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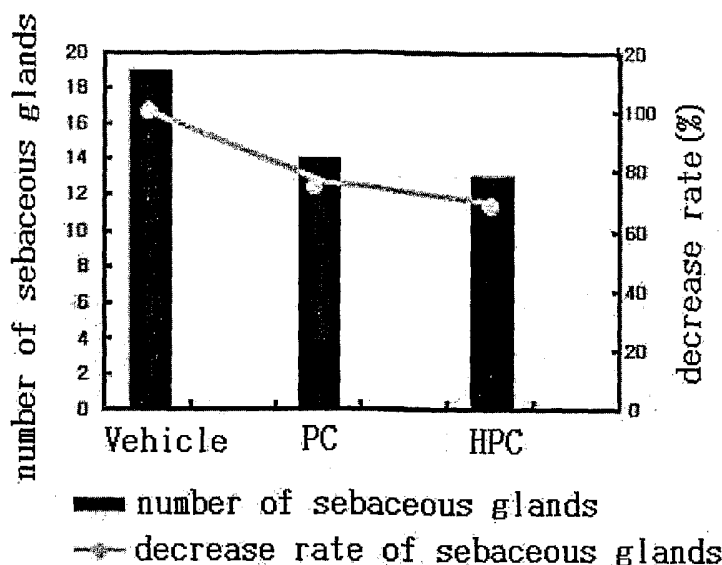
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(54) Title: COMPOSITION FOR INHIBITING DIFFERENTIATION OR GROWTH OF FAT CELLS

variation of the number of sebaceous glands



(57) Abstract: The present invention relates to a composition for inhibiting differentiation or growth of fat cells including sebocytes, which contains phosphatidylcholine (PC) and/or hydrogenated phosphatidylcholine (HPC) as an active ingredient. The composition is effective in treatment or prevention of fat cell-related diseases, particularly obesity or acne through inhibiting of differentiation or growth of fat cells such as sebocytes. Further, the composition can be used in combination with other compositions or other active ingredients in treatment or prevention of many fat cell-related diseases.



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COMPOSITION FOR INHIBITING DIFFERENTIATION OR GROWTH OF FAT CELLS

TECHNICAL FIELD

5 The present invention relates to a composition for inhibiting differentiation or growth of fat cells. On the other side, the invention relates to a composition for inhibiting sebum secretion. More specifically, the invention contains phosphatidylcholine or hydrogenated phosphatidylcholine as an active ingredient.

10 BACKGROUND ART

Lipid is one of main ingredients constituting a human body and used as an important energy source in the human body since it is oxidized mainly in the liver and thus generates many ATPs (adenosine triphosphates). The lipid is a main ingredient of the biomembrane and also a precursor material of steroidal hormone
15 (for example, sex hormone). Recently, the lipid is taken many interests as intermediaters in a signal transduction system of a living body. The lipid includes fatty acid, acylglycerol, phospholipid, sphingolipid, steroids, etc.

The lipid occupies 10% or more of an adult's weight on an average and about 30~40% of calories ingested by food. Many kinds of lipids are present in the
20 human body. Phospholipid is phosphorylated to third glycerol carbon of diacylglycerol and thus becomes a phosphatidic acid, which is a kind of main lipid-constituting cell membrane. The phospholipid exhibits a hydrophilic property by hydroxy group of a phosphorous part (head) and a hydrophobic property by a remaining part (tail). Accordingly, when the phospholipid is put in water, the
25 hydrophobic tails come together into an interior of the phospholipid lump and the

hydrophilic heads are arranged on an exterior of the lump, thereby causing a bilayer of phospholipid to be formed. Due to such a characteristic, phosphatidic acid becomes a main ingredient of the cell membrane. Other phospholipids, which have such a characteristic of dual-properties molecule and constitute the cell membrane, include cephalin (=phosphatidyl ethanolamine) and lecithin (=phosphatidyl choline), etc.

Fat ingredient ingested from the external is used as an ingredient constituting the human body or an energy source and remained fat ingredient is accumulated in fat cells. If a problem occurs in a fat metabolism, various variations occur in the human body and the extreme case is that the life may be lost.

The number and sizes of the fat cells significantly influence on an outward shape of the human body. The number of the fat cells is increased as growing up. When segmentation and proliferation of the fat cells are finished, about thirty billion of fat cells are present in the human body. In the times of adolescence, the fat cells are most flourishingly increased. Beyond the adolescence, the number of the fat cells is not increased and only the size thereof is increased. The energy left in the human body is accumulated in the fat cells as a fat form and thus the size of the fat cells is grown.

It is called obesity that fat is not effectively used, accumulated and thus increased beyond a normal range in the human body. The obesity generally occurs due to excessive intake of fat, except for some minor exceptions. That is, when intake energy is larger than consumption energy, a great quantity of neutral fat is accumulated in adipose tissues and thus the weight exceed a normal weight by 20% or more. Since the obesity is a cause of many diseases including adult disease, it should be attentive to the obesity.

The obesity can be classified into a fat cell-hypertrophic type and a fat cell-vegetative type. The hypertrophic type is that a size of fat cell is increased. It occupies most of adult obesity. When obesity occurs at an early age, the obesity becomes a fat cell-vegetative type that more fat cells are generated. That is, when
5 the number of fat cells is increased beyond a normal state in a year after one's birth, in the times of adolescence or during the period of pregnancy or an amount of fat is increased due to excessive intake of calories in 2~10 years in which the amount of fat in fat cells is not varied, the fat cell-vegetative type obesity frequently occurs.

The lipid ingredient that occupies most of membrane ingredients
10 constituting the human body is much present particularly in the skin and reveals various physiological functions as well as structural functions. Also, it performs some essential functions of vital phenomenon, such as to maintain homeostasis of the human body and to protect the body from an exterior environment.

The skin comprises epidermis, dermis, subcutaneous tissue and skin
15 appendage that the skin is varied.

Millions of cells, blood vessels and nerves are present in a form of complicated network per inch of the skin. In addition, the pores of hair follicles, sebaceous glands and sweat glands are present in the skin. Most of skin problems occur in the epidermis, which is a surface layer of the skin, but inherent problems
20 starts from the dermal layer. The dermis consists of collagen (a kind of protein) fiber. The collagen fiber makes the skin strong and maintains a shape of the skin and gives flexibility to the skin. The blood vessels, fat cells, sebaceous glands and sweat glands are wrapped by the collagen fiber. There are 95~100 sebaceous glands per cm² of the skin on the average. The sebaceous glands secrete sebum and thus
25 protect the skin.

Small bags (vesicles) are present in the sebaceous glands and conduits of the sebaceous glands are connected to the middle of the hair follicle. The sebaceous gland secretes sebum, thereby making the skin smooth and protecting moistures of the skin from evaporating. The sebaceous glands are present in all regions of the human body, particularly in the face, except for hands and foots. The sebum is a semi-liquid, oily ingredient and produced from the sebaceous gland. The sebum generally flows to an entry of the hair follicle along the conduit of the sebaceous gland.

When the sebum becomes to be hard and the hair follicle is blocked, a black head is generated.

The sebaceous gland is not controlled by the nerve and the secretion thereof is regulated by stimulation of male sex hormone (androgen). The male sex hormone is secreted in a small quantity from adrenal cortex of a man and a woman, stimulates the sebaceous gland or a development of the hair follicle and thus makes a horny layer be thickened. A secretion state of sebum is varied depending on the years. The sebum is flourishingly secreted during one year after one's birth, after which the function of secretion is degenerated. When it is 8~10 years old, the function of secretion is again matured. From 11~13 years old, the function is more developed. The function is most developed about 20~25 years old. After that, the function of secretion is almost not changed, and gradually decreased in old age. The sebum secretion is also varied according to seasons, ages, hormones and environments and increased or decreased depending on temperatures, thickness of sebaceous membrane and perspiration. About 1~2 g of the sebum is secreted on the average per day. The sebum is again produced in 3~4 hours after bathing, 1~2 hour(s) after shampooing and one hour after washing.

The sebum forms sebum film on the horny layer and a surface of hair and thus prevents moistures from evaporating, thereby maintaining humidification of the skin and hair and simultaneously giving gloss them. The sebum contains fatty acid. It is known that the fatty acid has functions of sterilizing pyogenic bacteria and trichophyton and controlling skin microorganism.

When the sebum is secreted on the skin surface, it is mixed with sweat and thus forms the sebum film like natural cream, since the sebum and sweat are mixed and thus forms an emulsion state. The emulsification of the sebum is very complicated. When the sebum and sweat are present in an appropriate amount, w/o type-emulsion state is formed. When moistures are increased due to perspiration, the state is changed to o/w type. When it comes back, the state becomes w/o type again. That is, the sebum film is changed to a w/o type or o/w type according to proportions of fat and moisture in the skin surface.

In addition, it is known that the sebum has functions of controlling percutaneous absorption and generating vitamin D, etc.

When the sebum having such functions is more secreted than normal state, acnes or dandruffs are formed and the skin type becomes oily. When the sebum is secreted too less, moistures are easy to evaporate. Thus, there is no gloss and the skin type becomes dry. In addition, the fatty acid having a sterilizing function is decreased and it is apt to be attacked by pyogenic bacteria and trichophyton.

The sebum serves to protect the skin. However, the sebaceous glands are excessively developed, the sebum secretion is increased and acnes may be generated. Accordingly, regarding the prevention and treating of the acnes, it is effective to inhibit the development of the sebaceous glands and thus to regulate the sebum secretion.

It is not well known of materials for treating lipid-related diseases such as obesity or acnes by inhibiting differentiation or growth of fat cells. In particular, with regard to the treating of acnes, it is almost not known of medicaments controlling the growth and differentiation of fat cells and thus treating the acnes.

5 Most of prior acne cures are antibiotics alleviating inflammation of the inflammatory acnes or killing acne bacilli or ones treating the acnes by perforating the blocked skin pores. That is, since most of cures can only alleviate the acnes through an antibacterial action and anti-inflammatory action, they cannot be a fundamental remedy for acnes.

10 The inflammatory acnes exacerbated by androgen (male sex hormone) and proliferating of acne bacilli is generally treated with antibiotics killing the acne bacilli and medicines perforating the blocked skin pore. There are various cures of local treatment that are applied on the acne region. Action mechanism thereof is different, respectively. For example, benzoyl peroxide has strong antibacterial and
15 anti-inflammatory actions but shows skin irritant property and cannot control the sebum secretion. There are various antibacterial action cures including erythromycin, tetracycline and clindamycin, etc. which inhibit propagation of bacilli. However, the medical treatments up to dates are effective when the acnes are not serious, but do not have a large influence on inpatients when the acnes are
20 serious or frequently recur. It is recently reported that the antibacterial action cures are inefficacious for acnes, which does not induce inflammation. Thus, these cures are believed to inhibit the inflammatory response caused by bacilli present, rather than to inhibit the propagation of bacilli itself. Accordingly, long-term uses thereof are limited. In addition, retinoid acid, which is used as a cure for inhibiting
25 abnormal hyperkeratosis, effectively reduces generated comedo, but excessively

stimulates the skin. Also, the retinoid acid does not have an effect of regulating the sebum secretion. Accordingly, up to dates, it is difficult to treat a cause of the acnes.

There is a systematic therapy such as an oral or injection administration as well as local treatment. However, they cause many adverse reactions. In addition, it is not easy to use the therapy, since it is often forbidden to medicate them to pregnant women or children under 12 years old.

In order to fundamentally treat the acnes, it is required to inhibit differentiation and growth of sebaceous cells, which is a kind of fat cells, and thus to reduce an excessive sebum secretion. However, the prior arts could not achieve such objects.

Meanwhile, in order to treat diseases such as obesity due to the hypertrophy of fat cells or the excessive proliferation of fat cells, it can be a fundamental therapy to inhibit differentiation and growth of fat cells. However, with regard to it, the prior arts could not suggest an effective means.

DISCLOSURE OF THE INVENTION

Accordingly, the present invention has been made to solve the above-mentioned problems occurring in the prior art. The object of the present invention is to provide composition capable of fundamentally treating or alleviating lipid-related diseases such as obesity or acnes. Specifically, the object of the invention is that obesity due to the hypertrophy of fat cells or the excessive proliferation of fat cells is fundamentally treated with inhibiting differentiation and growth of fat cells such as sebaceous cells and the acnes are fundamentally treated with inhibiting the sebum secretion.

In order to accomplish the above objects, there is provided a composition

for inhibiting differentiation or growth of fat cells comprising one or two selected from the group consisting of phosphatidylcholine and hydrogenated phosphatidylcholine as an active ingredient.

Preferably, the above fat cells may be sebaceous cells.

5 The composition according to the invention may be used for treating a disease caused by at least one of abnormal lipid metabolism, hypertrophy of fat cells, over-activation and abnormal proliferation of fat cells.

According to the invention, the above disease may be obesity or acne.

According to another embodiment of the invention, there is provided a
10 composition for inhibiting sebum secretion comprising one or two selected from the group consisting of phosphatidylcholine and hydrogenated phosphatidylcholine as an active ingredient.

According to an embodiment of the invention, there is provided a composition for treating or inhibiting obesity comprising one or two selected from
15 the group consisting of phosphatidylcholine and hydrogenated phosphatidylcholine as an active ingredient.

In the composition according to the invention, the above phosphatidylcholine may be derived from an egg or a soybean.

In the composition according to the invention, the phosphatidylcholine or
20 the hydrogenated phosphatidylcholine may be contained in an amount of 0.005~90 wt.% of the total composition.

In the composition according to the invention, the phosphatidylcholine or the hydrogenated phosphatidylcholine may be contained solely or in combination with other active ingredients.

25 According to an embodiment of the invention, there is provided a kit for

treating obesity comprising the above composition for inhibiting differentiation or growth of fat cells.

According to an embodiment of the invention, there is provided a kit for treating acnes comprising the above composition for inhibiting differentiation or growth of sebaceous cells or the composition for inhibiting the sebum secretion.

The above composition for inhibiting differentiation or growth of sebaceous cells or composition for inhibiting the sebum secretion is contained together with other compositions.

BRIEF DESCRIPTION OF THE DRAWINGS

The above and other objects, features and advantages of the present invention will be more apparent from the following detailed description taken in conjunction with the accompanying drawings, in which:

FIG. 1 is a photograph comparing the results of the treatment of phosphatidylcholine (PC) and hydrogenated phosphatidylcholine (HPC) on the backs of fuzzy rats with the result of the control group applied;

FIG. 2 shows a thin-layer chromatography analysis result of comparing biopsied tissues of the back of fuzzy rat applied with hydrogenated phosphatidylcholine with the control group;

FIGs. 3a and 3b are graphs quantitatively showing the result of Fig. 2;

FIG. 4 is a photograph comparing the result of applying cream formulations according to an embodiment of the invention to the backs of fuzzy rats with the control group;

FIG. 5 is a photograph showing the result of applying the composition of

liposome formulations according to an embodiment of the invention to the backs of fuzzy rats.

FIGs. 6a to 6c are photographs showing the back tissue of fuzzy rat, which is treated as Example 3, stained by H&E staining method, FIGs. 6d to 6f are photographs showing the back tissue of fuzzy rat, which is treated as Example 3, stained by fat staining method and FIG. 6g is a graph numerically showing the results of FIGs. 6a to 6f;

FIG. 7 is a graph comparing the number of activated sebaceous cells present in the back tissue of the fuzzy rat treated as Example 3 with the control group;

FIG. 8 is a photograph showing an effect of phosphatidylcholine and hydrogenated phosphatidylcholine on the gene expression of PPAR γ ;

FIG. 9 is a photograph comparing the states before and after differentiating into adipocytes;

FIG. 10 is a photograph showing variations of expression amounts of PPAR γ by PC and HPC treatments; and

FIG. 11a shows a variation of total amount of lipid by HPC treatment and FIG. 11b shows a variation of an amount of sebum in the human body by HPC treatment.

BEST MODE FOR CARRYING OUT THE INVENTION

Hereinafter, preferred embodiments of the present invention will be described with reference to the accompanying drawings. In the following description of the present invention, a detailed description of known functions and configurations incorporated herein will be omitted when it may make the subject

matter of the present invention rather unclear.

In the description herein, "treatment" is interpreted to have the broadest meaning. That is, it comprises disease prevention as well as alleviation or relief of the symptoms of disease. For example, it comprises acne prevention by inhibiting sebum secretion of a person having the oily skin and thus maintaining lipid balance. In addition, it comprises maintenance of not further making worse the disease.

In the present invention, "hydrogenated phosphatidylcholine" is phosphatidylcholine of which all unsaturated fatty acid is changed to saturated fatty acid.

In the present invention, "differentiation of fat cells" is a process that adipose tissue grows from preadipocyte to fat cell. That is, it is a process of differentiation from mesenchymal stem cell into mature fat cell via the preadipocyte. In this differentiation process, lipid-related gene expression is required, wherein PPAR- γ and CCAAT/enhancer-binding proteins (C/EBP) are engaged. Methods for confirming whether or not the differentiation of fat cells is are experimental approach by staining and a biochemistry approach. The confirmation can be done by comparing degrees of staining of Oil-red-O, in the staining method (Fig. 9). The left and right photographs of Fig. 9 show preadipocytes before differentiation and mature fat cells after differentiation, respectively. In addition, with specific genes expressed in differentiation into fat cells, it can confirm whether or not the differentiation occurred. For example, the genes include adipocyte fatty acid binding protein, PPAR γ and adipisin.

In the present invention, "growth of fat cells" is meant that the number of fat cells is increased during a period of fat cell increase such as the early days of birth and adolescence after the fat cells are differentiated, that sizes of fat cells

formed once are increased and that inactivated fat cells are activated. There is a hypothesis that an ability of increasing the number of fat cells is reduced before grown-up and then an increase of an amount of adipose tissue is mainly dependent on the increase of the sizes of the fat cells. However, the hypothesis of a particular
5 period in which fat cells are proliferated is partially true. It can be seen that both the number and sizes of fat cells are increased in some obesity started from an adult age. As such, it is possible to adjust the bioenergy balance by differentiating the fat cells and controlling the number and sizes of fat cells generated. If there is an abnormality in such mechanism, the disease occurs. Type II diabetes that exhibits
10 insulin resistance occurs mainly due to obesity. With regard to this, the material called as thiazolidinediones (TZD) is used as a cure. The TZD is known as a substrate for PPAR γ receptor, but reduces the insulin resistance in type II diabetes and more reacts on the insulin. However, when considering the substrate for PPAR γ receptor, an adverse reaction of the TZD can be expected that accelerates the
15 differentiation into the fat cells. Accordingly, material that enhances the insulin reaction such as the TZD but acts as an antagonist substrate for the PPAR γ receptor may be used as a good cure for the diabetes.

According to the invention, a content of phosphatidylcholine or hydrogenated phosphatidylcholine is preferably 0.005~90 wt.% of the total
20 composition. If the content is below 0.005 wt.%, the effect thereof is negligible, and if the content is above 90 wt.%, there occurs a problem in a stability of formulation.

The acne is an inflammatory disease of hair follicle happening to only the human being. It usually happens in the times of adolescence and disappears in the
25 mid-twenties. However, it happens even in the twenties or thirties. Most of the

acnes occur on the forehead and a region of the nose in which sebum is flourishingly secreted and a region of the jaws on which the hands are frequently touched. Sometimes, the acne occurs even on the chest and back, wherein it may remain for a long time even after the acnes on the face are disappeared. The acne
5 includes typical acne vulgaris, acne conglobata, premenstrual acne and preadolescent acne, etc. A main attack region and symptoms of the respective acnes are different according to the kinds and causes thereof. A treatment method is also different depending on the kinds of the acnes.

The cause or mechanism of acne generation is not clearly clarified, but it is
10 reported that various factors such as a male sex hormone action, a genetic cause, food, menses and stress are concerned with it and the acnes occurs through reciprocal reaction among these factors. In particularly, it is believed that excessive sebum secretion, keratinization of the pores, an abnormal multiplication of microorganism in a pilosebaceous unit and an inflammatory response is major
15 causes of the acne.

When men and women reach their adolescence, androgen, which is a male sex hormone, is flourishingly secreted and stimulates the sebaceous gland adjacent to the hair follicle. As a result of that, excessive differentiation and growth of the sebaceous gland occur and thus sebum is over-produced. In addition, it is made a
20 condition that *Propionibacterium acnes*, which are bacilli present in the hair follicle, are easy to propagate by itself. Accordingly, steatolytic enzyme secreted by the bacilli decomposes triacylglyceride of an interior of sebum, thereby producing free fatty acid. The free fatty acid stimulates horny cells of an entry of the pores and promotes differentiation of the horny cells, so that the horny layer is thickened and
25 thus hyperkeratosis occurs. The entry of the pores of the horny layer having

increased binding force is narrowed or plugged by the excessive sebum secretion and the thickened horny cells, so that comedo is formed. As a result of that, the wall of hair follicle is thinned due to the comedo formed by the continuously secreted sebum and the hyperkeratosis, and immunocytes that senses proliferation
5 of anaerobes as proliferation of anaerobes is increased are activated, so that an inflammatory response is caused and thus inflammatory acnes are produced.

As mentioned above, a first step of acne generation mechanism is over-differentiation and growth of the sebaceous glands by the androgen. Accordingly, in order to fundamentally treat the acnes, it is required to inhibit the over-
10 differentiation and growth of the sebaceous glands.

It is recently reported that phospholipids containing a large quantity of linoleic acid extracted from the soybean can be used as a cure for the acnes. Fifteen years ago, Downing suggested a hypothesis as follows: a proportion of fatty acids in the skin of the acne patient is different from that of the normal person and when
15 the content of linoleic acid is insufficient, if the unbalance is made up for through the supply of linoleic acid from an exterior, it is possible to reduce the generation of the acnes. According to a recent investigation, it is observed that when a diet deficient in the linoleic acid is done, the acne is generated. In addition, since the linoleic acid has an effect of inhibiting a synthesis of excessively secreted sebum, it
20 can be used for a fundamental treatment of the acnes.

Contrary to the existing method of treating the acnes by inhibiting antibacterial action or hyperkeratosis, the phospholipids, which basically controls a synthesis of excessively secreted lipids and has an effect of making up for unbalanced composition of fatty acid due to generation of free fatty acid isolated by
25 *P. acnes*, can be used for preventing and inhibiting the acnes.

The phospholipids include phosphatidylcholine (PC), phosphatidylethanolamine (PE), phosphatidylserine (PS) and sphingomyelin (SM) and occupy a half or more of all lipids of cell membrane. These phospholipids are unequally and dissymmetrically present in an inner layer and an outer layer of the cell membrane. An outer layer of plasma membrane mainly consists of phosphatidylcholine and sphingomyelin and an inner layer thereof consists of phosphatidylethanolamine and phosphatidylserine. Phosphatidylinositol (PI), which is the fifth most present phospholipid, is also present in the inner layer of the cell membrane. Although the phosphatidylinositol occupies small portions as an ingredient of the cell membrane, it plays a very important role in a signal transduction. Since the phosphatidylserine and the phosphatidylinositol are negatively charged on the head thereof and present in the inner layer of the cell membrane, a surface toward cytoplasm of the plasma membrane is negatively charged on the whole. Glycerol phospholipid has two fatty acids connected to glycerol. The fatty acids can be different each other. The third carbon of the glycerol is connected to phosphorous group to form phosphatidic acid, which is often in turn connected to other small molecules (phosphatidylethanolamine, phosphatidylcholine, phosphatidylserine or phosphatidylinositol).

Phosphatidylcholine is one of phospholipids and performs various good actions on the skin, such as effects of softening the skin, supplying moistures to the skin, protecting the skin and alleviating cutaneous disorder. Phosphatidylcholine has a structure in which fatty acid, phosphoric acid and choline are connected to the glycerin, wherein the fatty acid includes unsaturated fatty acid, which is essential fatty acid such as linoleic acid and linolenic acid. Unsaturated phosphatidylcholine is extracted mainly from the bean and the yolk of an egg, and comprises fatty acids

in portions of 12.9% palmitic acid, 4.4% stearic acid, 10.5% oleic acid, 66.5% linoleic acid and 5.7% linolenic acid. In particular, the linoleic acid among above fatty acids is also used as a cure for inflammatory acnes, but its mechanism is not clear yet. Acne patients are deficient in the content of linoleic acid, which is an essential fatty acid in the skin, so that a balance with other fatty acids is broken. At this time, there is a hypothesis that the linoleic acid supplied from the exterior compensates for the deficiency and thus alleviates the acne.

The linoleic acid has an effect of reducing production of sebum excessively secreted from the sebaceous glands. Accordingly, the phosphatidylcholine containing a large quantity of linoleic acid has a sufficient possibility of being used as a cure for the acnes.

According to the present invention, phosphatidylcholine and hydrogenated phosphatidylcholine in which all fatty acids of phosphatidylcholine are changed into saturated fatty acids was synthesized, and it was observed that they have an effect on the inhibition of sebum secretion and the growth of fat cells.

Sebaceous cells (sebocytes) are specific epithelial cells and secrete the sebum with rupturing after the end of differentiation. Dihydrotestosterone, which is one of androgens, is absolutely concerned with multiplication and growth of sebocytes. However, it is recently many reported that activation of peroxisome proliferator-activated receptor (hereinafter, referred to as PPAR) in the cell as well as male sex hormone plays an important role in lipid synthesis and metabolic mechanism.

The PPAR is an intranuclear hormone receptor activated by substrate binding and present as two types of retinoid X receptor and heterodimer. The activated receptor is bound to a PPAR response element, thereby controlling

peroxisome, microsome and genes related to lipid metabolism in mitochondria. The kinds of PPARs are various. Up to date, four kinds of PPARs, i.e., α , β , δ and γ are found. Particularly, PPAR γ is profoundly concerned with the lipid metabolism. It is known that PPAR γ is mostly distributed in the fat cell and secretes the sebum by accelerating differentiation of fat cell less differentiated and synthesizing the lipid. Accordingly, when any ingredient inhibits PPAR expression, it is meant that the ingredient can inhibit the differentiation of lipid cells. The substrate bound to PPAR γ includes various fatty acids and its metabolic body, for example, arachidonic acid and linolic acid. Prostaglandin J2 metabolic body and thiazolidinedione used for diabetes are also well known substrates bound to the receptors. It is known that PPAR γ has an anti-inflammatory action as well as the lipid metabolism-concerned controlling function. That is, when PPAR γ is activated, the generation of the cytokines, such as TNF α and IL-6, inducing inflammation is reduced. Accordingly, it may be expected that any material capable of reducing PPAR γ expression can reduce the number of fat cells by inhibiting differentiation of fat cells and thus be effective for the acnes and obesity. In addition, any material capable of increasing PPAR γ expression can be used for the acnes that inflammation excessively occurs around the comedo through bacterial propagation, by using it as inflammation alleviative material. It is known that the linoleic acid is bound to PPAR γ , increases the activation of the receptor thereof and thus alleviates the inflammation. The inventors found that phosphatidylcholine and hydrogenated phosphatidylcholine have an effect of inhibiting maturation and growth of sebocyte, which is a kind of fat cells. Also, it was found that hydrogenated phosphatidylcholine inhibits the differentiation of fat cells by hindering PPAR from generating mRNA. Accordingly, it can be seen that a composition comprising

phosphatidylcholine or hydrogenated phosphatidylcholine according to the present invention is effective against the acnes and obesity.

Laboratory animal used for confirming the effect of inhibiting the differentiation and growth of the sebocytes of each material was a Fuzzy rat and referred to as "WF/Pm-FZ". "WF" is derived from "Wistar Furth" group, "Pm" is indicative of Dr. Plam who made the species. "FZ" indicates that fuzzy gene is varied into recessive gene.

The phosphatidylcholine or hydrogenated phosphatidylcholine can be used in medicine as it is or a form of pharmaceutically acceptable salt. A kind of the above salt is not particularly limited to only if it is pharmaceutically acceptable. For example, hydrochloric acid, sulfuric acid, nitric acid, phosphoric acid, hydrofluoric acid, hydrobromic acid, formic acid, acetic acid, tartaric acid, lactic acid, citric acid, fumaric acid, maleic acid, succinic acid, metasulfonic acid, benzene sulfonic acid, toluene sulfonic acid, naphthalene sulfonic acid, etc. can be used.

In order to prepare the composition according to the present invention, when the phosphatidylcholine or hydrogenated phosphatidylcholine is formulated to pharmaceutical composition such as a cure for acne or obesity, various kinds of supplementary agent used in medicine as occasion demands, for example, carrier or other additives such as stabilizing agent, emollient and emulsifying agent may be added to it as long as the agent does not have a bad influence on the effective ingredient thereof.

In addition, the composition comprising the phosphatidylcholine or hydrogenated phosphatidylcholine according to the present invention may be provided by oral or parenteral administration. Transdermal administration is preferable as the parenteral route. Local application is most preferable. The

formulation includes ointment, cream, injection, powder, granulum, tablet and any formulation suitable for the pharmaceutical preparation.

In the pharmaceutical formulation, the content of phosphatidylcholine or hydrogenated phosphatidylcholine may be extensively changed according to the formulation and is based on the typical method. A preferred dosage of the pharmaceutical composition according to the present invention is 0.001~1000 mg/Kg·day.

The composition according to the invention may be dosed solely or equally to other medicine, or together with other medicine to aid it.

Meanwhile, the composition according to the invention may be cosmetics formulation such as cream, lotion, skin, essence, foundation, pack, cleansing cream, cleansing lotion, shampoo, and rinse. The cosmetics formulation can be used at any site such as the face, scalp, breast, and back, in which the acnes occur.

Hereinafter, the effect of the invention will be described more in details through the following embodiments.

Example

<Example 1. A preparation of high purity phosphatidylcholine (PC)>

The lecithin (1.5 kg) of soybean or yolk was dissolved to be 20 wt.% in solvent having hexane and isopropyl alcohol mixed in a ratio of 1:1, and then injected into a column (ID 15 × L 96 cm), which was filled with silica gel stabilized with the same solvent, by using a metering pump at a rate of 400 ml/min. After that, hexane/isopropyl alcohol solvents in which 5% water and 10% water are mixed were respectively flown six times and nine times. At the same time, each constituent fractions of phospholipid were identified in a discharge part of the

column with UV of 200 nm by using a spectrophotometer. Among them, the PC fraction was separately recovered. The recovered PC fraction was filtered with a 0.45 μm filter, vacuum-dried and then suspended in distilled water, freeze-dried. Resultantly, the soybean PC was obtained about 20% and the yolk PC was obtained about 67%.

<Example 2. A preparation of hydrogenated phosphatidylcholine (HPC)>

The high purity PC of soybean or yolk was dissolved in methanol/isopropyl alcohol/ethanol mixed solvent of about four times as much, then 4 % palladium catalyst was added to the solution and then heated to 50°C. After that, the double bond present in the high purity PC was reduced to convert it into saturated fatty acid under a hydrogen pressure of 15 atmospheres. The PC prepared like this was analyzed by gas chromatography. According to the result, it was identified that all were converted into corresponding saturated fatty acid.

<Example 3. An experiment of sebum secretion inhibition effect (animal experiment)>

Laboratory animal used for the present research was a male fuzzy rat (weight: about 300g) in 4 ~ 5 weeks after its birth, which over-secretes androgen by genetic variation. 7 weeks later, the fuzzy rat has a characteristic that sebum is secreted in its back site by the effects of androgen. A vehicle having solvent only was used as a control group and phosphatidylcholine and hydrogenated phosphatidylcholine were used as an experimental group, which were dissolved in solvent in an amount of 0.5% (mass/volume). The solvent was a material in which 1,3-butylene glycol, ethanol and distilled water were mixed in a ratio of 5:2:3 and

referred to as “vehicle”. All experimental materials were applied to the back region of the fuzzy rat with two times a day, i.e., in the morning and afternoon. The amount of application was about 1 ml. After checking the sebum secretion of the fuzzy rat applied by the vehicle, it was determined when the experiment would be ended. The experimental groups were observed with the naked eyes to check the sebum secretion inhibition effect per week. After the end of the experiment, it took photographs of states of the back region. The photographs are shown in Fig. 1.

As shown in Fig. 1, for the vehicle case, it was observed that yellow sebum was secreted in the back region of the fuzzy rat. However, for PC or HPC case, it was seen that the secretion amount of sebum was relatively small compared to the vehicle case.

Meanwhile, the present inventors quantitatively analyzed the fat cells through the thin-layer chromatography (TLC), based on the above results. The inventors performed a biopsy the backs of the rats of the vehicle control group and the above HPC treated experimental group. Dispase (GibcoBRL, USA), which is a digestive enzyme, was mixed with buffer solution of phosphate and the mixture was allowed for 12 hours at 4°C in the biopsied skin tissue. The epidermis was separated from the dermis and shaken in solution in which chloroform and methanol were present in a ratio of 2:1 and at the same time lipid was extracted. The extracted lipid was filtered with a 0.45 µm filter and then dissolved in chloroform solution (50µl). The prepared sample was dropped to the thin-layer chromatography (Merck, Germany) and developed in a development bath with development solution having composition as follows: hexane (to 19 cm), toluene (to 19 cm) and hexane: ether: acetic acid = 80: 20: 1. The development was developed twice. After that, 50% sulfuric acid solution was dispersed in sprinkles

and a process of color development was slowly performed in oven at 120°C. The results were shown in Fig. 2. Also, their results were quantitatively shown in Fig. 3a and Fig. 3b.

In Fig. 2, a, b, c and d are results regarding untreated epidermis, treated epidermis, treated skin and untreated skin, respectively. A red box at left indicates an internal standard (methyl oleate). Also, a', b' and c' indicates wax ester (WE), triglycerol (TG) and free fatty acid (FFA), respectively.

As shown in Fig. 2, all skin lipids were remarkably reduced in the HPC-treated group, which were definitely different from the non-treatment group. Figs. 3a and 3b quantitatively shows the results.

Fig. 3a is a graph showing a comparison of the treatment group and the non-treatment group regarding the total lipids, and Fig. 3b is a graph showing a comparison of the treatment group and the non-treatment group according to kinds of the lipids. According to the Figs., with regard to the total lipids, the treatment group was significantly reduced compared to the non-treatment group. In addition, regarding respective lipid of the free fatty acid (FFA), the wax ester (WE) and the triglycerol (TG), the treatment group was significantly reduced compared to the non-treatment group. Since the wax ester is lipid derived from the sebaceous glands, the above results show that the effect, which the HPC inhibits the sebum secretion, is excellent. In addition, the results regarding the FFA and TG show that the HPC has the effect of preventing or treating fat cells-related diseases such as obesity, etc. by inhibiting the differentiation and growth of the fat cells.

<Example 4. A preparation of cream formulations>

(Cream formulation 1)

Table 1

Ingredients	Contents
PC or HPC	0.5%
Propylene glycol	20.0%
Stearyl alcohol	6.5%
Cetyl alcohol	3.5%
Sorbitan monostearate	3.0%
Polysorbate 60	2.0%
Isopropyl myristate	1.0%
Anhydrous sodium sulfite	0.2%
Polysorbate 80	0.1%
Purified water	To 100

(Preparation method)

Stearyl alcohol, cetyl alcohol, sorbitan monostearate and isopropyl
5 myristate were introduced into a double-walled vessel and heated until the mixture
was completely dissolved. The mixture was added to a separately prepared mixture
of purified water propylene glycol and polysorbate 60 at 70~75°C while using a
homogenizer for liquid. The produced emulsion was continued to mix and cooled
below 25°C. Solution of PC or HPC, polysorbate 80 and solution of purified water
10 and solution of anhydrous sodium sulfite in purified water were subsequently added
to the emulsion while continuously mixing. Cream was homogenized and then used.

(Cream formulation 2)

Table 2

Ingredients	Contents
PC or HPC	0.5%
Hydroxypropyl beta-cyclodextrin	20.0%
Stearyl alcohol	2.5%
Cetyl alcohol	2.5%
Mineral oil	11.0%
Glycerol monostearate	2.5%
Glycerol	5.0%
Sorbate 60	2.0%
Polysorbate 60	3.5%
Purified water	To 100

(Preparation method)

An appropriate amount of hydrochloric acid was added until it was a
5 solution. An appropriate amount of sodium hydroxide was added until pH was 6.0.
An appropriate amount of purified water was added to be 100 mg. PC or HPC was
added to a solution of hydroxypropyl beta-cyclodextrin in purified water while
stirring. Hydrochloric acid was added to be solution and then sodium hydroxide
was added until pH was 6.0. Glycerol and polysorbate 60 were added while stirring
10 and the mixture was heated to 70°C. The mixture produced while slowly stirring
was added to a mixture of mineral oil, stearyl alcohol, cetyl alcohol, stearyl
monostearate and sorbate 60 at 70°C. After cooling below 25°C, remained amount
of purified water was added and the mixture was mixed until it was homogenized.

(Cream formulation 3)

Table 3

	INCI name	Contents
Water-phase part	Disodium EDTA	0.020
	Glycerine	4.000
	Butylene glycol	2.000
	Xanthan gum	0.030
	Triethanolamine	0.200
	Di-water	To 100
	Carbomer	0.1
Oil-phase part	Stearic acid	1.800
	Glyceryl stearate PEG-100 stearate	1.000
	Cetearyl alcohol	2.000
	Glyceryl stearate	2.000
	Sorbitan sequeioleate	0.300
	Polysorbate 60	1.200
	Mineral oil	7.000
	Isopropyl myristate	1.500
	Cetyl octanoate	1.500
	PC or HPC	0.5
	Dimethicone	0.400
	Preservative	Q.S.

(Preparation method)

The water-phase part and the oil-phase part were respectively heated to 75°C. After checking that the water-phase part and the oil-phase part were completely dissolved, the water-phase part was poured into a main kiln. And then, while the oil-phase part was slowly poured into the main kiln, the mixture was stirred at 3,500 rpm by a homomixer and at 30 rpm by a paddle mixer for 3 minutes and then cooled.

<Example 5. An experiment of sebum secretion inhibition effect of composition of cream formulation>

This experiment was practiced based on the method described in Example 3. Phosphatidylcholine (PC) and hydrogenated phosphatidylcholine (HPC) were used in experimental groups in a density of 0.5~30% (mass/volume), respectively. They were used in various formulations as follow. All experimental formulations were applied to the back region of the fuzzy rat with two times a day, i.e., in the morning and afternoon. The amount of application was about 1 ml. The experimental groups were observed with the naked eyes to check the sebum secretion inhibition effect per week. After the end of the experiment, it took photographs of states of the back region. The results are shown in Fig. 4

As it can be seen from Fig. 4, the cream formulations exhibited an excellent effect of inhibiting the sebum secretion, compared to the non-treatment group.

<Example 6. A preparation of liposome formulations>

(Liposome formulation 1)

Table 4

Ingredients	Contents
PC or HPC	30.0%
Cholesterol	5.0%
Ethyl alcohol	10.0%
Methyl paraffin	0.15%
Propyl paraffin	0.02%
Disodium edetate	0.15%
Sodium chloride	0.4%
Hydroxypropylmethylcellulose	1.2%
Purified water	To 100

(Preparation method)

Purified water was added to be 100 g. A mixture of PC or HPC, cholesterol and ethyl alcohol was stirred, heated at 55~60°C and homogenized until it was a solution, and it was added to a solution of methyl paraffin, propyl paraffin, disodium edetate and sodium chloride in purified water. Hydroxypropylmethylcellulose in purified water was added and continued to stir until it was swelled.

(Liposome formulation 2)

Table 5

Ingredients	Contents
PC or HPC	10.0%
Cholesterol	1.0%

Ethyl alcohol	7.5%
Hydroxypropylmethylcellulose	1.5%
Purified water	To 100

(Preparation method)

Sodium hydroxide (1N) was added to be pH 5.0. Purified water was added to be 100 g. A mixture of PC or HPC and cholesterol in ethyl alcohol was stirred and heated at 40°C until it was a solution. Alcoholic solution was slowly added to water solution while homogenizing for 10 minutes. Hydroxypropylmethylcellulose in purified water was added and mixed until it was swelled. Sodium hydroxide (1N) was added to the produced solution to regulate the pH 5.0, and remained purified water was diluted.

<Example 7. An experiment of sebum secretion inhibition effect of composition of liposome formulation>

The composition of liposome formulation of Example 6 was experimented after applying to the fuzzy rat in a manner of transdermal administration, like the method of Example 5. The results are shown in Fig. 5.

It could be seen that the liposome formulations also effectively inhibited the sebum secretion, although there were differences according to densities of PC or HPC.

<Example 8. A histological observation of sebocytes>

In order to do histological research, after finishing the experiment in Example 3, tissue of the back region of fuzzy rat was collected by a biopsy punch.

The collected tissue was attached to a slide in fixing fluid of formalin, and then variations of sizes and shapes of the sebaceous glands and the pores and thickness of the horny layer were observed through methods of hematoxylin & eosin (H&E) staining and fat staining method (Oil Red O). The results of H&E staining were shown in Figs. 6a to 6c, and the results of the fat staining method were shown in Figs. 6d to 6f. Also, in Fig. 6g, a graph numerically showing the above results was shown.

As can be seen from the above Figs., the development of the sebaceous glands was slowed down in the PC and HPC-treated groups, compared to the vehicle-treated group. It was also seen that the number of activated sebaceous glands and sizes of the sebaceous glands were reduced. Accordingly, it can be seen that PC and HPC effectively inhibited the differentiation and growth of the sebocytes.

Further, according to the measurements of the number of the sebocytes, a result was obtained as shown in Fig. 7. As shown in Fig. 5, it can be seen that the number of sebaceous glands was significantly reduced in the PC and HPC-treated group, compared to the vehicle-treated group. That is, it can be known that PC and HPC effectively inhibited the differentiation and maturity of the sebocytes.

<Example 9. Effects of HPC on mRNA transcription of PPAR γ >

In order to check that the control group and the experimental materials accelerate or inhibit an expression of PPAR γ concerning with the sebum generation, it was performed a reverse transcription-polymerase chain reaction (PCR). After the experiment period of two months was terminated, tissue of the back region of fuzzy rat to which each of the experimental materials was applied was collected,

and then total RNAs were extracted by using Rneasy Protect (Qiagen). A reverse transcription was performed to obtain cDNA. The extracted RNA (5 µg) and oligo-dT (1 µg) were mixed and reacted with an appropriate amount of distilled water at 70°C for 10 minutes to cause denaturation of RNA. After cooling at 4°C for 10 minutes, composition of cDNA synthetic reaction (5× reverse transcription buffer solution, 2.5 mM dNTPs, 0.1M DTT and 200 unit of MMLV reverse transcription enzyme) was tailored and reacted at 37°C for 60 minutes so that oligo-dT was bound to RNA to synthesize cDNA.

The PCR was performed to amplify PPAR γ mRNA to be checked. A reaction mixture contained synthesized cDNA (2µl), upstream primer (10 pmol/µl), downstream primer (10 pmol/µl), 10 mM dNTP, 10×reactant buffer solution and Taq Polymerase (5 unit/µl) in distilled water. The reaction mixture made was subject to 30 cycles (94°C 3 min., 94°C 30 sec., 60°C 3 min., 72°C 1 min., 72°C 9 min.). Upstream primer of PPAR γ was ATA AAG TCC TTC CCG CTG ACC AAA GCC and downstream primer thereof was GCG GTC TCC ACT GAG AAT AAT GAC AGC. β -actin was made to be a standard to compare relative expressions. Upstream primer of β -actin was CAT GCC ATC CTG CGT CTG GAC and downstream primer thereof was TAC TCC TGC TTG CTG ATC CAC ATC TGC. The results were shown in Fig. 8.

As shown in Fig. 8, mRNA generation of PPAR γ was remarkably reduced in tissues applied by HPC, compared to the non-treatment vehicle group. Based on this result, it is believed that HPC reduces mRNA generation of PPAR γ in the PPAR γ mechanism and thus inhibits the differentiation of fat cells and weakens the lipid generation.

<Example 10. Effects of PC and HPC on mRNA transcription of PPAR γ in fat cells originated in rat>

In order to check that the control group and the experimental materials accelerate or inhibit an expression of PPAR γ concerning with the differentiation of fat cells, it was performed a reverse transcription polymerase chain reaction (PCR) in a cell level. 3T3 L1, which is preadipocyte originated in a rat, was purchased from the American Type Culture Collection (ATCC) (ATCC No. CL-173) and cultured on culture medium under a condition of 37°C and 5% carbon dioxide, based on information of provider. While culturing it under a differentiation condition of fat cells (1 μ M dexamethasone, 0.5 mM isobutylmethylsanthine, 10 μ g/ml insulin) by Bernlohr (1984, Proc. Natl. Acad. Sci USA 91:5439-5472), the vehicle of control group and the experimental materials of PC and HPC were treated. After the end of the experiment, cells were obtained and collected and total RNAs were extracted by using Rneasy Protect (Qiagen, USA). From that, the reverse transcription PCR (Premix, Bioneer, Korea) was performed to obtain cDNA. Firstly, the extracted RNA (5 g) and oligo-dT (1 g) were mixed and reacted with appropriate amount of distilled water at 70°C for 10 minutes to cause denaturation of RNA. After cooling 4°C for 10 minutes, composition of cDNA synthetic reaction (5 $\times$ reverse transcription buffer solution, 2.5 mM dNTPs, 0.1M DTT and 200 unit of MMLV reverse transcription enzyme) was tailored and reacted at 37°C for 60 minutes so that oligo-dT was bound to RNA to synthesize cDNA.

The PCR was performed to amplify PPAR γ mRNA to be checked. A reaction mixture contained synthesized cDNA (2 μ l), forward primer (10 pmol/ μ l), reverse primer (10 pmol/ μ l), 10 mM dNTP, 10 $\times$ reactant buffer solution and Taq Polymerase (5 unit/ μ l) and was reacted in distilled water. The reaction mixture

made was subject to 30 cycles (94°C 3 min., 94°C 30 sec., 60°C 3 min., 72°C 1 min., 72°C 9 min.). Forward primer of PPAR γ was ATGACAGCGACTTGGCAATA and reverse primer thereof was GCAATTGGAAGAAGGGAAT. β -actin was made to be a standard to compare relative expressions. Upstream primer of β -actin was CATGCCATCCTGCGTCTGGAC and downstream primer thereof was TACTCCTGCTTGCTGATCCACATCTGC. After the end of the reaction, the expression was measured with performing 1.2% refined agarose electrophoresis and thus checking the bands. The result was shown in Fig. 10.

In Fig. 10, a, b, c, d, e and f are expression amounts of PPAR γ in preadipocytes, non-treated fat cells, HPC (20 μ M)-treated fat cells, HPC (5 μ M)-treated fat cells, PC (5 μ M)-treated fat cells and PC (20 μ M)-treated fat cells, respectively. As can be seen from Fig. 10, the expression amount of PPAR γ was high in non-treated fat cells (b). However, PPAR γ was not almost expressed in c and d cases treated with HPC. Also, the expression amount of PPAR γ was very low in PC-treated fat cells (e and f).

Accordingly, it was proven in cell level that HPC and PC, particularly HPC has a function of inhibiting differentiation and growth of fat cells.

<Example 11. An analysis of secreted lipid through a thin-layer chromatography (TLC) for measuring a sebum secretion inhibiting effect of HPC in the skin of human body>

As the above Examples, an analysis of the secreted lipid was already performed in the cell unit and animal experiments. In addition to this, a clinical testing on the human body was carried out. Cream formulations comprising HPC

and cream formulations comprising not HPC were medicated to man volunteers having no problems on health who were 25~30 years old. According to Downing method, it was measured the amount of lipid, which is secreted over at least 15 hours. Firstly, for 12 hours, to remove the secreted sebum and the lipid secreted from the lipid glands, non-woven fabric of bentonite clay to which water was added was patched to the forehead and allowed. After the lapse of the time, the fabric was removed and kept until the test was carried out. Cream formulation 1 was applied to regions 2 cm away from the center of the forehead (it was applied before the test (0 week), after 2 weeks, 4 weeks and 6 weeks, respectively, and extracted), and then the fabric was again patched and subject to absorbing the lipids being secreted for 3 hours. The absorbed lipids were extracted with ether and quantitatively analyzed as method of the Example 3. The result was shown in Figs. 11a and 11b.

Fig. 11a shows a variation of total amount of lipid and Fig. 11b shows a variation of an amount of sebum. Fig. 11a shows an amount of lipid secreted for 12 hours and amounts of secreted lipids of the non-treatment group and the treatment group for 3 hours after the 12 hours. The amount of lipid was gradually decreased as a time goes on.

Fig. 11b shows a variation of an amount of sebum, which was calculated based on 25% contents of wax ester. The sebum secretion was remarkably decreased in the HPC-treated group.

According to the above results, it was seen that the total lipid was decreased from 2 weeks after the formulations comprising HPC was treated and the actually secreted lipid was also significantly decreased.

By using the composition of the invention, fat cells-related disorders can be treated or prevented by inhibiting the differentiation and growth of fat cells such as

sebocytes. In particular, the obesity due to hypertrophy of fat cells or over-proliferation of fat cells can be fundamentally treated and the acnes can also be fundamentally treated or prevented by inhibiting the sebum secretion. The invention can also be effectively used in combination for preventing and treating
5 the fat cell-related disorders.

While the invention has been shown and described with reference to certain preferred embodiments thereof, it will be understood by those skilled in the art that various changes in form and details may be made therein without departing from the spirit and scope of the invention as defined by the appended claims.

10

WHAT IS CLAIMED IS:

1. A composition for inhibiting differentiation or growth of fat cells comprising one or two selected from the group consisting of phosphatidylcholine and hydrogenated phosphatidylcholine as an active ingredient.

5

2. The composition according to claim 1, wherein the fat cells is sebaceous cells.

3. The composition according to claim 1 or 2, wherein the composition is
10 used for treating a disease caused by at least one of abnormal lipid metabolism, hypertrophy of fat cells, over-activation and abnormal proliferation of fat cells.

4. The composition according to claim 3, wherein the disease is obesity or acne.

15

5. A composition for inhibiting sebum secretion comprising one or two selected from the group consisting of phosphatidylcholine and hydrogenated phosphatidylcholine as an active ingredient.

20

6. A composition for treating or inhibiting obesity comprising one or two selected from the group consisting of phosphatidylcholine and hydrogenated phosphatidylcholine as an active ingredient.

7. The composition according to one of claims 1, 5 and 6, the
25 phosphatidylcholine is derived from an egg or a soybean.

8. The composition according to claim 7, the phosphatidylcholine or the hydrogenated phosphatidylcholine is contained solely or in combination with other active ingredients.

5 9. A kit for treating obesity comprising the composition for inhibiting differentiation or growth of fat cells according to claim 1 or the composition for treating or inhibiting obesity according to claim 6.

10 10. A kit for treating a acne comprising the composition for inhibiting differentiation or growth of fat cells according to claim 2 or the composition for inhibiting sebum secretion according to claim 5.

15 11. The kit according to claim 9, further comprising other compositions for inhibiting differentiation or growth of fat cells or other compositions for treating or preventing obesity.

 12. The kit according to claim 10, further comprising other compositions for inhibiting sebum secretion or other compositions for inhibiting differentiation or growth of sebocytes.

20

AMENDED CLAIMS

[received by the International Bureau on 27 Sep 2004 (27.09.2004);
original claims 1-12 replaced by amended claims 1-4 (1 page)]

1. A composition for inhibiting differentiation or growth of fat cells comprising
one or two selected from the group consisting of phosphatidylcholine and hydrogenated
5 phosphatidylcholine as an active ingredient.
2. The composition according to claim 1, wherein the fat cells is sebaceous cells.
3. The composition according to claim 1 or 2, the phosphatidylcholine is
10 derived from an egg or a soybean.
4. The composition according to claim 3, the phosphatidylcholine or the
hydrogenated phosphatidylcholine is contained solely or in combination with other
active ingredients.

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Fig. 1

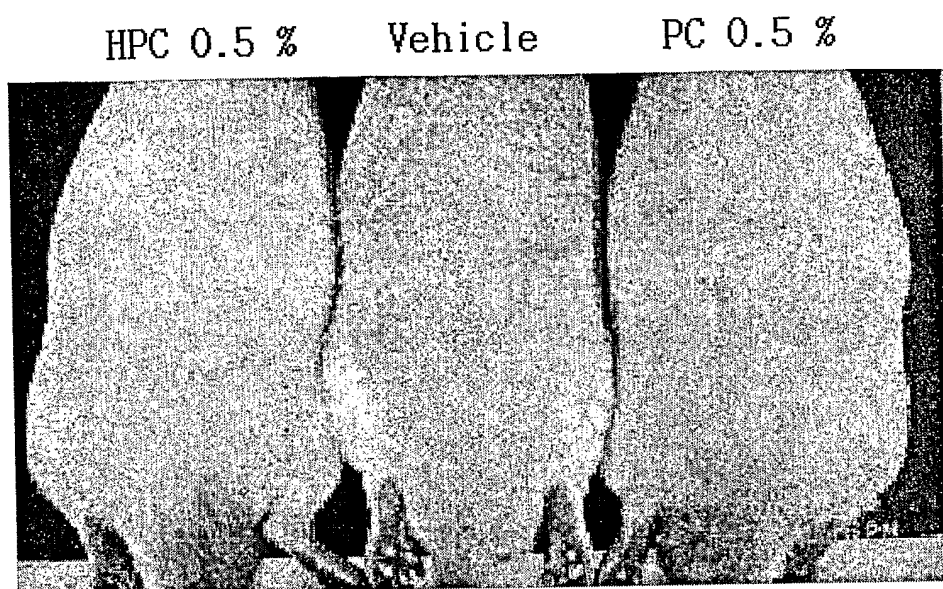
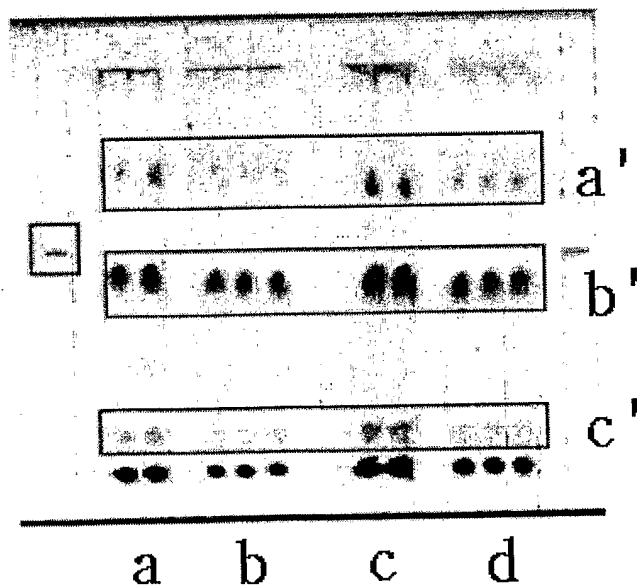
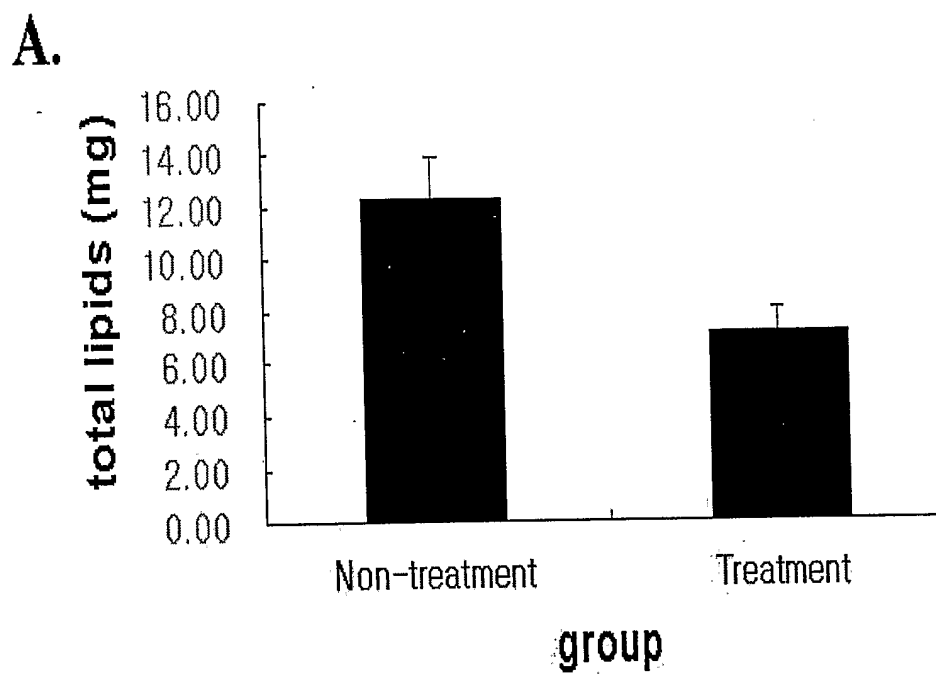


Fig. 2



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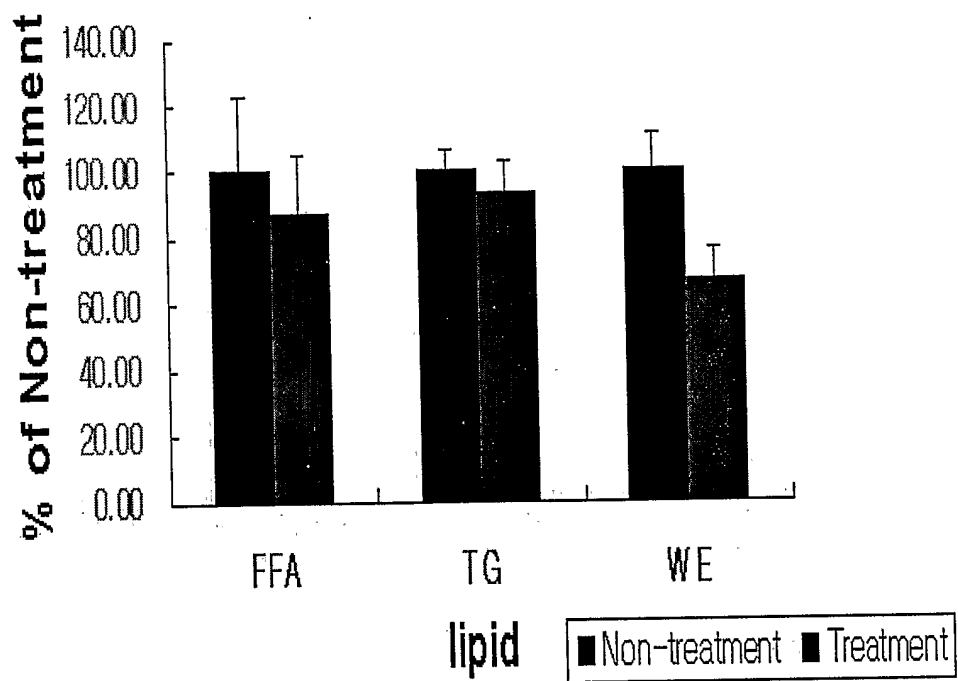
Fig. 3a



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Fig. 3b

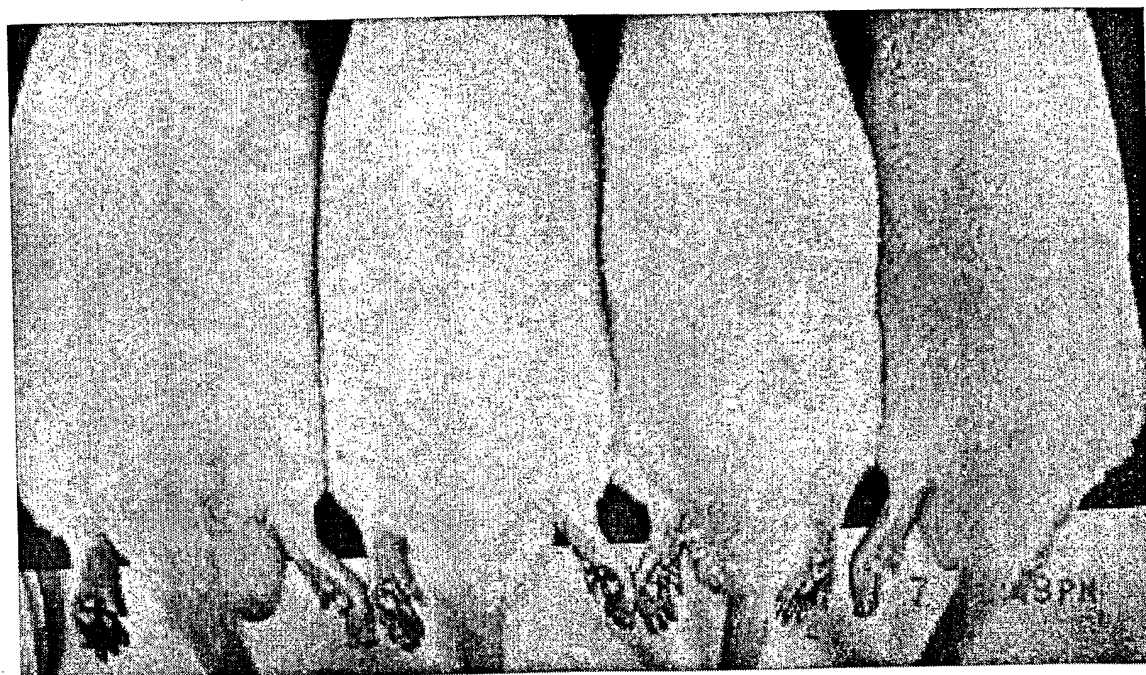
B.



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Fig. 4

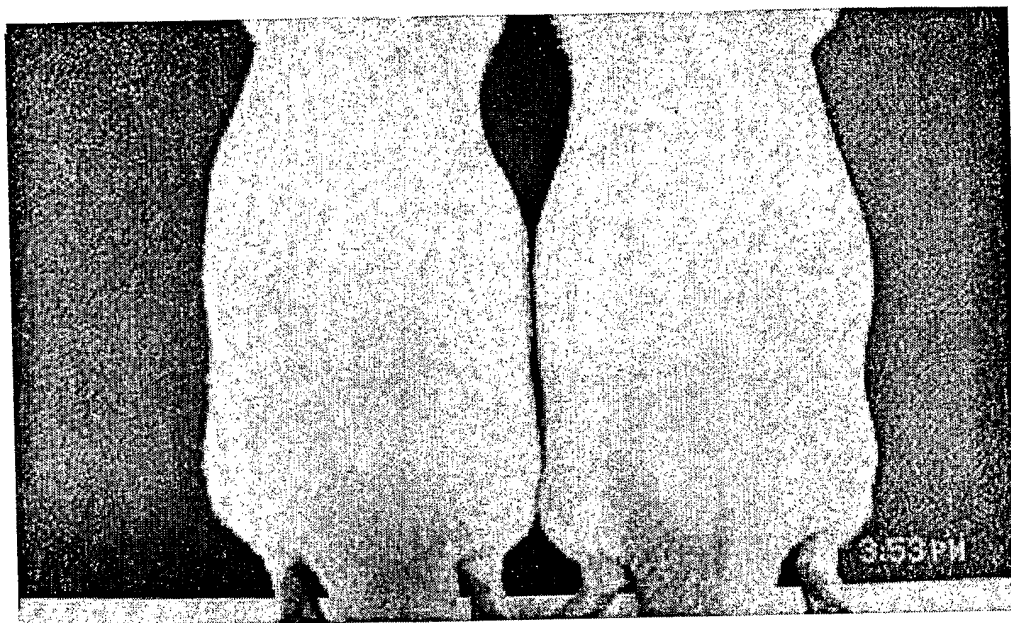
cream formulation 3 cream formulation 1
cream formulation 2 non-treatment group



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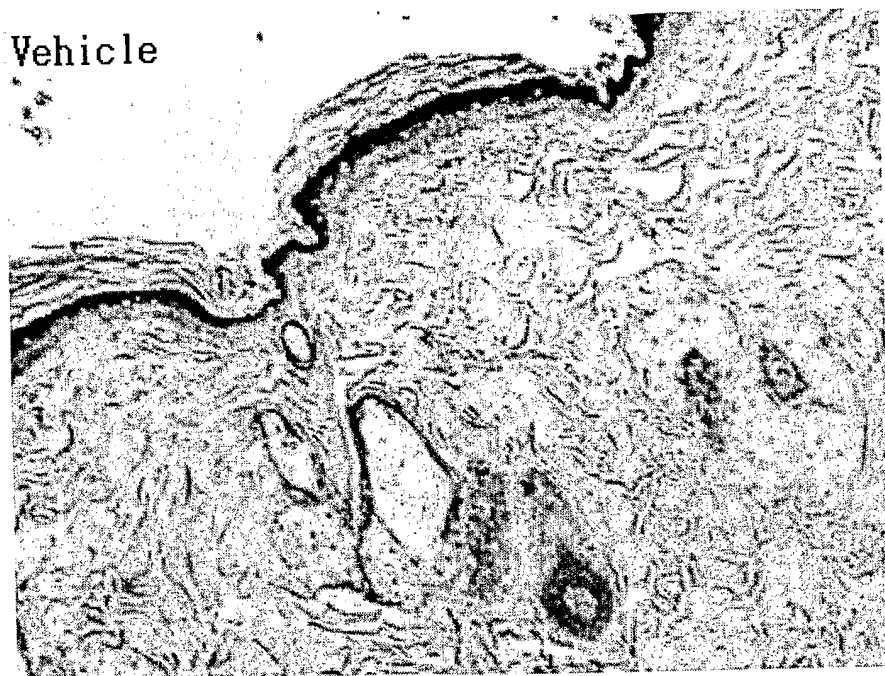
Fig. 5

liposome formulation 2 liposome formulation 1



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Fig. 6a



photograph of skin tissue of the vehicle group (H&E staining)

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Fig. 6b

PC



photograph of skin tissue of PC
formulation-treated group (H&E staining)

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Fig. 6c



photograph of skin tissue of the HPC
formulation-treated group (H&E staining)

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Fig. 6d



photograph of tissue of the
vehicle group

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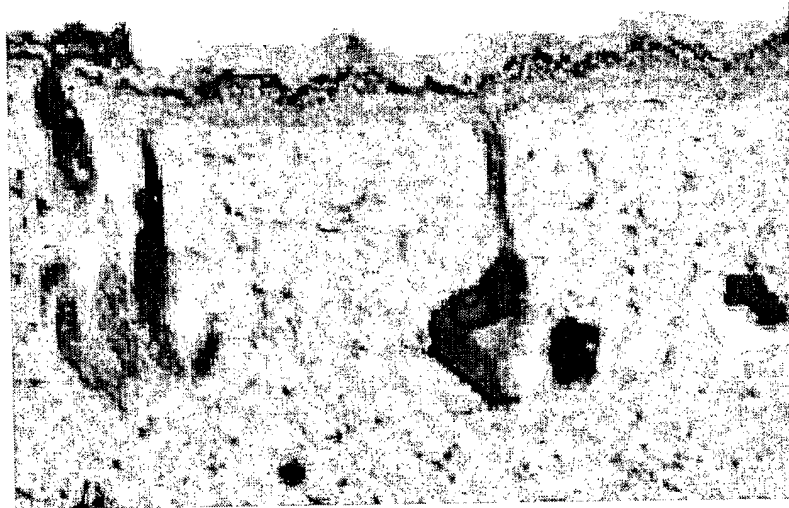
Fig. 6e



photograph of tissue of the PC-
treated group

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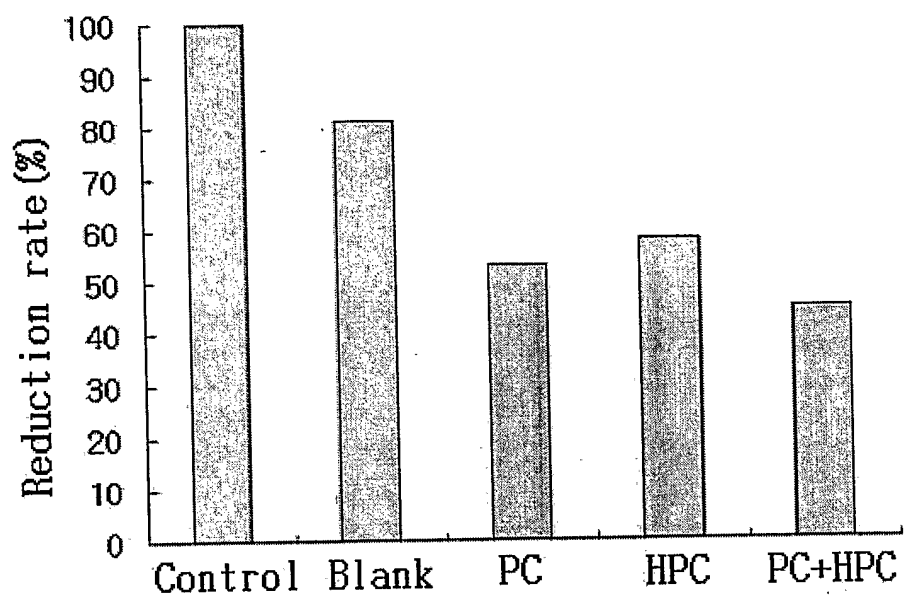
Fig. 6f



photograph of tissue of the HPC-
treated group

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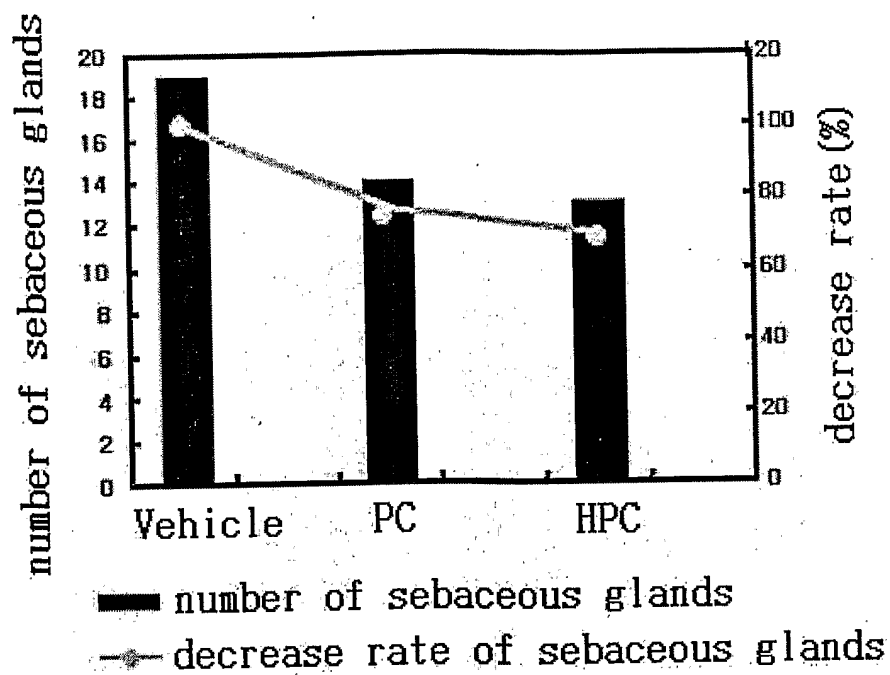
Fig. 6g



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Fig. 7

variation of the number of sebaceous glands



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Fig. 8

V: Vehicle

PC: phosphatidylcholine

HPC: hydrogenated phosphatidylcholine

V x5 HPC x5 (β -Actin)

V HPC

PPAR γ

Fig. 9



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Fig. 10

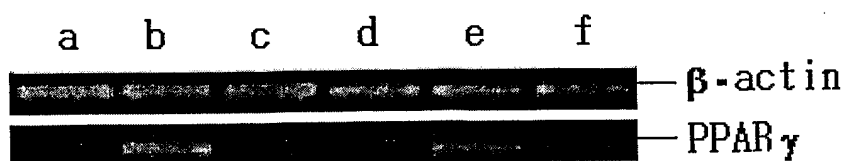
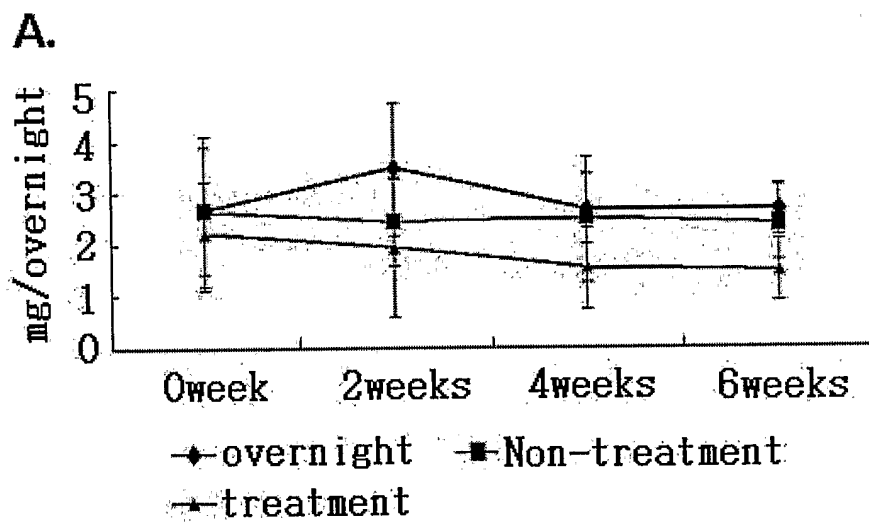
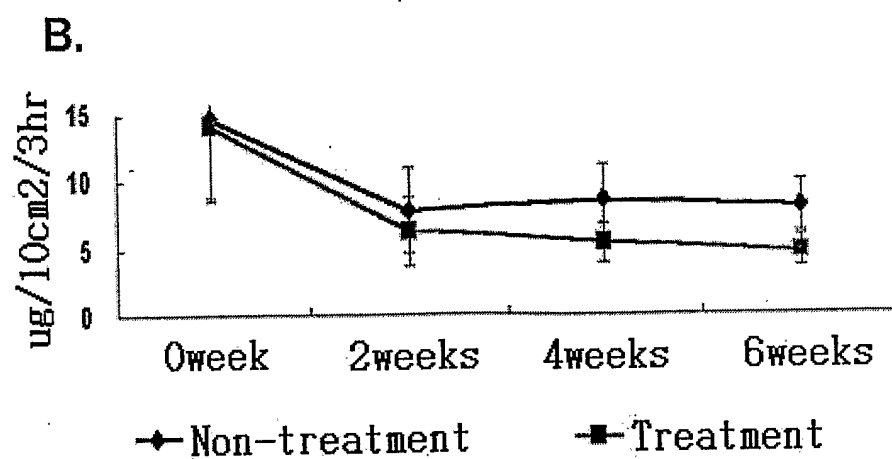


Fig. 11a



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Fig. 11b



INTERNATIONAL SEARCH REPORT

International application No.
PCT/KR2004/001314

A. CLASSIFICATION OF SUBJECT MATTER**IPC7 A61K 31/685**

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)

IPC7 A61k 31/685

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
Korean patents and applications for inventions since 1975

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

STN[CAPLUS, BIOSIS; phospholipid? and (adipocyte? or sebum? or obesity or PPAR or acne)]

Medline

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	JP 10-84880 A (TAKEDA SHOKUHIN KOGYO KK, HONEN CORP.) 07 April 1998 See abstract, claims, and Table 2. (Family: None)	3 - 9, 11
X	DE 19857491 A1 (KUHS G.M.B.H. & CO.) 15 June 2000 See abstract and examples 1, 7, 19. (Family: None)	3-5, 7, 8, 10, 12
X	GHYCZY et al., 'The treatment of acne vulgaris by phosphatidylcholine from soybeans, with a high content of linoleic acid', Journal of Applied Cosmetology, 1996, Vol.14(4), pp.137-145 See the whole document.	3-5, 7, 8, 10, 12
X	MORGANTI, 'Phospholipids in acne therapy', Cosmetic News, 2001, Vol.24(137), pp.89-92 See summary.	3-5, 7, 8, 10, 12
A	DAVIES et al., 'Oxidized alkyl phospholipids are specific, high affinity peroxisome proliferator-activated receptor gamma ligands and agonists', Journal of Biological Chemistry, 2001, Vol.276(19), pp. 16015-16023 See the whole document.	1 - 12

☐ Further documents are listed in the continuation of Box C.

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10 AUGUST 2004 (10.08.2004)

Date of mailing of the international search report

12 AUGUST 2004 (12.08.2004)

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