

【DESCRIPTION】

【Invention Title】

COMPOSITION FOR COMBATING OXIDATION AND ALLEVIATING
DETRIMENTAL EFFECTS ON SKIN CAUSED BY ASIAN DUST

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【Technical Field】

The present disclosure relates to a composition for combating oxidation and alleviating the detrimental effects on skin caused by Asian Dust, more particularly to a composition for combating oxidation and alleviating the detrimental effects on skin
10 caused by Asian Dust, which contains *Melia azedarach* bark and *Nelumbo nucifera* leaf as active ingredient.

【Background Art】

Skin is a part of the body exposed directly to the external environment. It
15 protects important organs of the body as a barrier, prevents excessive water loss and protects the body from infection. Although the skin provides strong protection even from external viruses, exposure to excessive UV, pollutants, etc. may lead to skin irritation such as erythema, edema, itchiness, etc. and inflammatory response. The skin trouble caused by these environmental factors is unattractive and reactive
20 oxygen species increased to such a level that the cell's antioxidant protection system cannot deal with are detrimental to DNAs, proteins, fats and sugars, negatively affect the cell's survival, growth and differentiation and may lead to cell death. Prolonged oxidative stress causes skin aging, cancer and inflammation (Ismail et al., Angiopoietin-1 reduces H₂O₂-induced increases in reactive oxygen species and

oxidative damage to skin cells. *J Invest Dermatol.* 130(5): 1307-17, 2010).

Asian Dust is a phenomenon which originates in the deserts of China, Mongolia, etc. whereby sand or yellow dust is carried eastward by winds and falls to the ground. Asian Dust affects Korea every spring. Asian Dust contains organic and inorganic matters and its physical properties and composition are various. It even includes metals that may have biological effects. Large particles included in the Asian Dust remain at the origin or around it and particulates with diameter of 10 μm or less (particulate matter 10; PM10) usually arrive in Korea. It is reported that the particulates can penetrate deep into the subsegmental bronchi and the gas-exchange regions of the lungs when inhaled and can damage the respiratory system (Yanagisawa et al., Gene expression analysis of murine lungs following pulmonary exposure to Asian sand dust particles. *Exp Biol Med* 232: 1109-1118, 2007, Kim et al., Cytotoxicity of yellow sand in lung epithelial cells. *J Biosci* 28(1): 77-81, 2003). The health effects of Asian Dust on humans have been studied a lot with regard to respiratory diseases. In an experimental study using mice, increase in oxidative stress-related indices was found in the lungs of mice to which Asian Dust was instilled (Naota et al., Pathological Study of Acute Pulmonary Toxicity Induced by Intratracheally Instilled Asian Sand Dust. *Toxicol Pathol.* 2010 Sep 30. [Epub ahead of print]).

Clinically, if the Asian Dust is exposed directly to the skin, it may cause contact dermatitis through allergic reactions. In addition, the dry and violent Asian Dust storm is known to cause skin dryness, keratinization, itchiness, acne, atopy, etc. by depriving the skin of moisture. Recently, regarding the detrimental effect of Asian Dust on the skin, it was reported for the first time that Asian Dust leads to increase in proinflammatory factors including the cytokines interleukin 6 and

interleukin 8 in keratinocytes (Choi et al., Asian dust storm particles induce a broad toxicological transcriptional program in human epidermal keratinocytes. *Toxicol Lett.* 15; 200(1-2): 92-9. 2010).

The keratinocytes present in the outermost layer of the skin play important roles in initiation, regulation and control of skin irritation response by interacting with fibroblasts, endothelial cells and dermal immune cells. Skin irritation is the most common detrimental effect related with subcutaneous inflammatory response and, clinically, 70% of contact dermatitis can be explained with skin irritation. Erythema, edema, crack, loss of water, exfoliation, itchiness and pain are typical symptoms of irritant or allergic contact dermatitis. They occur as a result of anomaly in the homeostasis of cells and inflammatory response. There have been few researches on skin irritation associated with Asian Dust. However, as the frequency of Asian Dust increases, development of a natural agent for external application to skin to prevent or treat skin irritation caused by Asian Dust is necessary.

【Disclosure】

【Technical Problem】

The present disclosure is directed to providing a composition for combating oxidation and alleviating the detrimental effects on skin caused by Asian Dust, which contains *Melia azedarach* bark and *Nelumbo nucifera* leaves as a novel skin irritation alleviating agent.

【Technical Solution】

In a general aspect, there is provided a composition for combating oxidation,

which contains *Melia azedarach* bark extract as an active ingredient.

In an exemplary embodiment of the present disclosure, the composition may further contain *Nelumbo nucifera* leaf extract.

5 In an exemplary embodiment of the present disclosure, the composition may suppress the production of reactive oxygen species or increase the expression of antioxidant protein genes.

In another general aspect, there is provided a composition for alleviating the detrimental effects on skin caused by Asian Dust, which contains *Melia azedarach* bark extract and *Nelumbo nucifera* leaf extract as an active ingredient.

10 In an exemplary embodiment of the present disclosure, the composition may increase the expression of GST, NQO1 or hBD3.

【Advantageous Effects】

A composition according to the present disclosure entails detoxification and
15 expulsion of toxins by increasing the expression of NQO1 which is an enzyme that detoxifies toxins and of GSTT1 which is an enzyme that expels toxins, protection of the skin by increasing the expression of the skin antimicrobial peptide hBD3 which is a natural immune factor and suppression of production of reactive oxygen species by increasing the expression of the antioxidant proteins GPx1, catalase, Trx1 and Prdx1.
20 Accordingly, the composition may be used as a pharmaceutical or cosmetic composition for preventing skin troubles caused by Asian Dust, protecting the skin and preventing and treating skin aging

【Description of Drawings】

25 Fig. 1 shows production of reactive oxygen species when normal human

keratinocytes are treated with a composition according to an exemplary embodiment of the present disclosure.

Figs. 2-4 show a result of analyzing the expression of NQO1 (Fig. 2), hBD3 (Fig. 3) and GSTT1 (Fig. 4) by polymerase chain reaction (RT-PCR) when normal human keratinocytes are treated with a composition according to an exemplary embodiment of the present disclosure.

Fig. 5 shows a result of analyzing the expression of GPx1, catalase, Trx1 and Prdx1 by polymerase chain reaction (RT-PCR) when normal human keratinocytes are treated with a composition according to an exemplary embodiment of the present disclosure.

【Best Mode】

Exemplary embodiments now will be described more fully hereinafter with reference to the accompanying drawings so that those of ordinary skill in the art can easily carry out the present disclosure.

The present disclosure provides a composition for combating oxidation, which contains *Melia azedarach* bark extract as an active ingredient.

The *Melia azedarach* bark refers to the bark or root bark of *Melia azedarach* (also called *Melia azedarach* L. var. *japonica* Makino) of the family *Meliaceae*. It is used to kill roundworms and threadworms and to treat rubella and scabies. It is also used to treat eczema and has diuretic and fever-alleviating effects. Although pharmacological effects against intestinal parasites such as tapeworm, roundworm, etc. and trichomonal vaginitis are reported, antioxidant effect has never been reported. The bark is curved with a tubular or semitubular shape. The external surface is grayish brown, with longitudinal fissures and transverse lenticels. The

peeled cork layer looks reddish brown. The snapped surface is yellowish white, fibrous and hard but easy to snap off. Young barks with many lenticels are good to be used as medicine. It is also called goryeonpi, ryeonpi, ryeongunmokpi, geumnyeongja, or the like.

5 The composition of the present disclosure may further contain *Nelumbo nucifera* leaf extract in addition to the *Melia azedarach* bark extract.

Nelumbo nucifera is a perennial aquatic plant with yellow flowers. It is widely used as food and also as various medicines in Eastern Asia, particularly in China. Specifically, the seed of *Nelumbo nucifera* is used to treat tissue
10 inflammation, cancer and leprosy, to detoxify poisons, or the like. And, the plumule of *Nelumbo nucifera* is reported to attenuate acute inflammation in vivo. Also, the leaf of *Nelumbo nucifera* is used in Chinese traditional medicine to stop bleeding and is known to alleviate hyperlipidemia in experimental animals.

The extraction method of the *Melia azedarach* bark or *Nelumbo nucifera* leaf
15 extract used in the present disclosure is not particularly limited. The extract may be obtained using water or an organic solvent. The organic solvent used in the present disclosure may be one or more selected from a group consisting of ethanol, methanol, butanol, ether, ethyl acetate and chloroform. Also, mixtures of these organic solvents with water may be used. In the examples described below,
20 extracts acquired from the Korea Plant Extract Bank.

Specifically, the *Melia azedarach* bark or *Nelumbo nucifera* leaf extract may be included in an amount of 1-30 wt% based on the total weight of the composition. If the total amount of the extract is less than 1 wt%, the desired effect may not be achieved. And, if the amount exceeds 30 wt%, a stability problem may occur.

25 The composition containing the *Melia azedarach* bark extract as an active

ingredient and the composition containing both the *Melia azedarach* bark extract and the *Nelumbo nucifera* leaf extract have antioxidant effect.

As can be seen from Test Example 1 and Fig. 1 which will be described later, the antioxidant effect can be achieved by suppressing the production of reactive oxygen species. Reactive oxygen species (ROS) are oxygen compounds having free electrons. To stabilize their unstable state, they attack lipoproteins of the cell membrane or intracellular DNAs, thereby causing cell damage, apoptosis, etc. and leading to various diseases and aging. Accordingly, skin aging can be prevented by reducing the reactive oxygen species.

As can be seen from Test Example 3 and Fig. 5 which will be described later, the antioxidant effect can be achieved by increasing the expression of antioxidant protein genes. The antioxidant protein gene includes GPx1, catalase, Trx1 and Prdx1, but is not limited thereto.

Glutathione peroxidase 1 (GPx1) is one of many glutathione peroxidases (GPx) and is encoded by the GPx1 gene in human. Glutathione peroxidases are involved in detoxification of hydrogen peroxide and are the most important antioxidant enzymes in human. For example, the protein may have the sequence Hs00829989_Gh.

Catalase is a common enzyme found in nearly all living organisms. It catalyzes the decomposition of hydrogen peroxide to water and oxygen. For example, the protein may have the sequence Hs00156308_m1.

Thioredoxin 1 (Trx1) is a cytoplasmic thioredoxin isoenzyme of the thioredoxin system which protects cells against oxidative and reductive stress. For example, the protein may have the sequence Hs01555212_g1.

Peroxiredoxin 1 (Prdx1) is a protein encoded by the PRDX 1 gene in human.

For example, the protein may have the sequence Hs00602020_Mh.

The present disclosure also provides a composition for alleviating the detrimental effects on skin caused by Asian Dust, which contains *Melia azedarach* bark extract and *Nelumbo nucifera* leaf extract as an active ingredient. More specifically, the composition of the present disclosure may alleviate the detrimental effects on normal keratinocytes in skin.

As can be seen from Test Example 2 and Figs. 2-4 which will be described later, the expression of the GST, NQO1 and hBD3 genes increase remarkably when both the *Melia azedarach* bark extract and the *Nelumbo nucifera* leaf extract are included as compared when either the *Nelumbo nucifera* leaf or the *Melia azedarach* bark extract is included alone. GSTT1 is an enzyme that expels toxins, NQO1 is an enzyme that detoxifies toxins and hBD3 is a skin antimicrobial peptide and natural immune factor. If the expression of detoxification-related enzymes including GST, NQO1 or hBD3 increases, skin protection can be achieved by alleviating skin irritation.

Glutathione S-transferases (GSTs) are classified into five classes. Glutathione S-transferase theta-1 (GSTT1), which is one of them, is an enzyme encoded by the GSTT1 gene and catalyzes the conjugation of reduced glutathione to a variety of electrophilic and hydrophobic compounds. For example, the protein may have the sequence Hs00184475_m1.

Quinone oxidoreductase 1 (NQO1) catalyzes the two-electron reduction of quinone. This enzyme provides protection from quinone-mediated carcinogenesis and activates bio-reductive alkylating agents. For example, the protein may have the sequence Hs01045994_m1.

Human beta defensin 3 (hBD3) induces migration and activation of antigen

presenting cells and functions as immune enhancer. For example, the protein may have the sequence hs00218678_m1.

The composition according to the present disclosure may be a cosmetic composition, a pharmaceutical composition or a health food composition.

5 Formulations of the cosmetic composition are not particularly limited. For example, it may be formulated into softening lotion, astringent lotion, nourishing lotion, nourishing cream, massage cream, essence, eye cream, eye essence, cleansing cream, cleansing lotion, cleansing foam, cleansing water, pack, beautifying essence, powder, body lotion, body cream, body oil, body essence, body cleanser,
10 hairdye, hair tonic, hair nourishing lotion, hair essence, hair serum, scalp treatment, hair treatment, hair conditioner, hair shampoo, hair lotion, toothpaste, gargle, hair fixative, hair restorer, lotion, ointment, gel, cream, patch, spray, etc.

The pharmaceutical composition may further contain a pharmaceutical adjuvant such as preservative, stabilizer, wetting agent, emulsifying promoter, salt
15 and/or buffer for control of osmotic pressure, etc. and other therapeutically useful substances and may be prepared into various formulations for oral or parenteral administration according to commonly employed methods.

Formulations for oral administration may include, for example, tablet, pill, hard and soft capsule, liquid, suspension, emulsion, syrup, granule, etc. and these
20 formulations may further contain, in addition to the active ingredient, a diluent (e.g., lactose, dextrose, sucrose, mannitol, sorbitol, cellulose or glycine) and a lubricant (e.g., silica, talc, stearic acid and its magnesium or calcium salt or polyethylene glycol). A tablet may contain a binder such as magnesium aluminum silicate, starch paste, gelatin, tragacanth, methyl cellulose, sodium carboxymethyl cellulose or
25 polyvinylpyrrolidone and, as occasion demands, it may contain a pharmaceutical

additive such as disintegrant (e.g., starch, agar, alginic acid or its sodium salt), absorbent, colorant, flavor, sweetener, etc. The tablet may be prepared by the common mixing, granulation or coating method.

And, formulations for parenteral administration may be in the form of an agent for external application to skin. For example, it may be in the form of lotion, ointment, gel, cream, patch or spray, but is not limited thereto.

Formulations of the health food composition are not particularly limited. For example, it may be formulated into tablet, granule, drink, caramel, diet bar, etc. Each formulation of the health food composition may contain, in addition to the active ingredient, other ingredients commonly used in the related art which are selected by those skilled in the art without difficulty. The additionally added ingredients may provide a synergic effect.

Determination of an administration dosage of the active ingredient is within the level of those skilled in the art. A daily administration dosage will vary depending on various factors including age and physical condition of the subject, presence of absence of complications, stage and severity of the condition to be treated, or the like. A general daily administration dosage for an adult of the composition may be 1-500 mg/kg, specifically 30-200 mg/kg. The administration may be made once or twice a day. However, the above-described administration dosage does not limit the scope of the present disclosure by any means.

【Mode for Invention】

Hereinafter, the present disclosure will be described in detail through examples. However, the following examples are for illustrative purposes only and it will be apparent to those of ordinary skill in the art that the scope of the present

disclosure is not limited by the examples.

[Test Example 1] Change in production of reactive oxygen species in human keratinocytes caused by Asian Dust

5 Normal human keratinocytes (epidermal neonatal keratinocytes) purchased from Lonza, Inc. (Walkersville, MD, USA) were subcultured in a CO₂ incubator under the condition of 37 °C and 5% CO₂ as recommended. The cell culture was incubated according to the instructions of Lonza, Inc. Specifically, the cells were cultured in 500 mL of KBM-2 medium (Clonetics CC-3103) supplemented with KGM-
10 2 Bullet kit [2 mL of bovine pituitary extract (BPE), 0.5 mL of human epidermal growth factor (hEGF), 0.5 mL of insulin, 0.5 mL of hydrocortisone, 0.5 mL of transferrin, 0.5 mL of epinephrine, 0.5 mL of GA-1000 (gentamycin sulfate + amphotericin B)].

 Untreated human keratinocytes were used as a control group. To a *Melia*
15 *azedarach* bark extract-treated test group, 10 ppm of *Melia azedarach* bark extract (acquired from the Korea Plant Extract Bank) was added. Also, 10 ppm of *Nelumbo nucifera* leaf extract (acquired from the Korea Plant Extract Bank) was added to a *Nelumbo nucifera* leaf extract-treated test group. To a *Melia azedarach* bark and *Nelumbo nucifera* leaf extracts-treated test group, 5 ppm of *Melia azedarach* bark
20 and extract and 5 ppm of *Nelumbo nucifera* leaf extract were added. 30 minutes after the pretreatment, 25 µg/mL of Asian Dust sample was added and the cells of each group were incubated for 24 hours. The Asian Dust sample was collected at the rooftop of the Amorepacific R&D Center (Yongin, Korea) during the Asian Dust season. 24 hours after the treatment with 25 µg/mL of Asian Dust, the cells were
25 washed twice with 10 mL of phosphate buffered saline (PBS) and

chemiluminescence was measured in 2 mL of PBS using a Glomax 20/20 luminometer 1 minute after adding 1 mM luminol and 20 units/mL horse-radish peroxidase (HRP). The result is shown in Fig. 1.

As seen from Fig. 1, the *Melia azedarach* bark extract and the *Nelumbo nucifera* leaf extract suppresses the production of reactive oxygen species in normal human keratinocytes increased by Asian Dust. In particular, the suppression effect is more significant when the cells are treated with both the *Melia azedarach* bark extract and the *Nelumbo nucifera* leaf extract.

[Test Example 2] Increased expression of toxin detoxification and expulsion factors and skin antimicrobial peptides as natural immune factor after treatment with *Melia azedarach* bark or *Nelumbo nucifera* leaf extract

Normal human keratinocytes (epidermal neonatal keratinocytes) purchased from Lonza, Inc. were subcultured in a CO₂ incubator under the condition of 37 °C and 5% CO₂ as recommended. The cell culture was incubated according to the instructions of Lonza, Inc. Specifically, the cells were cultured in 500 mL of KBM-2 medium (Clonetics CC-3103) supplemented with KGM-2 Bullet kit [2 mL of bovine pituitary extract (BPE), 0.5 mL of human epidermal growth factor (hEGF), 0.5 mL of insulin, 0.5 mL of hydrocortisone, 0.5 mL of transferrin, 0.5 mL of epinephrine, 0.5 mL of GA-1000 (gentamycin sulfate + amphotericin B)].

Untreated human keratinocytes were used as a control group. To a *Melia azedarach* bark extract-treated test group, 10 ppm of *Melia azedarach* bark extract (acquired from the Korea Plant Extract Bank) was added. Also, 10 ppm of *Nelumbo nucifera* leaf extract (acquired from the Korea Plant Extract Bank) was added to a *Nelumbo nucifera* leaf extract-treated test group. To a *Melia azedarach* bark and

Nelumbo nucifera leaf extracts-treated test group, 5 ppm of *Melia azedarach* bark and extract and 5 ppm of *Nelumbo nucifera* leaf extract were added. 30 minutes after the pretreatment, 25 µg/mL of Asian Dust sample was added and the cells of each group were incubated for 24 hours. 24 hours after the treatment with Asian Dust, the cells were washed twice with 10 mL of PBS and total RNA was isolated from the cells using TRIzol (Invitrogen). The isolated RNA was purified once again using an RNA kit (Qiagen) and the quality of RNA was analyzed using Bioanalyzer 2100 (Agilent). cDNA was synthesized from the isolated RNA using Superscript reverse transcriptase (RT) II kit (Invitrogen) and subjected to quantitative analysis by real-time reverse transcription polymerase chain reaction (Q-RT-PCR). The change in the expression patterns of GSTT1 (Hs00184475_m1), NQO1 (Hs01045994_m1) and hBD3 (hs00218678_m1) was evaluated using TaqMan gene expression assay kit (Applied Biosystems). The result is shown in Figs. 2-4.

As seen from Figs. 2-4, the *Melia azedarach* bark and *Nelumbo nucifera* leaf extracts increase the expression of NQO1 (Fig. 2) which detoxifies toxins, the skin antimicrobial peptide hBD3 (Fig. 3) which is a natural immune factor and GSTT1 (Fig. 4) which is an enzyme that expels toxins, in normal human keratinocytes. This effect is more significant when the *Melia azedarach* bark and *Nelumbo nucifera* leaf extracts are treated together.

Accordingly, it can be seen that a combined treatment of *Melia azedarach* bark and *Nelumbo nucifera* leaf extracts provides a synergic effect of improved detoxification and expulsion of toxins and increased expression of natural immune factors.

[Test Example 3] Increased expression of antioxidant protein genes after

treatment with *Melia azedarach* bark and *Nelumbo nucifera* leaf extracts

Normal human keratinocytes (epidermal neonatal keratinocytes) purchased from Lonza, Inc. were subcultured in a CO₂ incubator under the condition of 37 °C and 5% CO₂ as recommended. The cell culture was incubated according to the instructions of Lonza, Inc. Specifically, the cells were cultured in 500 mL of KBM-2 medium (Clonetics CC-3103) supplemented with KGM-2 Bullet kit [2 mL of bovine pituitary extract (BPE), 0.5 mL of human epidermal growth factor (hEGF), 0.5 mL of insulin, 0.5 mL of hydrocortisone, 0.5 mL of transferrin, 0.5 mL of epinephrine, 0.5 mL of GA-1000 (gentamycin sulfate + amphotericin B)].

Untreated human keratinocytes were used as a control group. To a *Melia azedarach* bark extract-treated test group, 10 ppm of *Melia azedarach* bark extract (acquired from the Korea Plant Extract Bank) was added. Also, 10 ppm of *Nelumbo nucifera* leaf extract (acquired from the Korea Plant Extract Bank) was added to a *Nelumbo nucifera* leaf extract-treated test group. To a *Melia azedarach* bark and *Nelumbo nucifera* leaf extracts-treated test group, 5 ppm of *Melia azedarach* bark and extract and 5 ppm of *Nelumbo nucifera* leaf extract were added. 24 hours after the treatment, the cells were washed twice with 10 mL of PBS and total RNA was isolated from the cells using TRIzol (Invitrogen). The isolated RNA was purified once again using an RNA kit (Qiagen) and the quality of RNA was analyzed using Bioanalyzer 2100 (Agilent). cDNA was synthesized from the isolated RNA using Superscript reverse transcriptase (RT) II kit (Invitrogen) and subjected to quantitative analysis by real-time reverse transcription polymerase chain reaction (Q-RT-PCR). The change in the expression patterns of GPx1 (Hs00829989_gH), catalase (Hs00156308_m1), Trx1 (Hs01555212_g1) and PRDX1 (Hs00602020_mH) was evaluated using TaqMan gene expression assay kit (Applied Biosystems).

When the normal human keratinocytes were treated with the *Melia azedarach* bark extract, the relative expression of GPx1, catalase, Trx1 and Prdx1 which are antioxidant enzymes that scavenge reactive oxygen species was about 2.5, 2.0, 1.9 and 1.9, respectively. When the cells were treated with the *Nelumbo nucifera* leaf extract, the relative expression was about 1.9, 1.7, 1.6 and 1.7, respectively. When the cells were treated with both the *Melia azedarach* bark and *Nelumbo nucifera* leaf extracts, the expression of GPx1, catalase, Trx1 and Prdx1 was the highest with about 3.0, 2.43, 2.48 and 2.4, respectively (Fig. 5).

Accordingly, it can be seen that the expression of the antioxidant enzymes increases when the cells are treated with the *Melia azedarach* bark extract alone and increases further when the cells are treated with both the *Melia azedarach* bark and *Nelumbo nucifera* leaf extracts.

Hereinafter, formulation examples of a composition containing *Melia azedarach* bark and *Nelumbo nucifera* leaf extracts according to the present disclosure will be described. However, the following formulation examples are for illustrative purposes only and it will be apparent to those of ordinary skill in the art that the scope of the present disclosure is not limited by the formulation examples.

[Formulation Example 1] Lotion

<i>Melia azedarach</i> bark and <i>Nelumbo nucifera</i> leaf extracts	3.00
L-Ascorbic acid 2-phosphate magnesium salt	1.00
Collagen (1% aqueous solution)	1.00
Sodium citrate	0.10
Citric acid	0.05

Licorice extract	0.20
1,3-Butylene glycol	3.00
Purified water	balance
(unit: wt%)	

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[Formulation Example 2] Cream

<i>Melia azedarach</i> bark and <i>Nelumbo nucifera</i> leaf extracts	1.00
Polyethylene glycol monostearate	2.00
Self-emulsifying glyceryl monostearate	5.00
Cetyl alcohol	4.00
Squalene	6.00
Glyceryl tri(2-ethylhexanoate)	6.00
Sphingoglycolipid	1.00
1,3-Butylene glycol	7.00
Purified water	balance

10

15

(unit: wt%)

[Formulation Example 3] Pack

<i>Melia azedarach</i> bark and <i>Nelumbo nucifera</i> leaf extracts	5.00
Polyvinyl alcohol	13.00
L-ascorbic acid 2-phosphate magnesium salt	1.00
Lauroyl hydroxyproline	1.00
Collagen (1% aqueous solution)	2.00
1,3-Butylene glycol	3.00
Ethanol	5.00

20

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Purified water	balance
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(unit: wt%)

[Formulation Example 4] Beautifying essence

5	<i>Melia azedarach</i> bark and <i>Nelumbo nucifera</i> leaf extracts	2.00
	Hydroxyethyl cellulose (2% aqueous solution)	12.00
	Xanthan gum (2% aqueous solution)	2.00
	1,3-Butylene glycol	6.00
	Concentrated glycerin	4.00
10	Sodium hyaluronate (1% aqueous solution)	5.00
	Purified water	balance

(unit: wt%)

【CLAIMS】

【Claim 1】

A composition for combating oxidation, which comprises *Melia azedarach* bark extract as an active ingredient.

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【Claim 2】

The composition according to claim 1, which further comprises *Nelumbo nucifera* leaf extract.

10 **【Claim 3】**

The composition according to claim 1 or 2, wherein the composition suppress the production of reactive oxygen species.

【Claim 4】

15 The composition according to claim 1 or 2, wherein the composition increases the expression of antioxidant protein genes.

【Claim 5】

20 The composition according to claim 4, wherein the antioxidant protein gene comprises GPx1, catalase, Trx1 and Prdx1.

【Claim 6】

A composition for alleviating the detrimental effects on skin caused by Asian Dust, which comprises *Melia azedarach* bark extract and *Nelumbo nucifera* leaf

extract as an active ingredient.

【Claim 7】

5 The composition according to claim 6, wherein the composition increases the expression of GST, NQO1 or hBD3.

【Claim 8】

10 The composition according to any one of claims 1, 2 and 6, wherein the composition alleviates the detrimental effects on normal keratinocytes in skin.

【Claim 9】

The composition according to any one of claims 1, 2 and 6, which comprises 1-30 wt% of the extracts based on the total weight of the composition.