Office de la Propriété Intellectuelle du Canada
Un organisme d'Industrie Canada

Canadian Intellectual Property Office
An agency of Industry Canada

CA 2385301 A1 2001/06/21

(21) 2 385 301

(12) DEMANDE DE BREVET CANADIEN
CANADIAN PATENT APPLICATION

(13) A1

(86) Date de dépôt PCT/PCT Filing Date: 2000/11/29
(87) Date publication PCT/PCT Publication Date: 2001/06/21
(85) Entrée phase nationale/National Entry: 2002/03/18
(86) N° demande PCT/PCT Application No.: JP 2000/008436
(87) N° publication PCT/PCT Publication No.: 2001/043716
(30) Priorité/Priority: 1999/12/14 (11/354031) JP

(51) Cl.Int.$^{7}$/Int.Cl.$^{7}$ A61K 7/48, A61K 31/197, A61P 17/00, A61K 7/00

(71) Demandeur/Applicant:
COSMO OIL CO., LTD., JP

(72) Inventeurs/Inventors:
ITOH, YOSHIYASU, JP;
TANAKA, TOHRU, JP

(74) Agent: RICHES, MCKENZIE & HERBERT LLP

(54) Titre : COMPOSITIONS DE PEELING
(54) Title: COMPOSITIONS FOR PEELING

(57) Abrégé/Abstract:
Compositions for peeling which contain a compound selected from among 5-aminolevulinic acid, its derivatives and salts thereof; and a method of improving the human or animal skin which involves the step of administering such a compound to the human or animal skin and the step of peeling the skin.

## ABSTRACT

A composition for peeling, comprising a compound selected from the group consisting of 5-aminolevulinic acid, derivatives thereof and salts thereof; and a method for improving the skin of human or animal, comprising: administering the compound selected from the group consisting of 5-aminolevulinic acid, derivatives thereof and salts thereof to the skin of human or animal, and carrying out peeling of the skin.

# DESCRIPTION

# PEELING COMPOSITION

## TECHNICAL FIELD

The present invention relates to a composition for peeling. More particularly, it relates to a composition for carrying out peeling by applying a chemical to the skin followed by irradiation with light.

## BACKGROUND ART

Peeling is a method for promoting the regeneration of new skin by peeling off old skin. It has been widely used in the field of cosmetic surgery in order to aesthetically improve the skin which is in bad conditions but do not necessarily require a medical treatment. Peeling is roughly classified into chemical peeling and laser peeling.

In the chemical peeling, chemicals such as phenol, glycolic acid and trichloroacetic acid are applied to the skin, and the skin is peeled off. It is further classified into superficial peeling in which the skin is peeled off superficially and deep peeling in which the skin is peeled off deeply. Since the operation is included in chemical burn, inflammation arises after the completion of the operation. When the operation is carried out only on the

skin surface layer to minimize the inflammation, only insufficient effects can be achieved. On the other hand, a strong operation frequently induces long-lasting pigmentation and rubefaction or even scar in some cases. The deep peeling is a highly invasive treatment which should be carried out under anesthesia and there arises postoperative pain after the operation.

The laser peeling is a therapeutic method wherein the skin surface is burnt by irradiating with laser beams instead of the application of chemicals. The operation should be carried out under anesthesia. Similar to the chemical peeling, the laser peeling is accompanied by inflammation. Since the operation frequently induces long-lasting pigmentation, rubefaction and scar formation particularly in case of Orientals, it should be extremely carefully applied to Orientals at the present stage.

As described above, the existing peeling methods may be associated with complications. When they are carried out while avoiding the complications, only insufficient effects can be obtained. Therefore, it has been required to establish an effective peeling method which imposes little burden on patients.

## DISCLOSURE OF THE INVENTION

An object of the present invention is to provide an effective peeling method which imposes little burden on patients.

The present invention relates to a composition for peeling, comprising a compound selected from the group consisting of 5-aminolevulinic acid, derivatives thereof, and salts thereof.

Also, the present invention relates to a method for improving the skin of human or animal skin, comprising: administering a compound selected from the group consisting of 5-aminolevulinic acid, derivatives thereof, and salts thereof to the human or animal skin; and carrying out peeling of the skin.

## BEST MODE FOR CARRYING OUT THE INVENTION

It is known that 5-aminolevulinic acid (hereinafter sometimes referred to as "5-ALA"), derivatives thereof and salts thereof used in the composition for peeling according to the present invention are useful as plant growth promoters, herbicides, insecticides, sensitizers in photodynamic therapy for cancer and the like (Japanese Patent No. 2613136, Japanese Patent No. 2896963, JP-W-61-502814, JP-A-2-138201). However, it has not been known that 5-ALAs are applicable to peeling.

The 5-ALA derivatives include 5-aminolevulinic acid esters, *N*-acyl-5-aminolevulinic acids, and *N*-acyl-5-aminolevulinic acid esters (hereinafter referred to as "5-ALA esters", "*N*-acyl-5-ALAs", and "*N*-acyl-5-ALA esters", respectively).

Examples of the 5-ALA esters include esters of 5-ALA with an alkyl group which may be substituted and has a linear, branched or cyclic structure and 1 to 24 carbon atoms. Examples of the substituent of the substituted alkyl group include a hydroxyl group, an alkoxy group, a phenyl group and the like. Preferred examples of the alkyl group which may be substituted include a methyl group, an ethyl group, an n-propyl group, an isopropyl group, an n-pentyl group, an n-hexyl group, a cyclohexyl group, an n-heptyl group, an n-octyl group, an n-nonyl group, an n-dodecyl group, an n-hexadecyl gorup, a benzyl group, a phenethyl group, a 3-phenylpropyl group, a hydroxyethyl gorup, an ethoxyethyl group and the like.

The *N*-acyl-5-ALAs include compounds wherein the amino group of 5-ALA has been acylated by an acyl group having 1 to 24 carbon atoms, such as an alkanoyl group, an aromatic acyl group, a benzyloxycarbonyl group and the like. Preferred examples of the acyl group include an acetyl group, an n-propanoyl group, an n-butanoyl group, an n-pentanoyl group, an n-hexanoyl group, an n-nonanoyl group, a benzyloxycarbonyl group and the like.

Examples of the *N*-acyl-5-ALA esters include compounds having the same ester and acyl group as described above. Preferable examples include combinations of a methyl ester group and a formyl group, a methyl ester group and an acetyl group, a methyl ester group and an n-propanoyl group, a methyl ester group and an n-butanoyl group, an ethyl ester group and a formyl group, an ethyl ester group and an acetyl group, an ethyl ester group and an n-propanoyl group, an ethyl ester group and an n-butanoyl group and the like.

Examples of the salts of 5-ALA or derivatives thereof include acid addition salts, such as hydrochloride, hydrobromide, hydroiodide, phosphate, nitrate, sulfate, acetate, propionate, toluenesulfonate, succinate, oxalate, lactate, tartarate, glycolate, methanesulfonate, butyrate, valerate, citrate, fumarate, maleate, malate and the like; metal salts, such as sodium salt, potassium salt, calcium salt and the like; ammonium salts; alkylammonium salts; and the like. These salts may be used as an aqueous solution or a powder and have the same effect as 5-ALA.

The 5-ALA, derivatives thereof or salts thereof as described above may form a hydrate or a solvate. They may be used alone or in combination of two or more.

The 5-ALA or its salts can be obtained by any method of chemical synthesis, microbial production and enzymatic production (for example, see WO98/54297, USP

5,380,935). The 5-ALA derivatives or salts thereof can be produced by a publicly known chemical synthesis method described in, for example, JP-A-4-9360. Products obtained by microbial or enzymatic methods and crude products obtained by chemical synthesis may be used as such without separating or purifying any more, so long as they are free from any contaminant harmful to humans or animals.

The composition for peeling according to the present invention may contain an additional component, so long as it contains the 5-ALA, derivatives thereof or salts thereof. The dosage form is not particularly limited, so long as it is an external preparation for skin. Examples include a dust, a solution, an ointment (including an oily ointment, an emulsified ointment, an water-soluble ointment and hydrogel), a dermatologic paste and the like. Examples of the additional component include a percutaneous absorption accelerator such as isopropyl myristate, a surfactant, alcohol, polyhydric alcohol and the like.

When an aqueous solution or an ointment is prepared, it is necessary to prepare it so as not to become alkaline in order to prevent 5-ALA, derivatives thereof or salts thereof from decomposition. The aqueous solution and the ointment are preferably prepared by adjusting pH 8 or lower, more preferably pH 7 or lower, and particularly preferably pH 6.1 or lower. When they become alkaline, decomposition can be prevented by eliminating oxygen. Taking this point

into consideration, they can be used together with a base component generally used for the solution and the ointment.

When peeling is carried out using the composition according to the present invention, the composition of the present invention is applied to the target skin, and the skin is exposed with light. Preferably, the skin is irradiated with light (i.e., photodynamic peeling). The term "photodynamic peeling" as used in the present invention means a peeling method in which a chemical is applied to the skin, followed by irradiation with light. The method for applying the composition of the present invention to the skin is not particularly limited, so long as the active ingredient can be absorbed via the skin. Examples include spraying, coating, packing, iontophoresis and the like. The dose to the skin of 5-ALA, derivatives thereof or salts thereof which are the active ingredient is generally from 10 mg to 10g, preferably from 100 mg to 5 g and more preferably from 1 g to 5 g in terms of 5-ALA hydrochloride per 100 $cm^2$ of the skin. From the viewpoint of efficiency, after the application of the composition according to the present invention till the light irradiation, the applied skin is preferably shaded.

The skin which is applied with the composition according to the present invention is not particularly limited. But, the composition is preferably applied to the skin which is to be improved from an esthetic viewpoint,

for example, skin having old horny substances or epiderm, skin with spots, freckles or acne, skin suffering from deterioration in tension or gloss, wrinkled skin and skin after suffering from comedo.

Next, the skin is irradiated with light. From the viewpoint of the percutaneous absorption and metabolism of the active ingredient, it is preferable to irradiate the skin with light for 2 to 48 hours, preferably for 4 to 24 hours, after the application of the composition according to the present invention. It is also preferable to perform the light-irradiation after removing off the composition according to the present invention remaining on the skin.

As the light for the irradiation, it is preferable to employ visible beams around 635 nm or laser beams of about 635 nm. The irradiation intensity is preferably from 1 to 100 $J/cm^2$, particularly from 3 to 10 $J/cm^2$. The irradiation time is preferably from 1 to 50 minutes, particularly from 10 to 30 minutes.

After the treatment in accordance with the present invention, the treated part is rubefied and swells up for about 3 days. Subsequently, the skin surface peels off spontaneously. As a result, the peeling effects (for example, fall-off of old horny substances or epiderm, removal of spots, freckles or acne, restoration of skin tension or gloss, relief of wrinkles, amelioration of

comedo) can be achieved within 3 or 4 days to 3 weeks, though the effects are different among individuals.

Although sufficient effects can be obtained by carrying out the treatment in accordance with the present invention once, it may be repeated plural times so as to further enhance the effects.

Although the mechanism of the photodynamic peeling according to the present invention still remains unknown, it is estimated as proceeding as follows. Death of cells and tissues is classified into two types, i.e., necrosis and apoptosis. The occurrence of necrosis, in which cell injury and death are caused by external factors such as chemicals or heat, induces inflammatory reactions such as vasodilation and migration of inflammatory cells toward the affected part. Subsequently, melanogenesis is accelerated in pigment cells. On the other hand, apoptosis means another form of cell death in which cells, so to speak, "suicide" because of intracellular factors. It is known that the cell death of this type is accompanied by no or little inflammation.

Chemical peeling and laser peeling are techniques based on the necrosis mechanism whereby epidermal cells are killed by chemicals or heat. Therefore, these treatments frequently induce troubles such as inflammation, pigmentation or scar formation which prolong over 3 month, 6 month or even 1 year or longer in some cases.

In contrast thereto, it is considered that epidermal cells are killed based on the apoptosis-like mechanism in photodynamic peeling. After externally administered, 5-ALA is absorbed in epidermal cells and then enters into the heme biosynthesis pathway in these cells. Next, it is converted into photosensitive substances such as protoporphyrin IX and accumulated in the cells. When exposed to exciting light, these photosensitive substances such as protoporphyrin IX form active oxygen and, in its turn, the cells "suicide". When 5-ALA salts or derivatives thereof are administered, 5-ALA would be induced therefrom in the body and the induced 5-ALA would trigger the same mechanism.

As described above, it is considered that the photodynamic peeling according to the present invention is based on the cell death by the apoptosis-like mechanism and, therefore, induces little troubles such as inflammation, pigmentation or rubefaction.

The present invention is explained in detail based on Examples. But they are described for only illustration, and the present invention is not limited thereto. Also, the following Examples were carried out under full informed consent on the basis of medical diagnosis.

Example 1

5-ALA hydrochloride was added to a hydrophilic ointment in accordance with the Japanese Pharmacopoeia (manufactured by Yoshida Pharmaceutical Co., Ltd.) to give a concentration of 20% by weight and mixed thoroughly.

Under informed consent, 10 g of the obtained ointment was applied to the left half of the face of a 22 years old female. Then the treated part was covered with Saran Wrap (manufactured by Asahi Kasei Corporation, the same applies hereinafter) and further with aluminum foil for blocking off light.

After 4 hours, the treated part was cleansed and irradiated with an excimer dye laser (PDTEDL1 manufactured by Hamamatsu Photonics K.K.) at a wavelength of 635 nm at an intensity of 5 $J/cm^2$ for 30 minutes.

After the irradiation, edematous erythema was formed at the affected part. But, it gradually disappeared from the day 3. At the same time, a thin scab was formed on the skin surface. The scab, i.e., old epidermal components, gradually peeled off in bathing and face cleansing, and peeling was completed 3 weeks after the operation.

A comparison between the left and right parts of the face indicated that skin tension, wrinkles, pigmentation, acne spots and the like had been obviously relieved. In a follow-up survey carried out 6 months after

the treatment, the treated part still remained in favorable conditions.

Example 2

Under informed consent, 5 g of an ointment prepared in the same manner as in Example 1 was applied to the right cheek (6 cm × 6 cm) of a 24 years old female. Then, the treated part was covered with Saran Wrap and further with aluminum foil for blocking off light.

After 12 hours, the treated part was cleansed and irradiated with an excimer dye laser (PDTEDL1 manufactured by Hamamatsu Photonics) at a wavelength of 635 nm at an intensity of 5 J/cm$^2$ for 15 minutes.

Although edematous erythema was formed at the affected part after the irradiation, it gradually disappeared from the day 3. At the same time, old epidermal components gradually peeled off in bathing and face cleansing, and peeling was completed on the day 6 after the operation.

A comparison with the corresponding part in the left cheek indicated that skin tension, wrinkles, pigmentation, acne spots and the like had been obviously relieved. In a follow-up survey carried out 6 months after the treatment, the treated part still remained in favorable conditions.

## INDUSTRIAL APPLICABILITY

An effective peeling treatment which imposes little burden on patients can be attained by using the composition for peeling of the present invention.

# CLAIMS

1. A composition for peeling, comprising a compound selected from the group consisting of 5-aminolevulinic acid, derivatives thereof and salts thereof.

2. The composition according to claim 1, which is a composition for photodynamic peeling.

3. The composition according to claim 1, wherein the 5-aminolevulinic acid derivatives are 5-aminolevulinic acid esters, *N*-acyl-5-aminolevulinic acids, or *N*-acyl-5-aminolevulinic acid esters.

4. The composition according to claim 1, which further comprises a percutaneous absorption accelerator.

5. A method for improving the skin of human or animal, comprising:

administering a compound selected from the group consisting of 5-aminolevulinic acid, derivatives thereof and salts thereof to the skin of human or animal; and

carrying out peeling of the skin.

6. The method according to claim 5, wherein said peeling is carried out by photodynamic peeling.

7. The method according to claim 6, wherein the 5-aminolevulinic acid derivatives are 5-aminolevulinic acid esters, *N*-acyl-5-aminolevulinic acids, or *N*-acyl-5-aminolevulinic acid esters.