is characterized by protection from antioxidant damage. In another embodiment, reducing the appearance of fine lines or reducing the appearance of wrinkles in the skin of the subject is characterized by inhibition of 5 free radical formation. In another embodiment, reducing the appearance of fine lines or reducing the appearance of wrinkles in the skin of the subject is characterized by enhanced collagen production. In another embodiment, reducing the appearance of fine lines or reducing the appearance of wrinkles is characterized by enhanced moisture retention. In another embodiment, frequency and depth of wrinkles can be measured using Visioscan 10 imaging. In another embodiment, skin elasticity can be measured using a cutometer. In another embodiment, skin hydration can be measured using a dermal phase meter.

In various embodiments, the composition is formulated for topical application to the skin, such as the skin surrounding or including the eyes, mouth, nose, forehead, ears, neck, 15 hands, feet, hair, and/or overall body. For example, the topical skin care composition may be in the form of a solution, serum, cream, lotion, body milk, emulsion, balm, gel, soap, conditioner, powder and the like. Alternatively, the topical skin care composition may be in the form of a shampoo, conditioner, serum, or toner. In other embodiments, the composition is formulated for topical application to hair or scalp.

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In other embodiments, the composition is provided as an active ingredient in a composition formulated for topical application to the skin. In other embodiments, the composition is provided as an active ingredient in a composition formulated for topical application to hair or scalp. In other embodiments, the composition is provided as an active 25 ingredient in a composition formulated for use in wound healing. In other embodiments, the composition is provided as an active ingredient in a composition formulated for cosmetic use. In other embodiments, the composition is provided as an active ingredient in a composition formulated for use as a conditioning product for a subject seeking to improve their appearance. Various skin-related conditions include appearance of aging, wrinkles, fine 30 lines, thinness, diminished elasticity or suppleness, dry skin, undesirable appearance of pores, pronounced appearance of stretch marks and scars, undesirable color tone and hue, dermatitis, eczema, sunburn, inflammation, pruritic lesions, inflammatory and non-inflammatory lesions of the skin of a subject. Other conditions related to hair include

baldness (i.e., alopecia), reduced shaft volume, structural deformations (e.g., split ends), low elasticity, brittleness, dullness, dryness, slow growth, among others.

EXAMPLES

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Example 1

Sources of natural ingredients

Natural ingredients may be purified or isolated from a variety of plant and animal sources. For example, black mamaku, is an extract prepared from a black tree fern (*Cyathea medullaris*) endemic to New Zealand. Cehami, is an indigenous herb (*Centipeda Cunninghamii*) found in Australia. Pomegranate is a fruit-bearing tree (*Punica granatum*) from Central Asia. *Croton lechleri*, also known as Sangre de Grado (Dragon's blood), is a plant native to South America, whose latex is used by native peoples for wound healing. Cacao (*Theobroma cacao*) is the fruit-bearing tree whose seeds are used to make cocoa powder and chocolate. Maqui (*Aristotelia chilensis*) is a tree which produces berries and is found in Argentinian rainforest. Green tea (*Camellia sinensis*) has long been cultivated in Asia as a source of ingredients that provide various health benefits. Vanilla is obtained from orchids of the genus *Vanilla*, a native plant from Mexico. Fucoidan describes sulfated fucans obtained from algae, such as *Undaria pinnatifida*. *Aphanizomenon flos-aquae* (AFA) is a blue-green algae known to contain a variety of antioxidants and other biologically active compounds. Aloe may be obtained from species of the genus *Aloe*, a leafy plant originating from Africa, examples of *Aloe* species include *Aloe barbadensis* and *A. Africana*. Mushroom polysaccharides are glucans found mainly in various species of mushrooms such as *Cordyceps sinesis*, *Hericium erinaceous*, and *Ganoderma lucidum*. Colostrum is a fluid secreted by the mammary glands of female mammals during the first few days of lactation, a primary source being bovine animals. The specific structures or forms from which natural ingredients are derived from these species are obtained (e.g., leaf, bean, fruit, bark, etc.) are described in Tables 2 and 3.

Example 2

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Natural ingredients isolated from plants increase fibroblast proliferation

As shown in Figure 1, the following plants increased the proliferation of neonatal dermal fibroblasts: AFA (96%), aloe (87%), fucoidan from *Undaria pinnatifida* (55%), cehami (30%), cacao (54%), maqui (58%) and vanilla (25%). As shown in Figure 2, these results were extendible into adult fibroblasts cultured *in vitro*. Individual compounds

enhanced cell proliferation ranging from an increase of over 10% up to nearly 70% across various compounds when compared to untreated controls, thereby demonstrating a substantial increase in dermal fibroblast proliferation above baseline.

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Example 3

Synergetic effects achieved using combinations of natural ingredients

As shown in Figure 3, when blended together some of these ingredients showed a significant level of synergy in enhancing the proliferation of adult dermal fibroblasts. While AFA enhanced the proliferation of adult dermal fibroblasts by 26%, and aloe, fucoidan and 10 cehami individually reduced proliferation by 37%, 22% and 30%. Importantly, blends of these various ingredients enhanced proliferation by 42% to 86%. The strongest response was seen with blends of AFA, fucoidan and cehami (86%), aloe and fucoidan (74%), aloe, fucoidan, and cehami (71%), fucoidan and cehami (70%), AFA, aloe and fucoidan (70%), and a blend of all four ingredients (63%). Blending of AFA with cacao and vanilla also 15 created a synergy that increased the proliferation adult dermal fibroblasts by 30% and 15%. A vital component of skin, collagen, was also expressed at higher levels, following application of individual ingredients. As shown in Figure 4, collagen production by primary adult human dermal fibroblasts in culture was increased by fucoidan from *Undaria pinnatifida*, bovine colostrum extract, cytokines, cehami (*Centipeda cunninghamii*), and amla (20 *Phyllanthus emblica*). Whereas control untreated fibroblasts secreted approximately 0.3 µg/mL of collagen, treated samples secreted amounts ranging from 0.8 to over 1.2 µg/mL of collagen.

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Example 4

Variable effects of natural ingredients in neonatal and adult dermal fibroblasts

As shown in Figure 5A, an *in vitro* cellular bioassay wound healing model was applied, wherein adult human dermal fibroblasts were cultured until formation of a dense monolayer, a scratch was created through the monolayer, and following treatment with individual compounds, recovery was observed after 36 hours. Both neonatal and adult skin 30 fibroblasts were grown to confluence in wells and subjected to the scratch test prior to exposure to extracts from various plants in order to study the effect of herbal extracts on stem cell proliferation and migration. Results are shown in Table 1 and Figure 6. AFA, aloe, fucoidan, cehami, black mamaku, berries, cacao and green tea all significantly enhanced stem cell proliferation and migration. Vanilla, pomegranate and *Croton lechleri* showed moderate

effect. Application of a scoring system, as shown in Figure 5B, demonstrated that a variety of individual ingredients contained significant wound healing properties, with individual ingredients providing an improvement ranging from 20% up to nearly 60% scratch recovery compared to untreated controls.

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Table 1 Effect of Different Natural Ingredients on Proliferation and Migration

	Neonatal dermal fibroblasts		Adult dermal fibroblasts	
	Proliferation	migration	Proliferation	migration
AFA	++++	+++	++++	++
Aloe	++++	+++	++++	++
Fucoidan	++++	0	++++	0
Black mamaku	+	+	++++	-
Cehami	++	+	+++	-
Pomegranate	++	+	0	0
Croton lechleri	+	++	0	0
Berries*	++++	+++	+++	++
Cacao	+++	+++	0	++
Maqui	+++	++	+++	0
Green Tea (PE)	0	+	++	+++
Vanilla Burboun	++	0	0	++

* Wild Bilberry, Wild Ligonberry, Black Currant, Aronia, Pomegranate, Wild Blueberry, Concorde Grape, Sour Cherry, Wild Elderberry, Wild Cranberry, Red Raspberry, Black Raspberry

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Example 5

Effective anti-wrinkle results via topical application of a composition containing a combination of natural ingredients

A combination of natural ingredients was tested over 7, 14, and 28 day periods for conditioning of skin and wrinkle reduction. This combination, provided in a serum containing Coconut oil, Nilotica butter, Rosa mosqueta, Olivem 1000, Cocoa butter, Soy lecithin, Chilean Soapnut, NovHyal, Fucoidan Maritech, Moringa, Aloe vera, Genistein, Black mamaku, Cehami, Amla, AFA, Pomegranate, Sangre de drago, Guar, Vanilla, Colostrum, Cytokines, Maqui, Synergy berries, Green Tea Extract, Vanilla 20 fold extract, Cacao, Olive extract (Hydroxytyrolosol), Vitamin E (Sunflower), Samambaia, Bulgarian Rose, Jasmine, Sweet Orange, Ylang-ylang, Tangerine Essential Oil, Bitter Orange extract, and Honeysuckle, demonstrated effective results following both short term (e.g., 7 day) and longer term (e.g., 28 day application). For example, application of the serum over a short 7 day period resulted in a 74% wrinkle reduction as shown by the representative individual depicted in Figure 7A, and an 81% wrinkle reduction by the individual shown in Figure 7B,

as measured across the entirety of the individual's facial features. Common areas where wrinkles are pronounced, such as the base of the eyelids and forehead, similarly benefited from application of the serum. For example, as depicted in Figure 8, skin appearance is improved following 28 day application of combined ingredients, when using a combination 5 of ingredients twice daily for 28 days. In another representative example using Visioscan imaging, as depicted in Figure 7A, 28 day application led to an 82% reduction in wrinkles located near the base of the lower eyelid. A similarly remarkable reduction of about 75% was observed on the forehead, as depicted in Figure 7B.

Wrinkle reduction was further correlated to a variety of improved aesthetic features, 10 including moisture retention and improved skin elasticity. These improvements were measurable and statistically significant. For example, Figure 10A depicts measurements of skin properties. Visioscan image analysis system showed that combinations of ingredients applied over a 28-day period demonstrated dramatic decreases in surface roughness, with highly significant decreases appearing at 7 days (** indicating statistical significance, 15 p<0.001), and remained highly significant at 14 and 28 days of product use (** indicating statistical significance, p<0.02). Average reductions were 9.8%, 17.4%, and 25.3% after 7, 14, and 28 days of use, respectively, with maximum % improvement reaching 39.3%. Likewise, Figure 10B demonstrates that measurement of skin elasticity/flexibility via cutometer indicated an increase in biological elasticity on the test sites treated with the test 20 product. The increases were statistically significant from baseline after 14 and 28 days of use (as indicated by *), and averaged a 10% increase in elasticity, with maximum % improvement reaching 31.9%. Similarly, Figure 10C shows that when using Nova Dermal Phase meter for skin hydration measurement, combinations of ingredients dramatically increase the skin moisture content. The increases were statistically significant from baseline 25 after 14 and 28 days of use (as indicated by *) with average increases of 29.7% and 30.5%.

In an additional example demonstrating the results of these features together, Figure 11 shows that following serum application over 7, 14, and 28 day periods, measurements indicated up to a 25% reduction in wrinkles, up to a 30% increase in moisture retention and up to a 10% increase in elasticity.

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Example 6

Biochemical activity of combined ingredients affecting improved skin appearance

Without being bound by any particular theory, it is suggested that the improved appearance of skin results from minimizing biochemical activity associated with skin aging.

Two well-known factors influencing skin aging including the insults associated with oxidation, and damage caused by free radical exposure. To characterize the effects of individual compounds on these key biochemical processes, the inventors investigated the effects of individual compounds on cellular antioxidant protection using CAP-e bioassay. As shown in Figure 12, the *in vitro* data demonstrated potent antioxidant bioavailability by amla (*Phyllanthus emblica*), pomegranate (*Punica granatum*), Sangre de drago (*Croton lechleri*), and green tea (*Camellia sinensis*). At higher concentrations, nearly all tested compounds exhibited inhibitory activity of oxidative damage. Most notably, amla (*Phyllanthus emblica*), pomegranate (*Punica granatum*), and Sangre de drago (*Croton lechleri*), all showed potent inhibitory activity, even with concentrations as low as 0.21 g/L.

Similarly, the inventors observed individual compounds inhibiting free radical formation, as shown in Figure 13. The inhibitory effect of each ingredient on free radical formation by inflammatory cells placed under oxidative stress conditions was tested. The *in vitro* data demonstrated reduced free radical production by individual compounds, including fucoidan from *Undaria pinnatifida*, Shea butter, rosa mosqueta (*Rosa rubiginosa*), maqui berry (*Aristotelia chilensis*), vanilla (*Vanilla planifolia*), and green tea (*Camellia sinensis*). Various concentrations of individual compounds demonstrated pronounced reduction of up to 70% of free radical formation.

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Example 7

Compositions containing different combinations of natural ingredients

As shown as examples in Tables 2-6, different combinations of natural ingredients may be added together to vary the contents of a compositions for a skin care conditioning product. Table 2 provides general examples, with specific compositions compatible with the present invention being described in Tables 3-6. Across various compositions, the combinations of ingredient should constitute approximately about 12-15% of the final composition volume, as shown across the various examples.

Table 2 Various Examples of Variable Amounts of Natural Ingredients in Different Compositions

Ingredients	INCI	% (w/w)	g-ml / 50 ml	Form	Part used
Coconut oil	<i>Cocos Nucifera</i>	0 - 10%	0 – 5.0	oil	endosperm/ fruit
Nilotica butter	<i>Vitellaria nilotica</i>	0 – 3.6%	0 – 1.8	butter	nut
Rosa mosqueta	<i>Rosa Rubiginosa</i>	0 – 3.6%	0 – 1.8	oil	fruit
Olivem 1000	<i>Ceteryl Olivate</i>	0 – 6.0%	0 – 3.0	oil	fruit
Cocoa butter	<i>Theobroma cacao</i>	0 – 3.6%	0 – 1.8	butter	bean
Soy lecithin	<i>Glycine Max</i>	0 – 1.6%	0 - 0.8	oil	bean
Chilean Soapnut	<i>Quillaja Saponaria</i>	0 – 3.0%	0 – 1.5	Syrup	bark
NovHyal	<i>NAG6P</i>	0 – 4.0%	0 – 2.0	Aqueous	biotech
Fucoidan Maritech	<i>Laminaria Japonica</i>	0 – 3.0%	0 – 1.5	Powder	whole plant
Moringa	<i>Moringa Oleifera</i>	0 – 3.0%	0 – 1.5	Powder	leaf
Aloe vera	<i>Aloe Barbadensis</i>	0 – 2.0%	0 – 1.0	Powder	leaf (gel)
Genistein	soy	0 – 1.33%	0 – 0.66	Powder	bean
Black mamaku	<i>Cyathea Medularis</i> <i>Centipeda</i>	0 – 1.33%	0 – 0.66	Powder	frond
Cehami	<i>Cunninghamia</i>	0 – 1.33%	0 – 0.66	Powder	leaves
Amla	<i>Phyllanthus emblica</i> <i>Aphanizomenon flos-aquae</i>	0 – 1.33%	0 - 0.66	Powder	fruit
AFA	<i>Punica Granatum</i>	0 - 0.66%	0 - 0.33	Powder	whole plant
Pomegranate	<i>Croton lechleri</i> <i>Cyamopsis tetragonolobus</i>	0 - 0.66%	0 - 0.33	Powder	fruit
Sangre de drago	<i>Vanilla planifolia</i>	0 – 1.33%	0 - 0.66	Powder	sap
Guar	<i>first milk</i>	0 – 1.33%	0 - 0.66	Powder	seed
Vanilla	<i>E. coli</i>	0 - 0.20%**	0 - 0.10	Aqueous	cultures
Maqui	<i>Aristotelia Chilensis</i>	0 – 0.50%	0 - 0.25	Powder	berries
Synergy berries±	12 berries	0 – 0.50%	0 - 0.25	Powder	berries
Green Tea Extract	<i>Camellia sinensis</i>	0 – 0.50%	0 - 0.25	Powder	leaf
Vanilla 20 fold extract	<i>Vanilla planifolia</i>	0 – 0.44%	0 - 0.22	Powder	bean
Cacao	<i>Theobroma Cacao</i>	0 - 0.4%	0 - 0.20	Powder	bean
Olive extract	<i>Olea europaea</i>	0 – 2.0%	0 – 1.0	Powder	fruit
(Hydroxytyrolosol)	<i>Helianthus annuus</i>	0 – 1.6%	0 - 0.80	oil	seed
Vitamin E (Sunflower)					
Samambaia	<i>Polypodium leucotomos</i>	0 – 1.0%	0 - 0.50	Powder	leaf
Bulgarian Rose	<i>Rosa Damascena</i> <i>Jasminum Grandiflorum</i>	0 - 0.44%	0 - 0.22	oil	flower
Jasmine	<i>Citrus Sinensis</i>	0 - 0.48%	0 - 0.24	oil	flower
Sweet Orange	<i>Cananga Odorata</i>	0 - 0.80%	0 - 0.40	oil	peel
Ylang-ylang		0 - 0.24%	0 - 0.12	oil	flower
Tangerine Essential Oil	<i>Citrus Reticulata</i>	0 - 0.02%	0.01	oil	peel
Bitter Orange extract	<i>Citrus aurantium</i>	0 – 2.5%	0 – 1.25	Aqueous	seed
Honeysuckle	<i>Lonicera Japonica</i>	0 – 2.5%	0 – 1.25	Aqueous	flower

*aFGF, bFGF, KGF2, VEGF, SCF 1/3 **varies depending on stock concentration. For example a 0.2%

concentration assumes 50 ppm stock solution.

± Wild Bilberry, Wild Ligonberry, Black Currant, Aronia, Pomegranate, Wild Blueberry, Concorde Grape, Sour Cherry, Wild Elderberry, Wild Cranberry, Red Raspberry, Black Raspberry

Table 3 Specific Example of Natural Ingredients for a Serum Composition

Ingredients	INCI	% (w/w)	g-ml / 50 ml	Form	Part used
Coconut oil	<i>Cocos Nucifera</i>	5.0%	2.5	oil	endosperm/fruit
Nilotica butter	<i>Vitellaria nilotica</i>	2.0%	1.0	butter	nut
Rosa mosqueta	<i>Rosa Rubiginosa</i>	1.8%	0.9	oil	fruit
Olivem 1000	<i>Ceteryl Olivate</i>	3.0%	1.5	oil	fruit
Cocoa butter	<i>Theobroma cacao</i>	1.8%	0.9	butter	bean
Soy lecithin	<i>Glycine Max</i>	0.80%	0.4	oil	bean
Chilean Soapnut	<i>Quillaja Saponaria</i>	1.5%	0.75	Syrup	bark
NovHyal	<i>NAG6P</i>	1.8%	.9	Aqueous	biotech
Fucoidan Maritech	<i>Laminaria Japonica</i>	2.0%	1	Powder	whole plant
Moringa	<i>Moringa Oleifera</i>	1.0%	0.5	Powder	leaf
Aloe vera	<i>Aloe Barbadensis</i>	1.0%	0.5	Powder	leaf (gel)
Genistein	soy	0.66%	0.33	Powder	bean
Black mamaku	<i>Cyathea Medularis</i>	0.33%	0.165	Powder	frond
Cehami	<i>Centipeda Cunninghamii</i>	0.33%	0.165	Powder	leaves
Amla	<i>Phyllanthus emblica</i>	0.33%	0.165	Powder	fruit
AFA	<i>Aphanizomenon flos-aquae</i>	0.33%	0.165	Powder	whole plant
Pomegranate	<i>Punica Granatum</i>	0.33%	0.165	Powder	fruit
Sangre de drago	<i>Croton lechleri</i>	0.33%	0.165	Powder	sap
Guar	<i>Cyamopsis tetragonolobus</i>	0.66%	0.33	Powder	seed
Vanilla	<i>Vanilla planifolia</i>	0.66%	0.33	Powder	bean
Colostrum	first milk	0.33%	0.165	Powder	milk
Cytokines	<i>E. coli</i>	0.10%	0.05	Aqueous	cultures
Maqui	<i>Aristotelia Chilensis</i>	0.25%	0.125	Powder	berries
Synergy berries	12 berries	0.25%	0.125	Powder	berries
Green Tea Extract	<i>Camellia sinensis</i>	0.25%	0.125	Powder	leaf
Vanilla 20 fold extract	<i>Vanilla planifolia</i>	0.22%	0.11	Powder	bean
Cacao	<i>Theobroma Cacao</i>	0.20%	0.1	Powder	bean
Olive extract (Hydroxytyrolosol)	<i>Olea europaea</i>	2.0%	1.0	Powder	fruit
Vitamin E (Sunflower)	<i>Helianthus annuus</i>	0.8%	0.4	oil	seed
Samambaia	<i>Polypodium leucotomos</i>	0.5%	0.25	Powder	leaf
Bulgarian Rose	<i>Rosa Damascena</i>	0.22%	0.11	oil	flower
Jasmine	<i>Jasminum Grandiflorum</i>	0.24%	0.12	oil	flower
Sweet Orange	<i>Citrus Sinensis</i>	0.40%	0.2	oil	peel
Ylang-ylang	<i>Cananga Odorata</i>	0.12%	0.06	oil	flower
Tangerine Essential Oil	<i>Citrus Reticulata</i>	0.01%	0.005	oil	peel
Bitter Orange extract	<i>Citrus aurantium</i>	1.25%	0.625	Aqueous	seed
Honeysuckle	<i>Lonicera Japonica</i>	1.25%	0.625	Aqueous	flower

Table 4 Specific Example of Natural Ingredients for a Serum Composition

Ingredients	INCI	% (w/w)	g-ml / 50 ml	Form	Part used
Coconut oil	<i>Cocos Nucifera</i>	5.0%	2.5	oil	endosperm/ fruit
Nilotica butter	<i>Vitellaria nilotica</i>	1.8%	0.9	butter	nut
Rosa mosqueta	<i>Rosa Rubiginosa</i>	1.8%	0.9	oil	fruit
Olivem 1000	<i>Ceteryl Olivate</i>	3.0%	1.5	oil	fruit
Cocoa butter	<i>Theobroma cacao</i>	1.8%	0.9	butter	bean
Soy lecithin	<i>Glycine Max</i>	0.80%	0.4	oil	bean
Chilean Soapnut	<i>Quillaja Saponaria</i>	1.5%	0.75	Syrup	bark
NovHyal	<i>NAG6P</i>	2.0%	1	Aqueous	biotech
Fucoidan Maritech	<i>Laminaria Japonica</i>	1.5%	0.75	Powder	whole plant
Moringa	<i>Moringa Oleifera</i>	1.50%	0.75	Powder	leaf
Aloe vera	<i>Aloe Barbadensis</i>	1.0%	0.5	Powder	leaf (gel)
Genistein	soy	0.66%	0.33	Powder	bean
Black mamaku	<i>Cyathea Medularis</i>	0.66%	0.33	Powder	frond
Cehami	<i>Centipeda Cunninghamii</i>	0.66%	0.33	Powder	leaves
Amla	<i>Phyllanthus emblica</i>	0.66%	0.33	Powder	fruit
AFA	<i>Aphanizomenon flos-aquae</i>	0.33%	0.165	Powder	whole plant
Pomegranate	<i>Punica Granatum</i>	0.33%	0.165	Powder	fruit
Sangre de drago	<i>Croton lechleri</i>	0.33%	0.165	Powder	sap
Guar	<i>Cyamopsis tetragonolobus</i>	0.66%	0.33	Powder	seed
Vanilla	<i>Vanilla planifolia</i>	0.66%	0.33	Powder	bean
Colostrum	first milk	0.33%	0.165	Powder	milk
Cytokines	<i>E. coli</i>	0.10%	0.05	Aqueous	cultures
Maqui	<i>Aristotelia Chilensis</i>	0.25%	0.125	Powder	berries
Synergy berries	12 berries	0.25%	0.125	Powder	berries
Green Tea Extract	<i>Camellia sinensis</i>	0.25%	0.125	Powder	leaf
Vanilla 20 fold extract	<i>Vanilla planifolia</i>	0.22%	0.11	Powder	bean
Cacao	<i>Theobroma Cacao</i>	0.20%	0.1	Powder	bean
Olive extract	<i>Olea europaea</i>	1.0%	0.5	Powder	fruit
(Hydroxytyrolosol)	<i>Helianthus annuus</i>	0.8%	0.4	oil	seed
Vitamin E (Sunflower)					
Samambaia	<i>Polypodium leucotomos</i>	0.5%	0.25	Powder	leaf
Bulgarian Rose	<i>Rosa Damascena</i>	0.22%	0.11	oil	flower
Jasmine	<i>Jasminum Grandiflorum</i>	0.24%	0.12	oil	flower
Sweet Orange	<i>Citrus Sinensis</i>	0.40%	0.2	oil	peel
Ylang-ylang	<i>Cananga Odorata</i>	0.12%	0.06	oil	flower
Tangerine Essential Oil	<i>Citrus Reticulata</i>	0.01%	0.005	oil	peel
Bitter Orange extract	<i>Citrus aurantium</i>	1.25%	0.625	Aqueous	seed
Honeysuckle	<i>Lonicera Japonica</i>	1.25%	0.625	Aqueous	flower

Table 5 Specific Example of Natural Ingredients for a Serum Composition

Ingredients	INCI	% (w/w)	g-ml / 50 ml	Form	Part used
Coconut oil	<i>Cocos Nucifera</i>	5.0%	2.5	oil	endosperm/ fruit
Nilotica butter	<i>Vitellaria nilotica</i>	1.8%	0.9	butter	nut
Rosa mosqueta	<i>Rosa Rubiginosa</i>	1.3%	0.65	oil	fruit
Olivem 1000	<i>Ceteryl Olivate</i>	3.0%	1.5	oil	fruit
Cocoa butter	<i>Theobroma cacao</i>	1.8%	0.9	butter	bean
Soy lecithin	<i>Glycine Max</i>	0.80%	0.4	oil	bean
Chilean Soapnut	<i>Quillaja Saponaria</i>	2.0%	1.0	Syrup	bark
NovHyal	<i>NAG6P</i>	2.0%	1.0	Aqueous	biotech
Fucoidan Maritech	<i>Laminaria Japonica</i>	0.5%	0.25	Powder	whole plant
Moringa	<i>Moringa Oleifera</i>	2.0%	1.0	Powder	leaf
Aloe vera	<i>Aloe Barbadensis</i>	0.5%	0.25	Powder	leaf (gel)
Genistein	soy	0.66%	0.33	Powder	bean
Black mamaku	<i>Cyathea Medularis</i>	0.66%	0.33	Powder	frond
Cehami	<i>Centipeda Cunninghamii</i>	0.66%	0.33	Powder	leaves
Amla	<i>Phyllanthus emblica</i>	0.66%	0.33	Powder	fruit
AFA	<i>Aphanizomenon flos-aquae</i>	0.66%	0.33	Powder	whole plant
Pomegranate	<i>Punica Granatum</i>	0.66%	0.33	Powder	fruit
Sangre de drago	<i>Croton lechleri</i>	0.66%	0.33	Powder	sap
Guar	<i>Cyamopsis tetragonolobus</i>	0.66%	0.33	Powder	seed
Vanilla	<i>Vanilla planifolia</i>	0.66%	0.33	Powder	bean
Colostrum	first milk	0.33%	0.165	Powder	milk
Cytokines	<i>E. coli</i>	0.10%	0.05	Aqueous	cultures
Maqui	<i>Aristotelia Chilensis</i>	0.50%	0.25	Powder	berries
Synergy berries	12 berries	0.25%	0.125	Powder	berries
Green Tea Extract	<i>Camellia sinensis</i>	0.25%	0.125	Powder	leaf
Vanilla 20 fold extract	<i>Vanilla planifolia</i>	0.22%	0.11	Powder	bean
Cacao	<i>Theobroma Cacao</i>	0.20%	0.1	Powder	bean
Olive extract	<i>Olea europaea</i>	1.0%	0.5	Powder	fruit
(Hydroxytyrolosol)	<i>Helianthus annuus</i>	0.8%	0.4	oil	seed
Vitamin E (Sunflower)					
Samambaia	<i>Polypodium leucotomos</i>	0.75%	0.375	Powder	leaf
Bulgarian Rose	<i>Rosa Damascena</i>	0.24%	0.12	oil	flower
Jasmine	<i>Jasminum Grandiflorum</i>	0.22%	0.11	oil	flower
Sweet Orange	<i>Citrus Sinensis</i>	0.40%	0.2	oil	peel
Ylang-ylang	<i>Cananga Odorata</i>	0.12%	0.06	oil	flower
Tangerine Essential Oil	<i>Citrus Reticulata</i>	0.01%	0.005	oil	peel
Bitter Orange extract	<i>Citrus aurantium</i>	1.0%	0.5	Aqueous	seed
Honeysuckle	<i>Lonicera Japonica</i>	1.0%	0.5	Aqueous	flower

Table 6 Specific Example of Natural Ingredients for a Serum Composition

Ingredients	INCI	% (w/w)	g-ml / 50 ml	Form	Part used
Coconut oil	<i>Cocos Nucifera</i>	5.0%	2.5	oil	endosperm/ fruit
Nilotica butter	<i>Vitellaria nilotica</i>	2.0%	1.0	butter	nut
Rosa mosqueta	<i>Rosa Rubiginosa</i>	2.0%	1.0	oil	fruit
Olivem 1000	<i>Ceteryl Olivate</i>	3.0%	1.5	oil	fruit
Cocoa butter	<i>Theobroma cacao</i>	2.0%	1.0	butter	bean
Soy lecithin	<i>Glycine Max</i>	0.40%	0.2	oil	bean
Chilean Soapnut	<i>Quillaja Saponaria</i>	1.3%	0.65	Syrup	bark
NovHyal	<i>NAG6P</i>	1.5%	0.75	Aqueous	biotech
Fucoidan Maritech	<i>Laminaria Japonica</i>	2.0%	1.0	Powder	whole plant
Moringa	<i>Moringa Oleifera</i>	1.50%	0.75	Powder	leaf
Aloe vera	<i>Aloe Barbadensis</i>	1.0%	0.5	Powder	leaf (gel)
Genistein	soy	0.66%	0.33	Powder	bean
Black mamaku	<i>Cyathea Medularis</i>	0.66%	0.33	Powder	frond
Cehami	<i>Centipeda Cunninghamii</i>	0.66%	0.33	Powder	leaves
Amla	<i>Phyllanthus emblica</i>	0.33%	0.165	Powder	fruit
AFA	<i>Aphanizomenon flos-aquae</i>	0.66%	0.33	Powder	whole plant
Pomegranate	<i>Punica Granatum</i>	0.66%	0.33	Powder	fruit
Sangre de drago	<i>Croton lechleri</i>	0.66%	0.33	Powder	sap
Guar	<i>Cyamopsis tetragonolobus</i>	0.33%	0.165	Powder	seed
Vanilla	<i>Vanilla planifolia</i>	0.33%	0.165	Powder	bean
Colostrum	first milk	0.33%	0.165	Powder	milk
Cytokines	<i>E. coli</i>	0.10%	0.05	Aqueous	cultures
Maqui	<i>Aristotelia Chilensis</i>	0.50%	0.25	Powder	berries
Synergy berries	12 berries	0.50%	0.25	Powder	berries
Green Tea Extract	<i>Camellia sinensis</i>	0.50%	0.25	Powder	leaf
Vanilla 20 fold extract	<i>Vanilla planifolia</i>	0.22%	0.11	Powder	bean
Cacao	<i>Theobroma Cacao</i>	0.20%	0.1	Powder	bean
Olive extract	<i>Olea europaea</i>	0.25%	0.125	Powder	fruit
(Hydroxytyrolosol)	<i>Helianthus annuus</i>	0.8%	0.4	oil	seed
Vitamin E (Sunflower)					
Samambaia	<i>Polypodium leucotomos</i>	0.5%	0.25	Powder	leaf
Bulgarian Rose	<i>Rosa Damascena</i>	0.22%	0.11	oil	flower
Jasmine	<i>Jasminum Grandiflorum</i>	0.24%	0.12	oil	flower
Sweet Orange	<i>Citrus Sinensis</i>	0.40%	0.2	oil	peel
Ylang-ylang	<i>Cananga Odorata</i>	0.12%	0.06	oil	flower
Tangerine Essential Oil	<i>Citrus Reticulata</i>	0.01%	0.005	oil	peel
Bitter Orange extract	<i>Citrus aurantium</i>	1.25%	0.625	Aqueous	seed
Honeysuckle	<i>Lonicera Japonica</i>	1.25%	0.625	Aqueous	flower

The various methods and techniques described above provide a number of ways to carry out the invention. Of course, it is to be understood that not necessarily all objectives or advantages described may be achieved in accordance with any particular embodiment described herein. Thus, for example, those skilled in the art will recognize that the methods 5 can be performed in a manner that achieves or optimizes one advantage or group of advantages as taught herein without necessarily achieving other objectives or advantages as may be taught or suggested herein. A variety of advantageous and disadvantageous alternatives are mentioned herein. It is to be understood that some preferred embodiments specifically include one, another, or several advantageous features, while others specifically 10 exclude one, another, or several disadvantageous features, while still others specifically mitigate a present disadvantageous feature by inclusion of one, another, or several advantageous features.

Furthermore, the skilled artisan will recognize the applicability of various features from different embodiments. Similarly, the various elements, features and steps discussed 15 above, as well as other known equivalents for each such element, feature or step, can be mixed and matched by one of ordinary skill in this art to perform methods in accordance with principles described herein. Among the various elements, features, and steps some will be specifically included and others specifically excluded in diverse embodiments.

Although the invention has been disclosed in the context of certain embodiments and 20 examples, it will be understood by those skilled in the art that the embodiments of the invention extend beyond the specifically disclosed embodiments to other alternative embodiments and/or uses and modifications and equivalents thereof.

Many variations and alternative elements have been disclosed in embodiments of the present invention. Still further variations and alternate elements will be apparent to one of 25 skill in the art. Among these variations, without limitation, are the sources of natural ingredients and constituent products, combinations of natural ingredients therein, the manufacturing techniques used to create cosmetic products, and the particular use of the products created through the teachings of the invention. Various embodiments of the invention can specifically include or exclude any of these variations or elements.

30 In some embodiments, the numbers expressing quantities of ingredients, properties such as concentration, reaction conditions, and so forth, used to describe and claim certain embodiments of the invention are to be understood as being modified in some instances by the term “about.” Accordingly, in some embodiments, the numerical parameters set forth in the written description and attached claims are approximations that can vary depending upon

the desired properties sought to be obtained by a particular embodiment. In some embodiments, the numerical parameters should be construed in light of the number of reported significant digits and by applying ordinary rounding techniques. Notwithstanding that the numerical ranges and parameters setting forth the broad scope of some embodiments 5 of the invention are approximations, the numerical values set forth in the specific examples are reported as precisely as practicable. The numerical values presented in some embodiments of the invention may contain certain errors necessarily resulting from the standard deviation found in their respective testing measurements.

In some embodiments, the terms “a” and “an” and “the” and similar references used 10 in the context of describing a particular embodiment of the invention (especially in the context of certain of the following claims) can be construed to cover both the singular and the plural. The recitation of ranges of values herein is merely intended to serve as a shorthand method of referring individually to each separate value falling within the range. Unless otherwise indicated herein, each individual value is incorporated into the specification as if it 15 were individually recited herein. All methods described herein can be performed in any suitable order unless otherwise indicated herein or otherwise clearly contradicted by context. The use of any and all examples, or exemplary language (e.g. “such as”) provided with respect to certain embodiments herein is intended merely to better illuminate the invention and does not pose a limitation on the scope of the invention otherwise claimed. No language 20 in the specification should be construed as indicating any non-claimed element essential to the practice of the invention.

Groupings of alternative elements or embodiments of the invention disclosed herein are not to be construed as limitations. Each group member can be referred to and claimed individually or in any combination with other members of the group or other elements found 25 herein. One or more members of a group can be included in, or deleted from, a group for reasons of convenience and/or patentability. When any such inclusion or deletion occurs, the specification is herein deemed to contain the group as modified thus fulfilling the written description of all Markush groups used in the appended claims.

Preferred embodiments of this invention are described herein, including the best mode 30 known to the inventors for carrying out the invention. Variations on those preferred embodiments will become apparent to those of ordinary skill in the art upon reading the foregoing description. It is contemplated that skilled artisans can employ such variations as appropriate, and the invention can be practiced otherwise than specifically described herein. Accordingly, many embodiments of this invention include all modifications and equivalents

of the subject matter recited in the claims appended hereto as permitted by applicable law. Moreover, any combination of the above-described elements in all possible variations thereof is encompassed by the invention unless otherwise indicated herein or otherwise clearly contradicted by context.

5 Furthermore, numerous references have been made to patents and printed publications throughout this specification. Each of the above cited references and printed publications are herein individually incorporated by reference in their entirety.

10 In closing, it is to be understood that the embodiments of the invention disclosed herein are illustrative of the principles of the present invention. Other modifications that can be employed can be within the scope of the invention. Thus, by way of example, but not of limitation, alternative configurations of the present invention can be utilized in accordance with the teachings herein. Accordingly, embodiments of the present invention are not limited to that precisely as shown and described.

CLAIMS

1. A purified skin care composition comprising:
 - one or more of the following components selected from the group consisting of: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof; and
 - a cosmetically acceptable carrier.
2. The purified skin care composition of claim 1, wherein the purified skin care composition comprises an emulsion substantially free of artificial ingredients.
3. A method of conditioning the skin of a subject, comprising:
 - providing a quantity of a purified composition, wherein the composition comprises one or more of the following components selected from the group consisting of: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof

thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof; and

administering a quantity of the purified composition to the subject in an amount sufficient to condition the skin of the subject.

4. The method of conditioning of claim 3, wherein conditioning the skin of the subject comprises an increase in progenitor cell, stem cell, and/or dermal fibroblast proliferation.
5. The method of conditioning of claim 3, wherein conditioning the skin of the subject comprises an increase in progenitor cell, stem cell, and/or dermal fibroblast mobilization.
6. The method of conditioning of claim 3, wherein conditioning the skin of the subject comprises an increase in progenitor cell, stem cell, and/or dermal fibroblast migration.
7. The method of conditioning of claim 3, wherein conditioning the skin of the subject comprises an increase in antioxidant protection.
8. The method of conditioning of claim 3, wherein conditioning the skin of the subject comprises inhibition of free radical formation.
9. A method of improving skin appearance in a subject, comprising:
providing a quantity of a purified composition, wherein the composition comprises one or more of the following components selected from the group consisting of: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe*

or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof; and

administering a quantity of the purified composition to the subject in an amount sufficient to improve skin appearance in a subject.

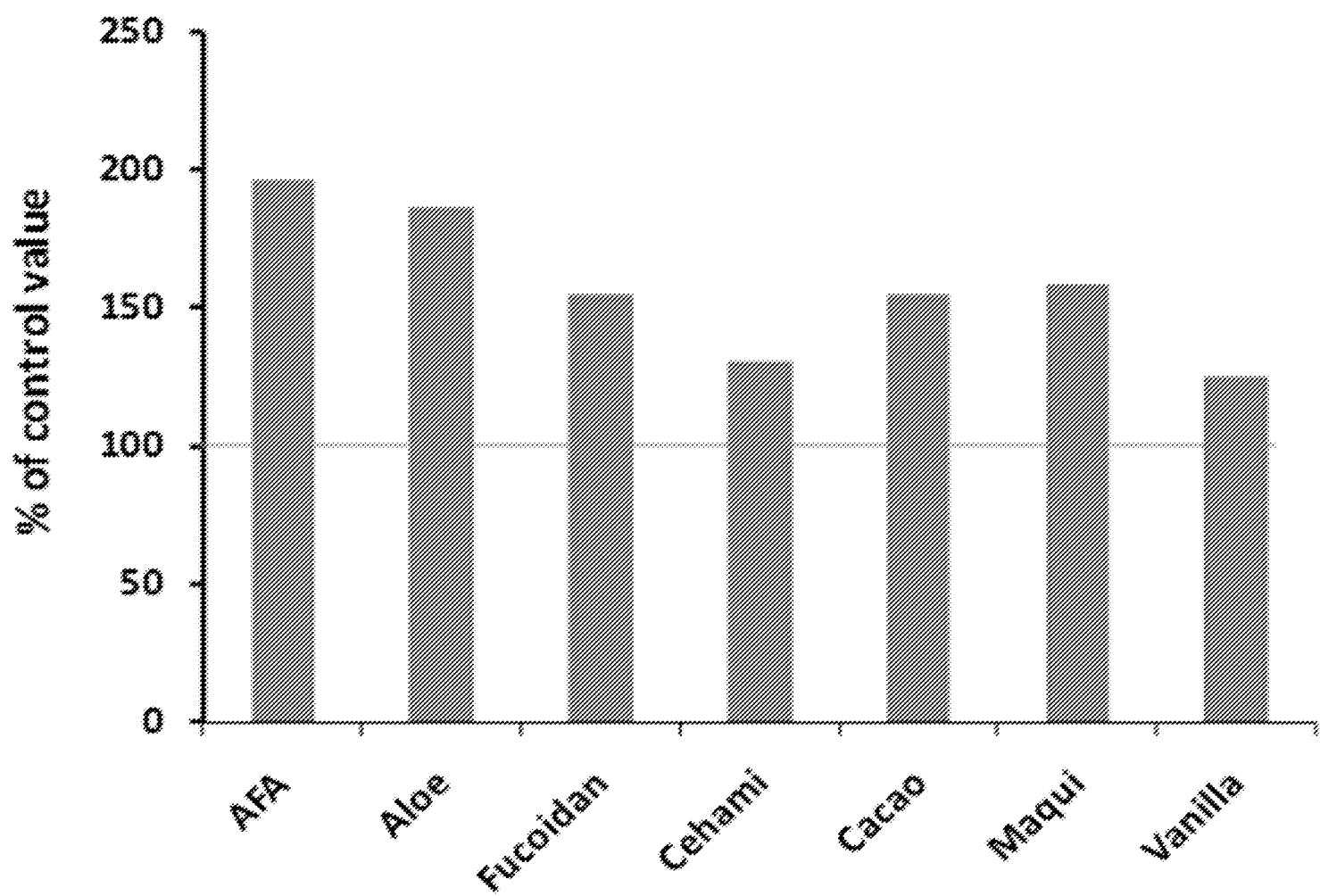
10. The method of claim 7, wherein the improved skin appearance comprises improved skin tone.
11. The method of claim 7, wherein the improved skin appearance comprises improved skin elasticity.
12. The method of claim 7, wherein the improved skin appearance comprises enhanced skin thickness.
13. The method of claim 7, wherein the improved skin appearance comprises improved skin hydration.
14. A method of reducing age-related features in the skin of a subject, comprising:
providing a quantity of a purified composition, wherein the composition comprises one or more of the following components selected from the group

consisting of: fucoidan or extracts thereof, *Moringa oleifera* or extracts thereof, *Aloe* or extracts thereof, soy or extracts thereof, *Cyathea medularis* or extracts thereof, *Centipeda cunninghamii* or extracts thereof, *Phyllanthus emblica* or extracts thereof, *Aphanizomenon flos-aquae* or extracts thereof, *Punica granatum* or extracts thereof, *Croton lechleri* or extracts thereof, colostrum or extracts thereof, *Citrus aurantium* or extracts thereof, *Lonicera japonica* or extracts thereof, *Olea europaea* or extracts thereof, *Polypodium leucotomos* or extracts thereof, *Camellia sinensis* or extracts thereof, *Aristotelia chilensis* or extracts thereof, berries or extracts thereof, *Theobroma cacao* or extracts thereof, *Cocos nucifera* or extracts thereof, *Vitellaria nilotica* or extracts thereof, *Rosa rubiginosa* or extracts thereof, *Ceteryl olivate* or extracts thereof, *Theobroma cacao* or extracts thereof, *Glycine max* or extracts thereof, *Citrus sinensis* or extracts thereof, *Quillaja saponaria* or extracts thereof, *Helianthus annuus* or extracts thereof, NAG6P, *E. coli* or extracts thereof, *Rosa damascena* or extracts thereof, *Jasminum grandiflorum* or extracts thereof, *Cananga odorata* or extracts thereof, *Citrus reticulata* or extracts thereof, and/or *Vanilla* or extracts thereof; and

administering a quantity of the purified composition to the subject in an amount sufficient to reducing age-related features in the skin of a subject.

15. The method of claim 14, wherein the age-related features comprise wrinkles.
16. The method of claim 14, wherein the age-related features comprise fine lines.
17. The method of claim 14, wherein the age-related features comprise dark patches of skin or age spots.

Figure 1



ure 2

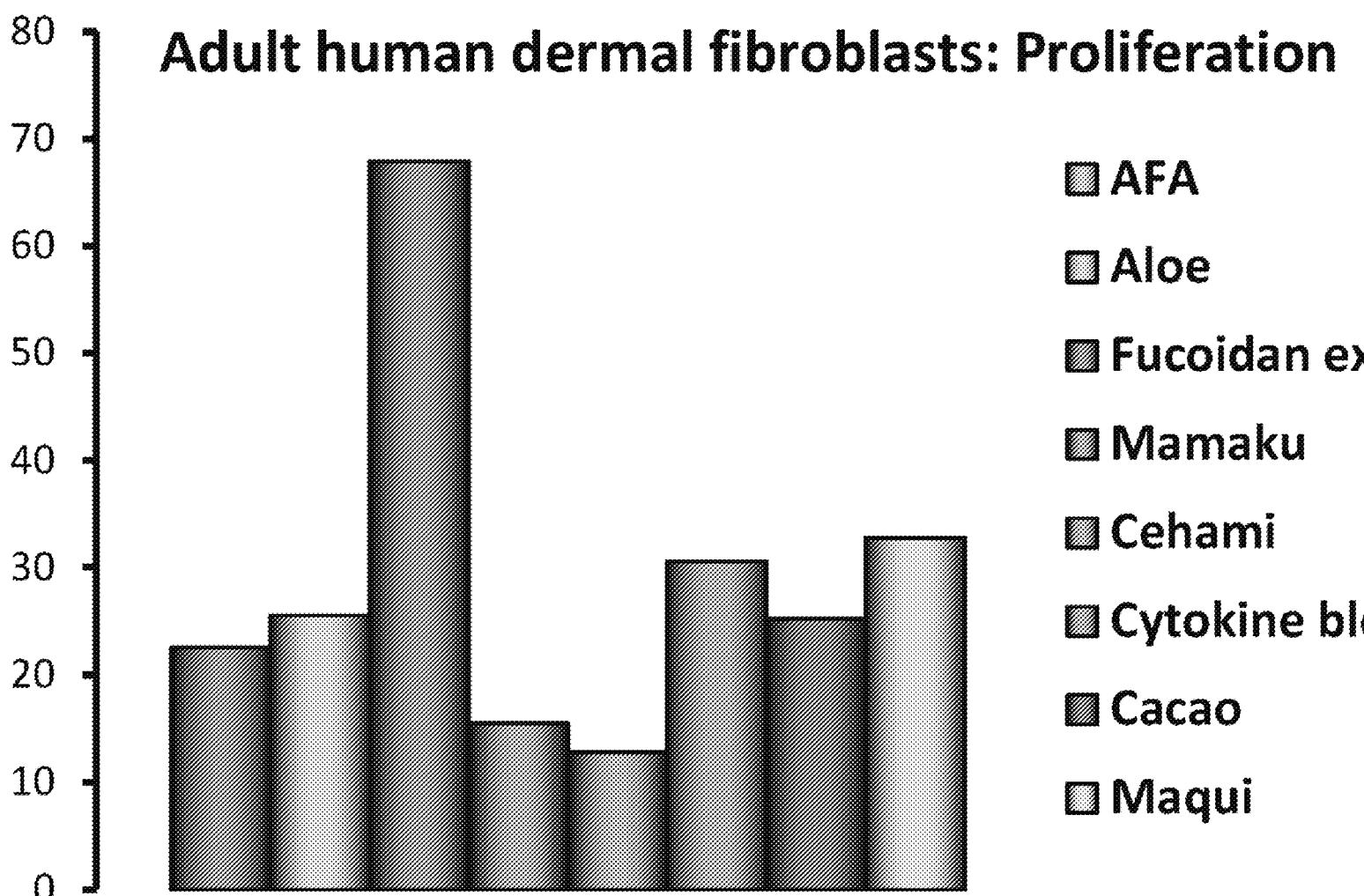
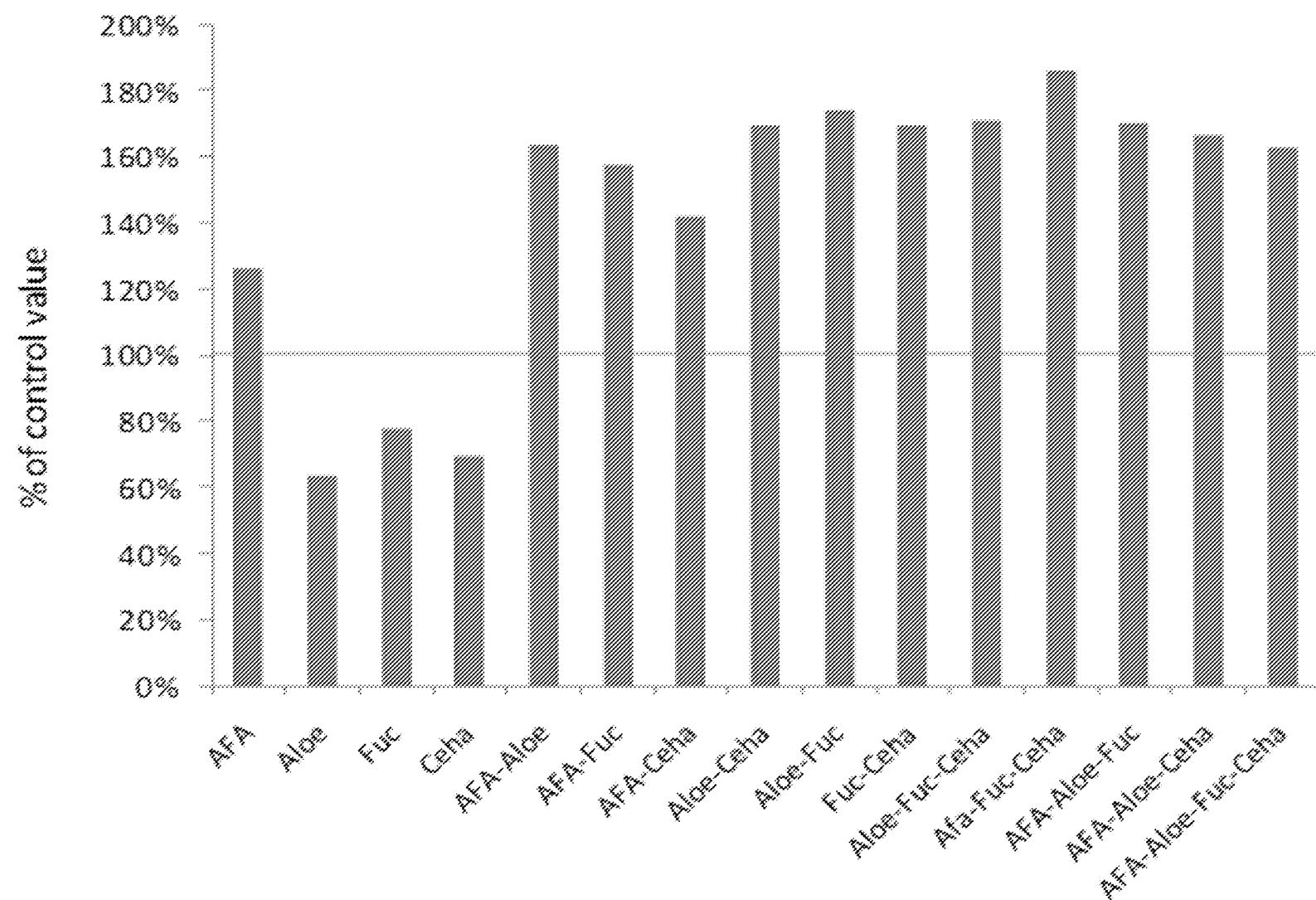
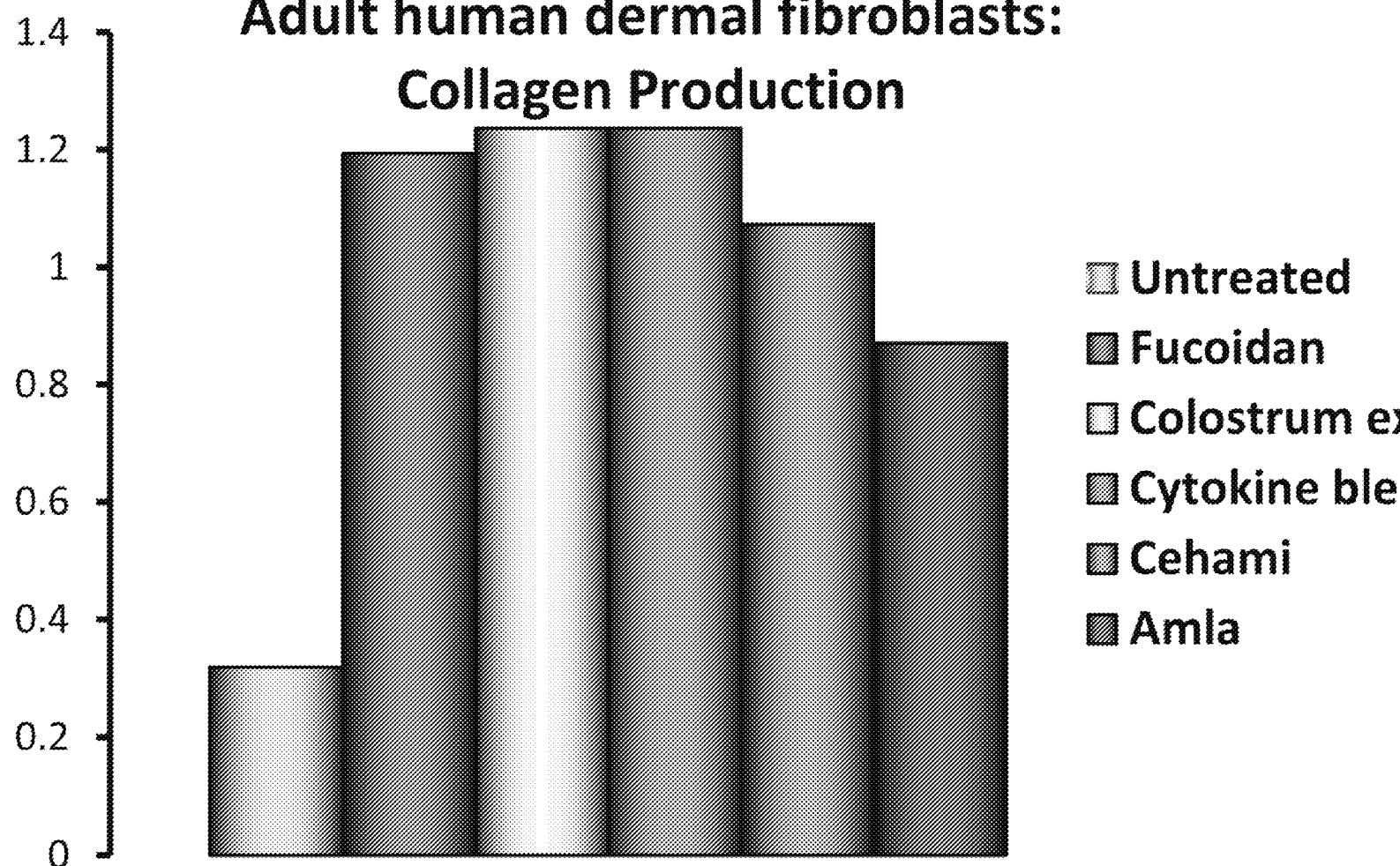


Figure 3

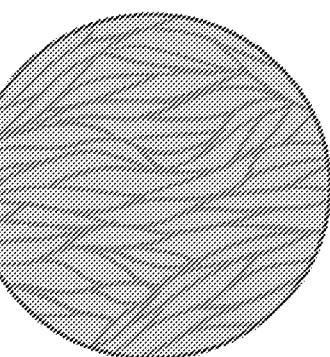


ure 4

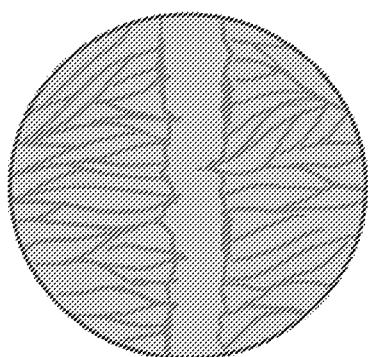
Adult human dermal fibroblasts: Collagen Production



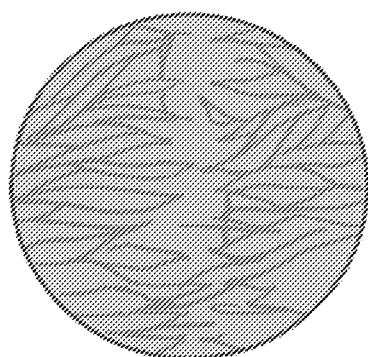
ure 5



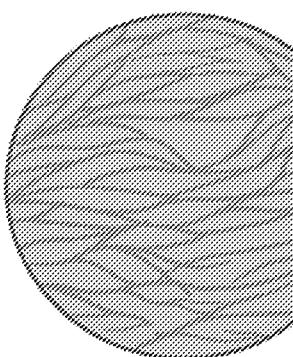
Dense cellular
monolayer
Score: 10



New in vitro
'wound'
Score: 0



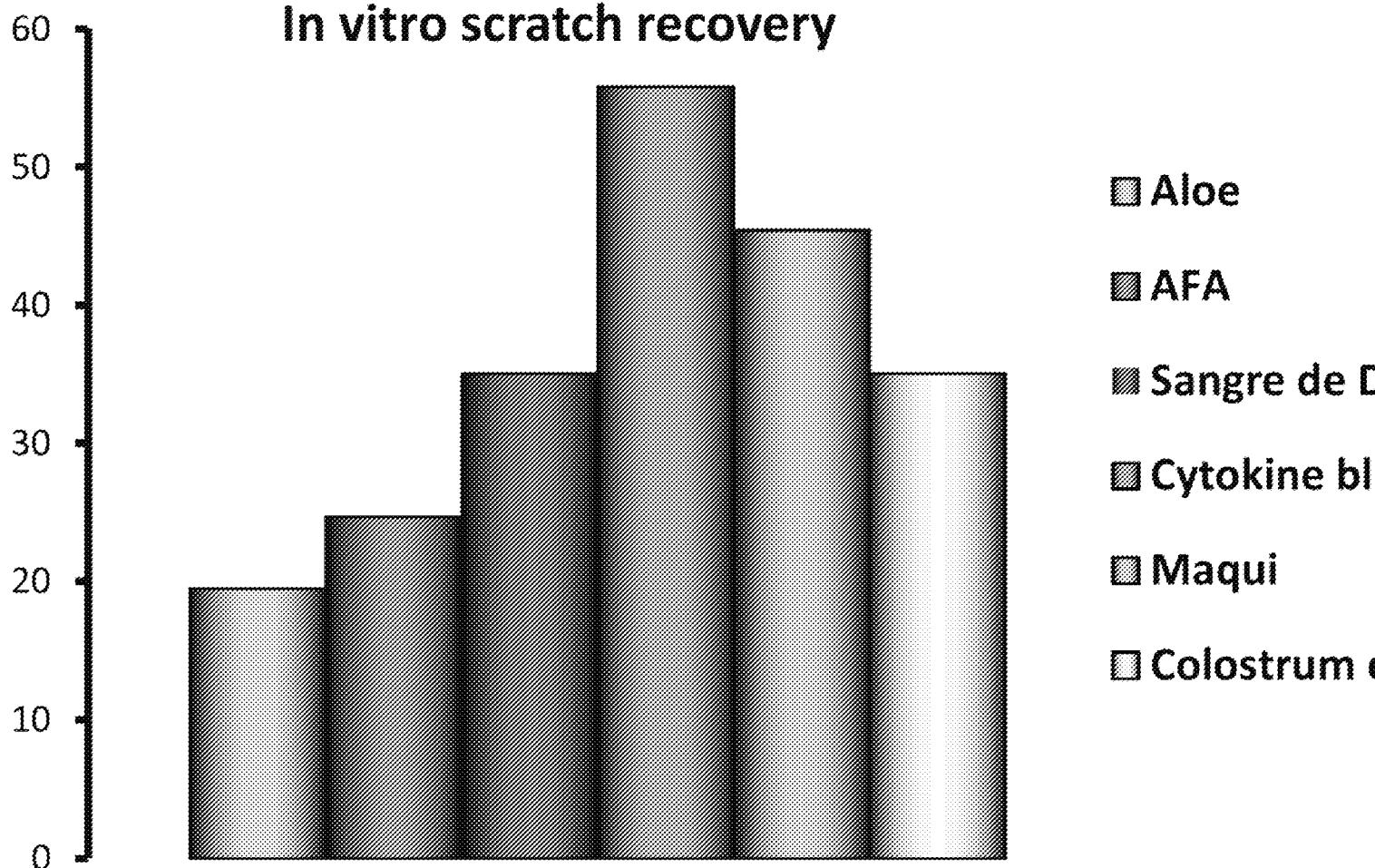
Recovering
monolayer
Score: 5

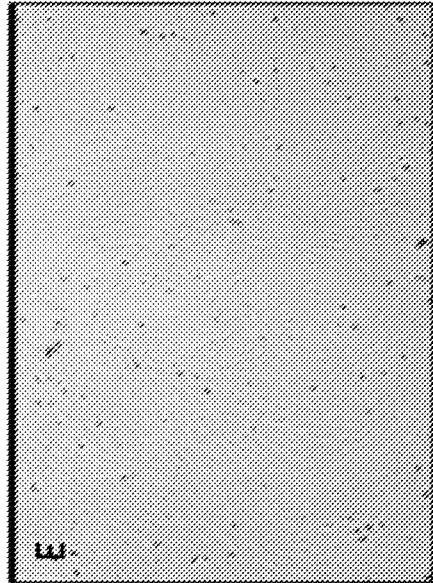
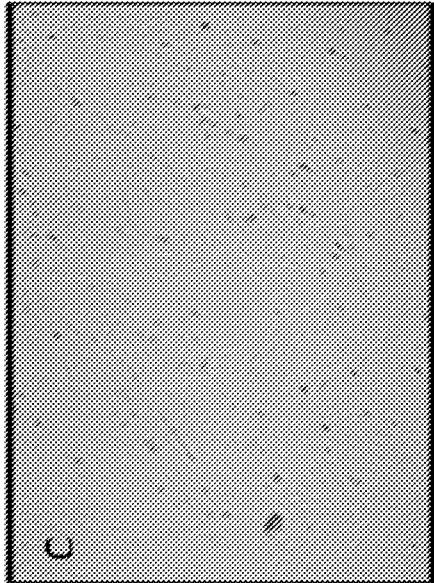
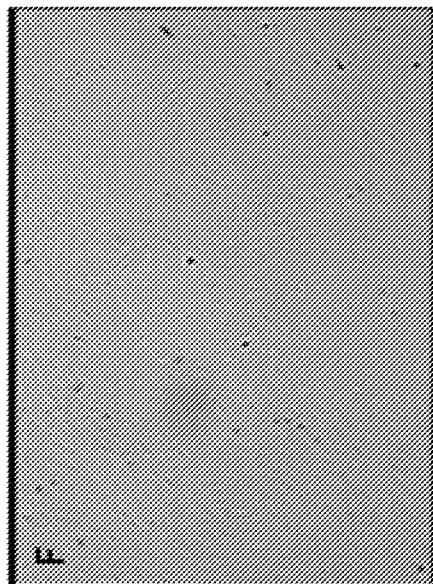
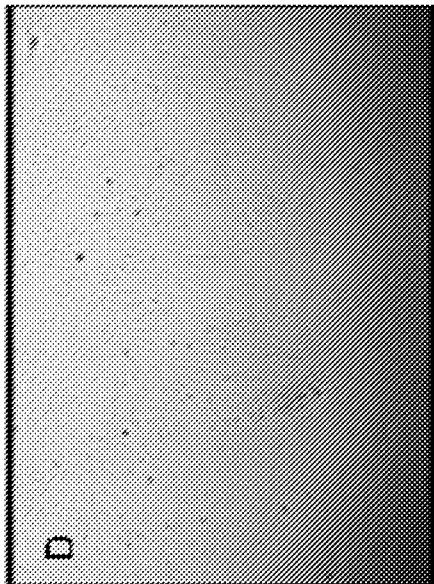
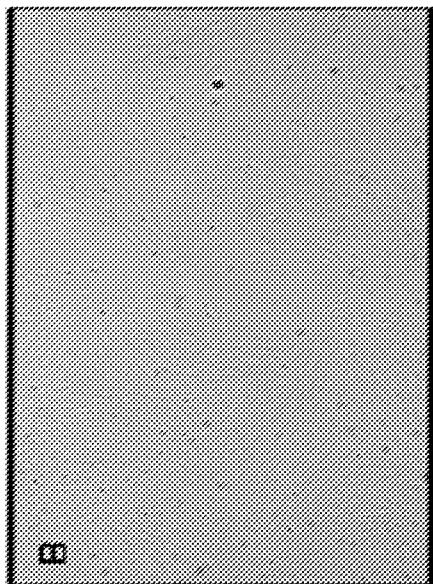


Recovering
monolayer
Score: 9

ure 5

Adult human dermal fibroblasts: In vitro scratch recovery





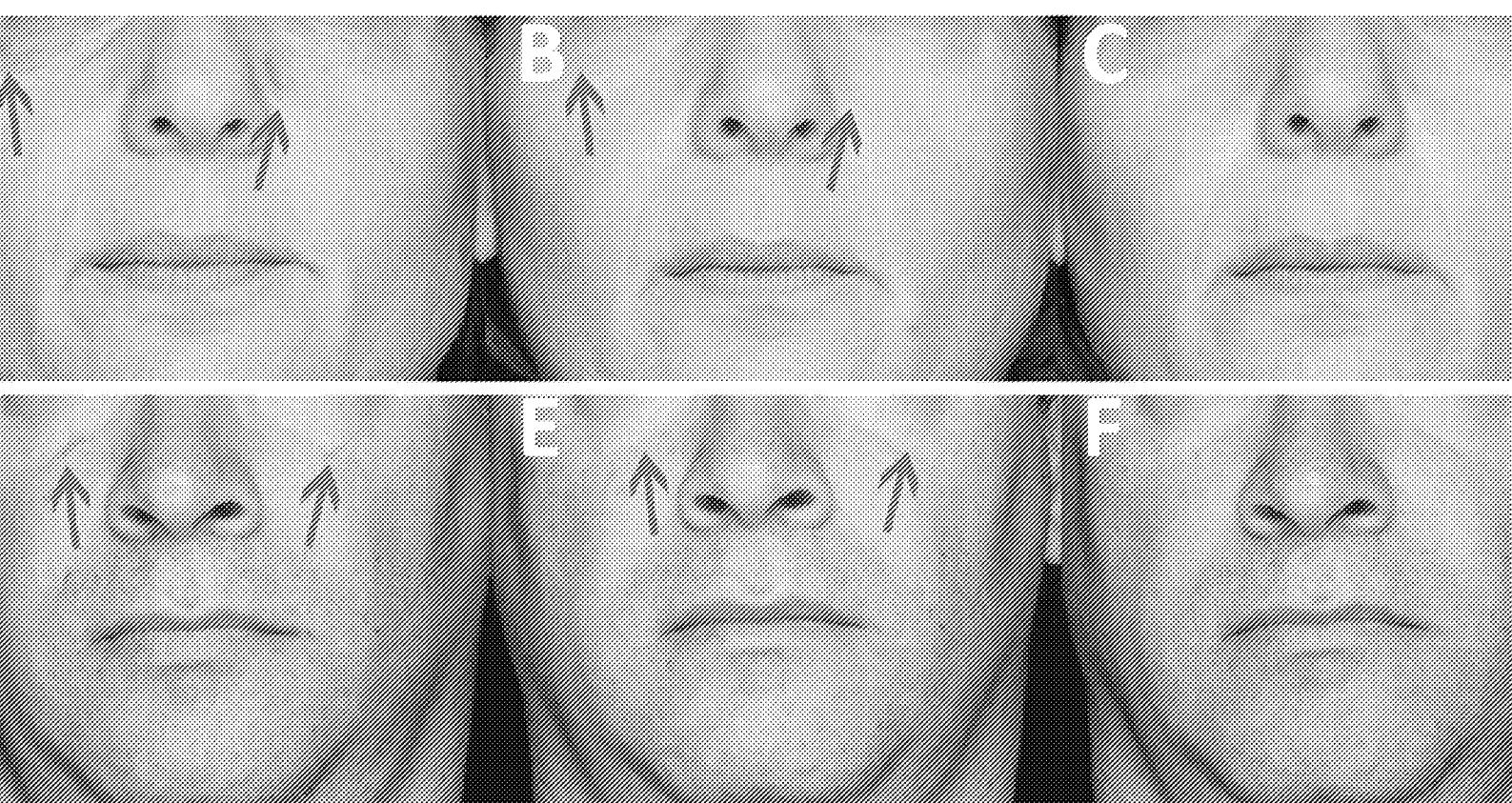
discrete / 1 day

A

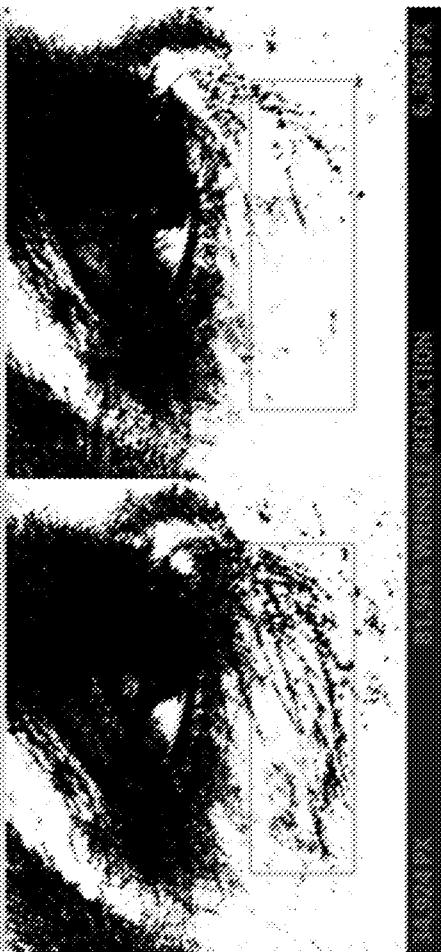


B

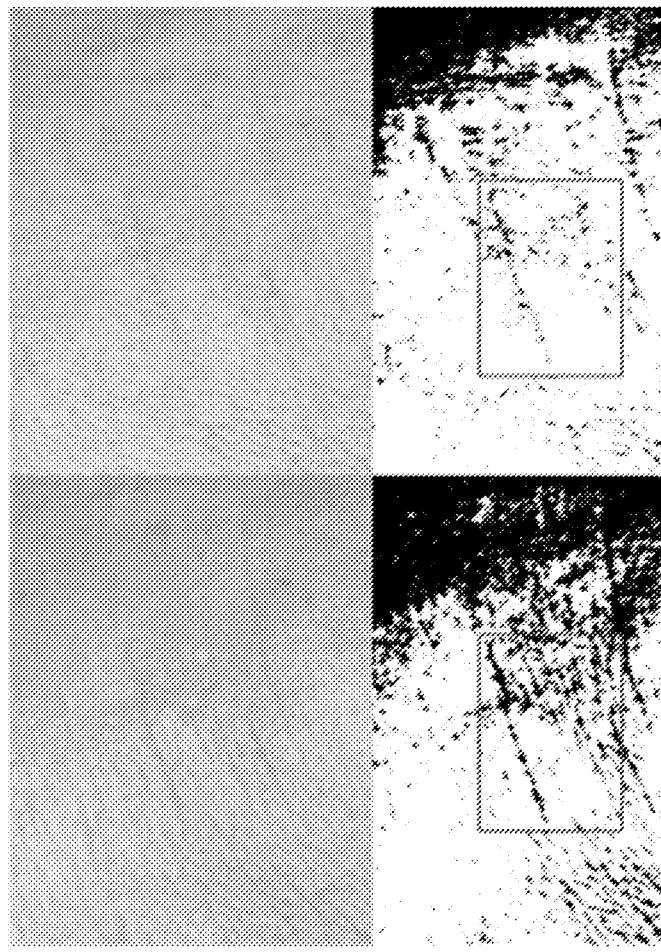
Figure 8



82% reduction in wrinkles



A



B

Figure 10

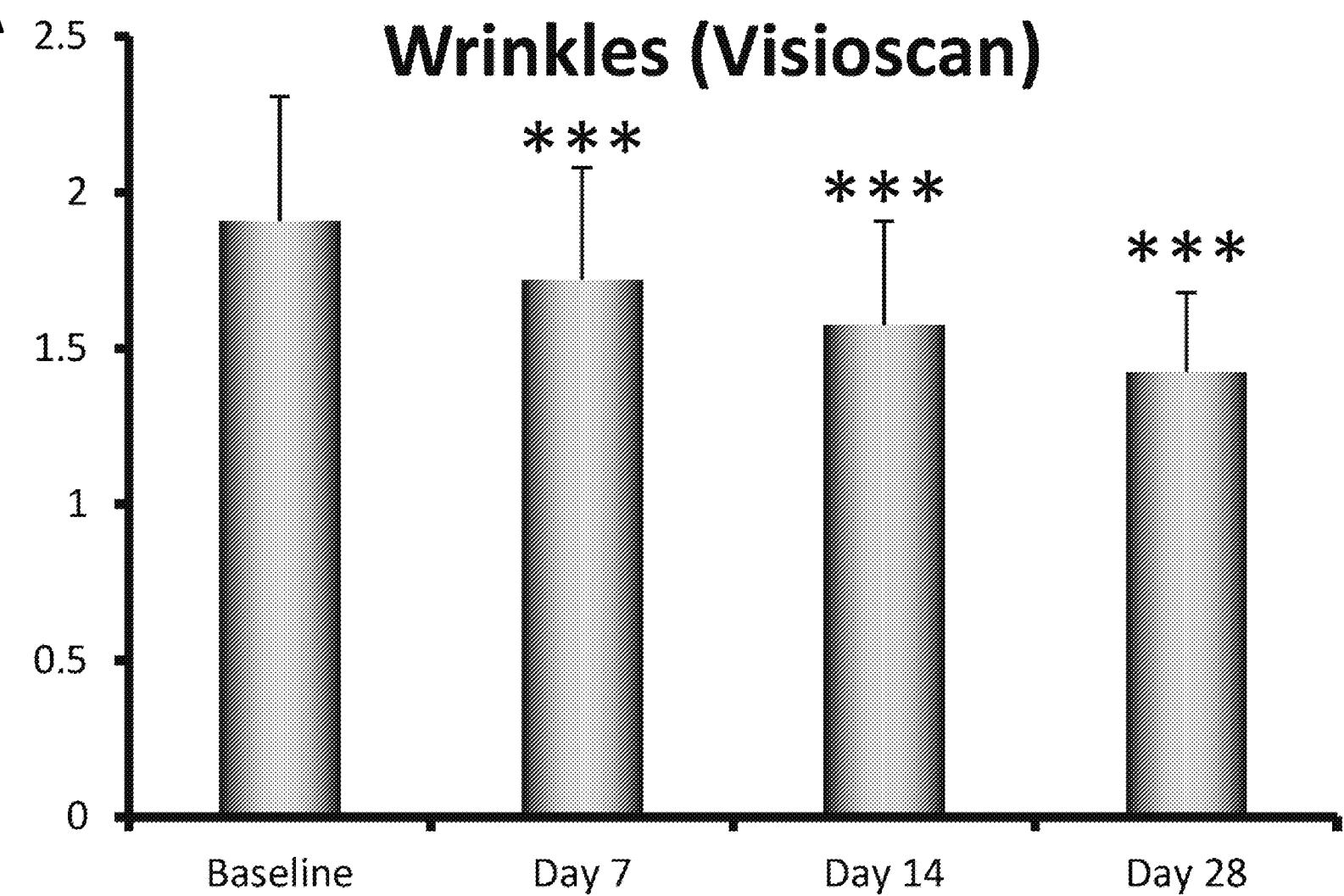


Figure 10

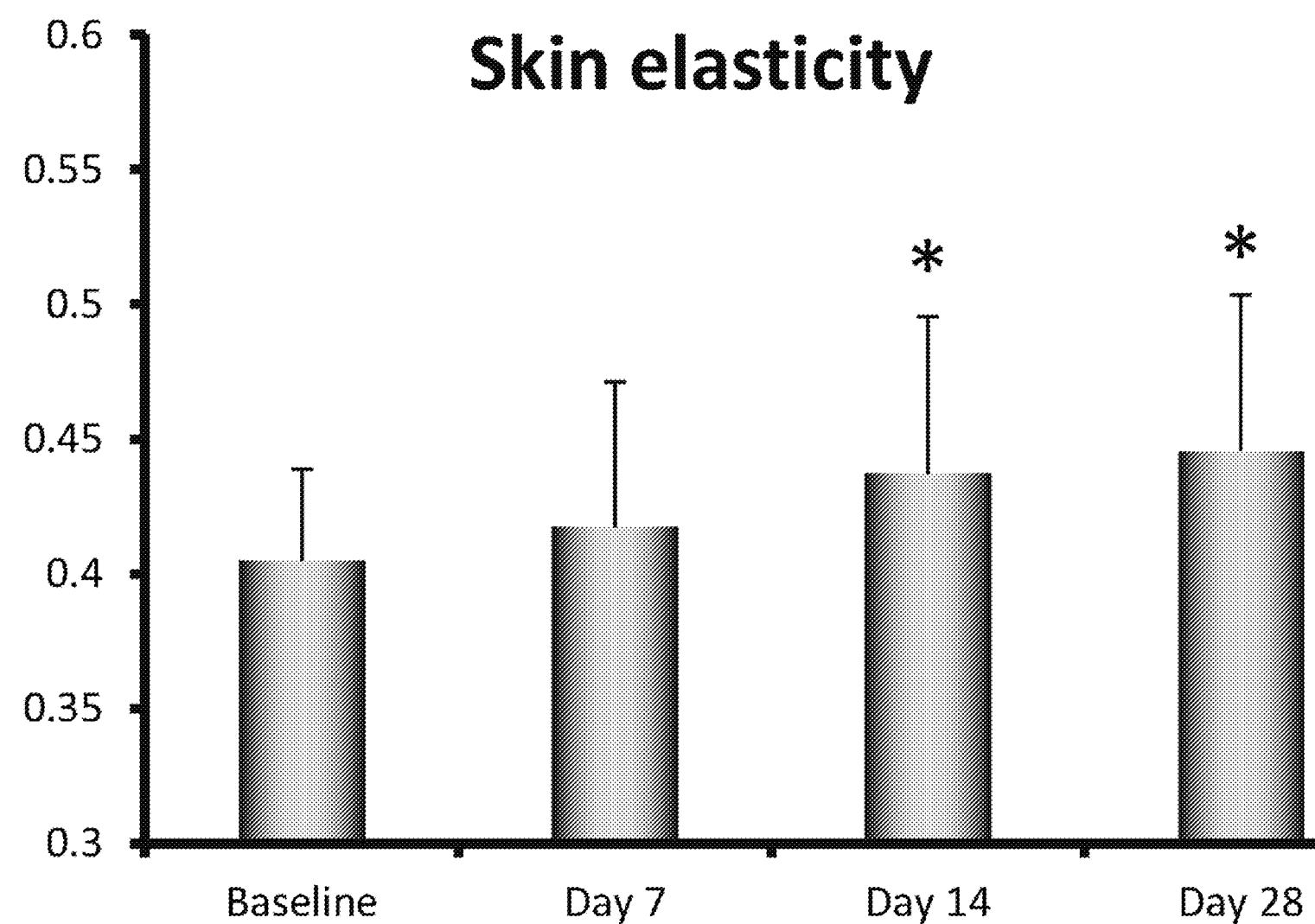
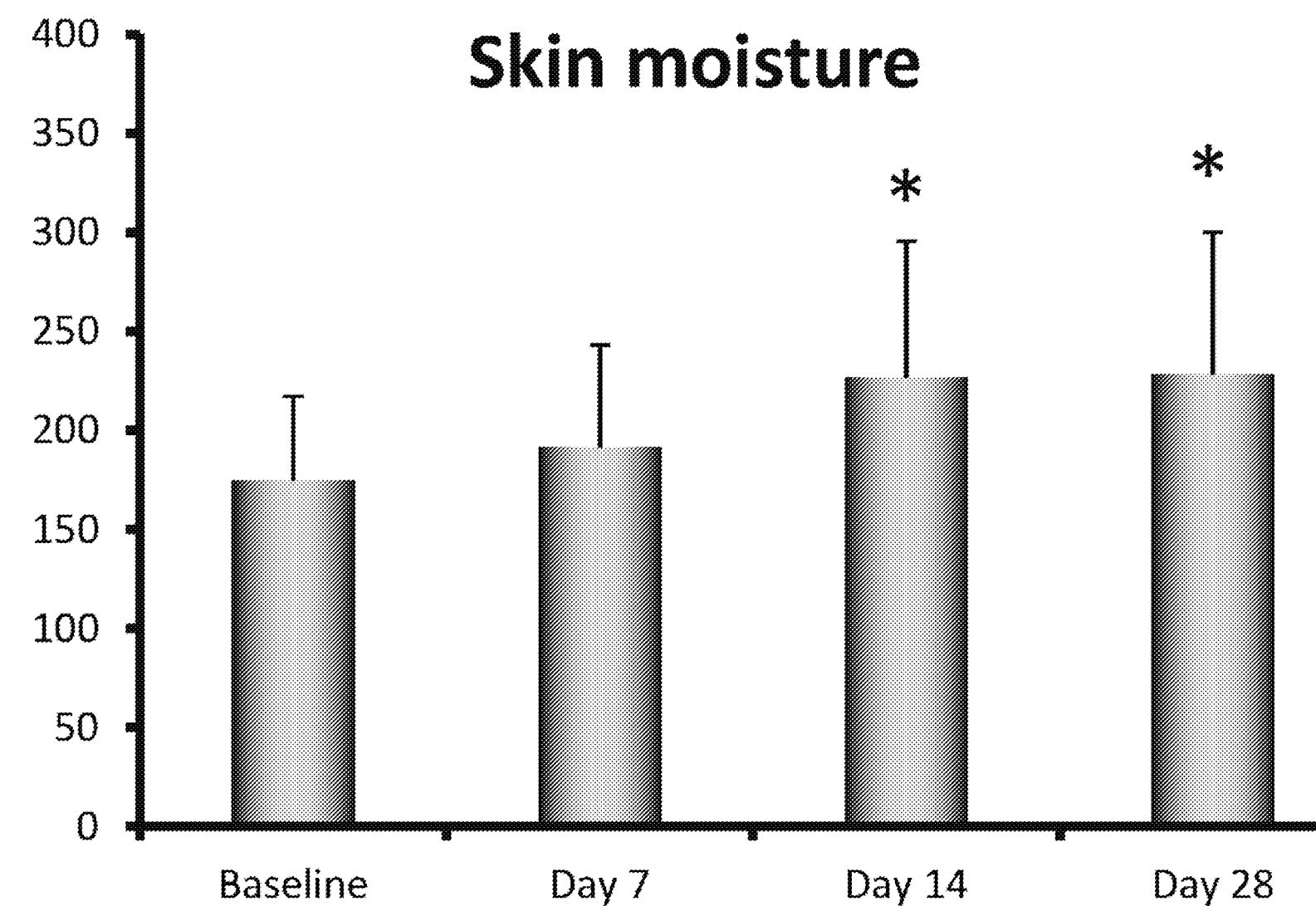
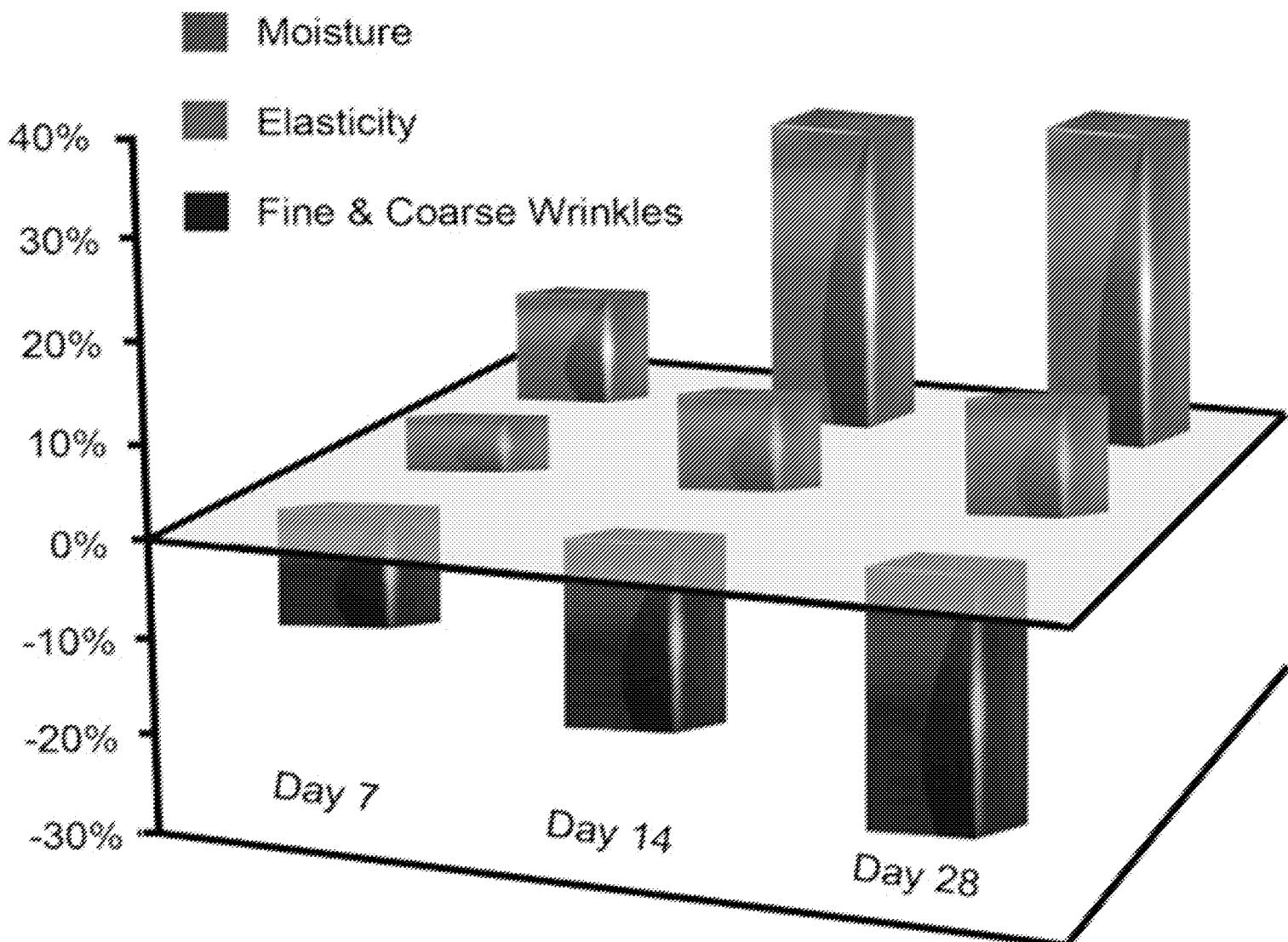


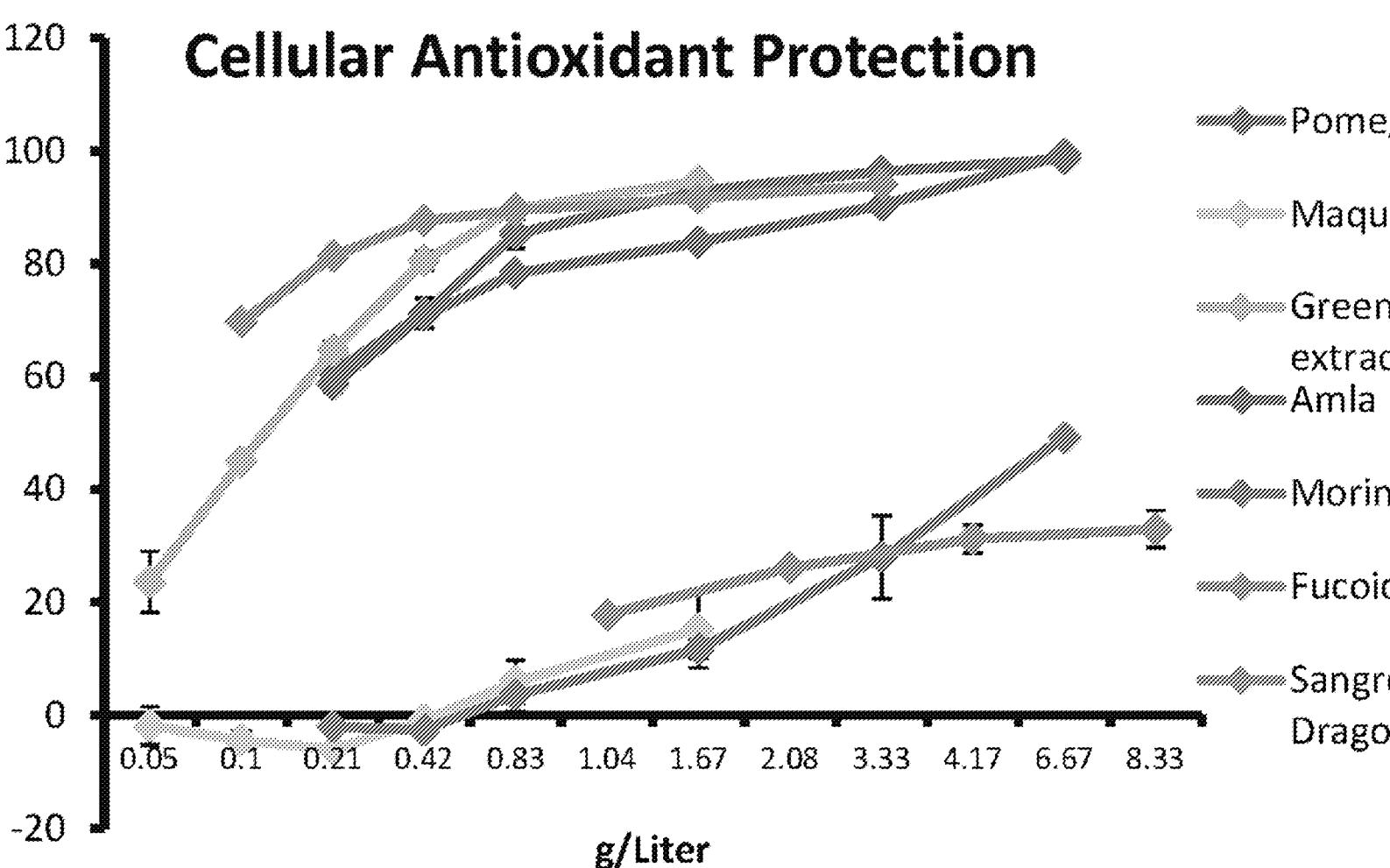
Figure 10



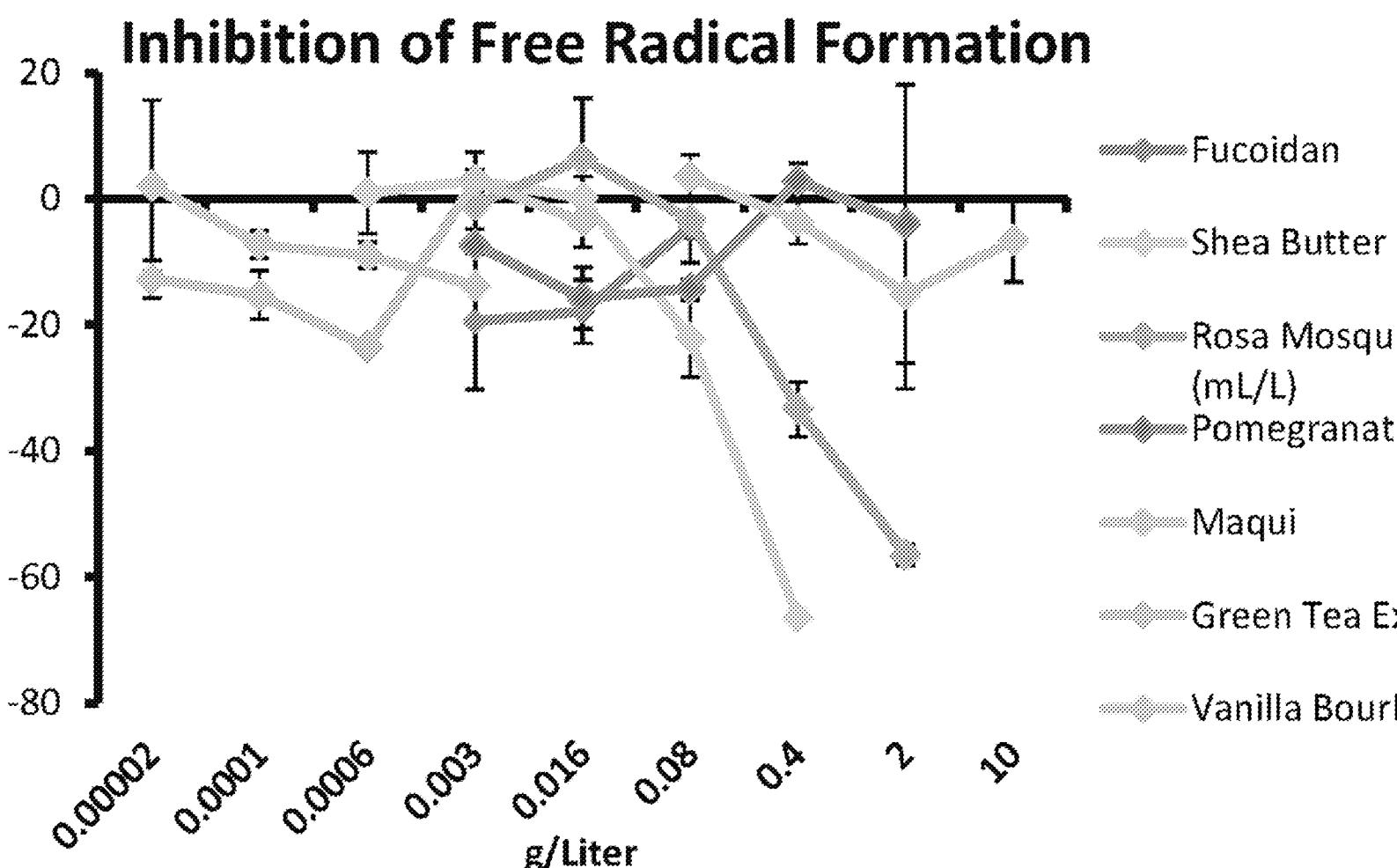
ure 11



ure 12



ure 13



INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 12/49619

A. CLASSIFICATION OF SUBJECT MATTER
 IPC(8) - A01N 65/00 (2012.01)
 USPC - 424/725

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
 USPC-424/725

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
 USPC-424/725,58,74

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
 Google, Google Scholar, Thomson Innovation, skin care, cosmetic, topical, anti-aging, anti-wrinkle, skin, moisturizer, conditioning,fucoidan, Moringa oleifera, Aloe, soy, Cyathea medularis, Centipeda cunninghamii, Phyllanthus emblica, Aphanizomenon flos aquae, Punica granatum, Croton lechleri, colostrum, Citrus aurantium, Lonicera japonica, Ole

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 2010/0233128 A1 (Panasenko) 16 September 2010 (16.09.2010) entire document esp. para [0004], [0019]-[0021], [0034]-[0036], [0044], [0053]-[0054], [0060]-[0061], [0073], [0082]	1-4, 7, 9-17
X	US 2007/0122492 A1 (Behr et al.) 31 May 2007 (31.05.2007) entire document esp. para[0095], [0102], [0111],[0167], [0179], [0193], [0197], [0291]	3, 5-6
X	US 2007/0178061 A1 (Venturi et. al.) 02 August 2007 (02.08.2007) entire document esp. para[0001], [0003], [0014]-[0016], [0033]	3, 8

Further documents are listed in the continuation of Box C.

* Special categories of cited documents:

“A” document defining the general state of the art which is not considered to be of particular relevance
 “E” earlier application or patent but published on or after the international filing date
 “L” document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
 “O” document referring to an oral disclosure, use, exhibition or other means
 “P” document published prior to the international filing date but later than the priority date claimed

“T” later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
 “X” document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
 “Y” document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
 “&” document member of the same patent family

Date of the actual completion of the international search 14 September 2012 (14.09.2012)	Date of mailing of the international search report 28 SEP 2012
Name and mailing address of the ISA/US Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-3201	Authorized officer: Lee W. Young PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 12/49619

Search Words,

skin care, cosmetic, topical, anti-aging, anti-wrinkle, skin, moisturizer, conditioning, fucoidan, *Moringa oleifera*, *Aloe*, soy, *Cyathea medularis*, *Centipeda cunninghamii*, *Phyllanthus emblica*, *Aphanizomenon flos aquae*, *Punica granatum*, *Croton lechleri*, colostrum, *Citrus aurantium*, *Lonicera japonica*, *Olea europaea*, *Polypodium leucotomos*, *Camellia sinensis*, *Aristolochia chilensis*, berries, *Theobroma cacao*, *Cocos nucifera*, *Vitellaria nilotica*, *Rosa rubiginosa*, *Ceteryl olivate*, *Glycine max* or *Quillaja saponaria*, *Helianthus annuus*, NAG6P, *E. coli*, *Rosa damascena*, *Jasminum grandiflorum*, *Cananga odorata*, *Citrus reticulata*, *Vanilla*, emulsion, oil, water, coconut oil, cocoa butter, olive oil, progenitor cell, stem cell, dermal fibroblast, antioxidant, free radical

摘要

本發明所述乃應用於和皮膚相關的各種天然成分。個別複合物為生產膠原蛋白，增殖成纖維細胞，抗氧化保護和抑制自由基提供有益作用。組合這些個別複合物能表現出協同效應，導致皮膚水分含量，抗皺和皮膚彈性的顯著改善。現提供各種組合成分的例子，可應用於調理皮膚及製作化妝品以改善外觀。