Abstract: The present invention relates to a composition for the prevention and treatment of asthma, which contains α-lipoic acid as an active ingredient. The inventive composition reduces not only airway hyperreactivity, a typical symptom of asthma, but also inflammation occurring in asthma. Accordingly, the inventive composition will be useful for the prevention and treatment of asthma.
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COMPOSITION FOR PREVENTING AND TREATMENT OF ASTHMA

COMPRISING ALPHA-LIPOIC ACID AS AN EFFECTIVE COMPONENT

Background of the Invention

Technical Field

The present invention relates to a composition for the prevention and treatment of asthma.

Background Art

Asthma is a chronic allergic disease with a prevalence rate of 5-10% and its attack rate increases rapidly. Etiological factors of asthma include chronic allergic inflammation of the airway, and airway hyperreactivity, and their clinical features include recurrent attacks of difficult breathing, and syndromes of cough and phlegm. The exact cause of asthma and a mechanism by which asthmatic attack occurs is still unclear, but it is guessed that genetic, developmental and environmental factors are important causes of asthma.

As therapeutic agents of bronchial asthma, steroidal agents and bronchodilators have been mainly used till now, but they temporarily relieve asthma symptoms and cannot provide a complete recovery. Also, the steroidal agents have a high possibility of side effects in long-term use.

Accordingly, there is an urgent need for the development of new therapeutic agents which can solve shortcomings with the existing asthma therapeutic agents while maximizing a therapeutic effect on asthma.

Meanwhile, \( \alpha \)-lipoic acid (also known as thiocyst acid, 6,8-dithiooctanoic acid, or 1,2-dithiolane-3-pentanoic acid), which is a trace component contained in
extracts from the liver and yeast, appeared as a new vitamin, since it was known to be a growth factor of lactic acid. This trace component was first isolated from the liver by L. J. Reed, et al. in the year 1951 and named “α-lipoic acid”. Then, its structure was established by synthesis.

Recently, α-lipoic acid and its reduced form, dihydrolipoic acid, were known to have a function as an antioxidant and to react with reactive oxygen species, such as peroxide, hydroxyl and peroxy radicals and single oxygen. Moreover, they are known to have a function of protecting cellular membranes by reaction with vitamin C or glutathione and to effectively inhibit oxidative stress.

The therapeutic effect of α-lipoic acid on bronchial asthma has not been known yet.

**Disclosure of Invention**

The present inventors have found that α-lipoic acid allows asthma to be treated without side effects, thereby completing the present invention.

Accordingly, an object of the present invention is to provide a composition which has excellent preventive and therapeutic effects on asthma without causing side effects.

The present invention provides a composition for the prevention and treatment of asthma.

The inventive composition contains α-lipoic acid as an active ingredient.

The inventive composition shows bronchial hyperreactivity inhibitory activity.

The inventive composition shows anti-allergic inflammatory activity.

Concretely, the anti-allergic inflammatory activity of the inventive
composition is caused by a reduction in inflammatory cells in the airway, a reduction in the ratio of eosinophils in the total blood cells of bronchoalveolar lavage fluid, and a reduction of immunoglobulin E.

Although a mechanism that the inventive composition inhibits bronchial hyperreactivity or shows anti-inflammatory activity was still not established, it is believed that the inventive composition can inhibit inflammatory cytokine causing an allergic inflammatory reaction, thus inhibiting the various pathophysiological characteristics of bronchial asthma, such as eosinophilic allergic airway inflammation, allergens, particularly immunoglobulin, and airway hyperreactivity.

In addition to α-lipoic acid as an active ingredient, the inventive composition may also comprise at least one additional active ingredient having the same similar function as α-lipoic acid.

In addition to such active ingredients, the inventive composition may contain pharmaceutically suitable, physiologically acceptable adjuvants, including solvents, disintegrants, sweeteners, binders, coating agents, swelling agents, lubricants, flavoring agents, solubilizers.

For administration, the inventive composition may also contain at least one pharmaceutically acceptable carrier, in addition to the active ingredients as described above.

Examples of the pharmaceutically acceptable carrier include saline solution, sterile water, Ringer’s solution, buffered saline solution, dextrose solution, maltodextrin solution, glycerol, ethanol and a mixture of two or more thereof. If necessary, the inventive composition may also contain other conventional additives, such as antioxidants, buffers and bacteriostatic agents.

Moreover, the inventive composition may additionally contain diluents,
dispersants, surfactants, binders and lubricants in order to formulate it into injection formulations, such as aqueous solution, suspension and emulsion, pills, capsules, granules and tablets. Furthermore, the inventive composition may preferably be formulated depending on particular diseases and its components, using the method described in Remington’s Pharmaceutical Science (latest edition), Mack Publishing Company, Easton PA, which is a suitable method in the relevant field of art.

The inventive composition may be administered in the conventional manner via the subcutaneous, intravenous, intraarterial, intraabdominal, intramuscular, intrasternal, percutaneous, intranasal, inhalation, topical, rectal, oral, intraocular or intradermal route.

The inventive composition is preferably administered at the amount of less than 600 mg per day for adults, but it was confirmed that the composition is relatively safe event at 30 mg/kg/day. The amount of the composition administered can vary depending on various factors, including the kind and severity of diseases, the kind and content of an active ingredient and other components contained in the composition, the kind of a formulation, and patient’s age, weight, general health condition, sex and diet, and administration time, administration route, the secretion % of the composition, administration period, and the kind of drugs used in combination.

The inventive composition was proven to have no particular side effects on a living body.

The inventive composition prepared as such can be advantageously used for the prevention and treatment of asthma without particular side effects, and it may be used alone or in combination with diet therapy, exercise therapy, liposuction, hormone therapy, chemical therapy and other methods using biological reaction
regulators.

In another aspect, the present invention provides the use of $\alpha$-lipoic acid as an active ingredient in the preparation of a composition for the prevention and treatment of asthma.

**Brief Description of Drawings**

FIG. 1 shows a reduction in airway hyperreactivity in asthma-induced mice administered with the inventive composition.

A: an intra-abdominal administration group.

B: an oral administration group.

FIG. 2 shows a reduction in airway inflammation lesion in asthma-induced mice administered with the inventive composition.

A: a graph showing lesion scores in the tissue of an intra-abdominal administration group.

B: a photograph showing the staining of bronchoalveolar tissue in an oral administration group.

FIG. 3 shows a reduction in immunoglobulin E in asthma-induced mice administered with the inventive composition.

A: an intra-abdominal administration group.

B: an oral administration group.

FIG. 4 shows a reduction in eosinophils in asthma-induced mice administered with the inventive composition.

A: an intra-abdominal administration group.

B: an oral administration group.
Best Mode for Carrying Out the Invention

The present invention will hereinafter be described in further detail by examples. It will however be obvious to a person skilled in the art that the present invention is not limited to or by the examples.

Example 1: Airway hyperreactivity inhibitory activity of α-lipoic acid-containing composition according to the present invention

To prepare asthma models, 6-week old mice were selected and sensitized with ovalbumin in the following manner. First, at days 1 and 14 of an experiment, 10 μg of egg albumin was administered into the abdominal cavity of mice together with 2 mg of alum. At days 28, 29 and 30 of the experiment, 1% ovalbumin solution prepared by dilution in phosphate buffered saline was nebulized with an ultrasonic nebulizer (UltraNeb 2000) for 30 minutes each day, thus preparing asthma models displaying bronchial hyperreactivity.

In the above procedure, the mice were divided into two groups each consisting of 9-10 animals. The two groups were administered with α-lipoic acid via oral and intra-abdominal routes, respectively, in order to examine the airway hyperreactivity inhibitory effect of α-lipoic acid.

For the intra-abdominal administration group, a composition containing α-lipoic acid at a concentration of 25 mg/ml was administered intra-abdominally such that α-lipoic acid was administered at the amount of 1 mg, 4 mg, 16 mg or 64 mg each day. For the oral administration group, a composition comprising feed containing α-lipoic at 0.125 wt%, 0.25 wt%, 0.5 wt% or 1 wt% was administered orally each day. In addition to such two groups, not only an asthma control group where any drug had not been administered and bronchial hyperreactivity had occurred, but also a normal control group where any drug had not been administered
and bronchial hyperreactivity had occurred, were provided.

At day 31 of the experiment, the animal groups were subjected to a methacholine challenge test to measure bronchial hyperreactivity. As an index of bronchial hyperreactivity, enhanced pause (Penh) value was measured by a mouse body volume plethysmograph (Allmedicus Co., Korea) in the following manner.

Methacholine was nebulized at gradually increasing concentrations of 2.5, 5, 10 and 20 mg/ml for 3 minutes for each concentration, and then Penh value was measured for 3 minutes. The Penh value measured after nebulization of each concentration of methacholine was statistically processed after expressing an increase compared to basal value (value measured after initially nebulizing physiological saline solution for 3 minutes), as a percentage, and the results are shown in FIG. 1.

As shown in FIG. 1A, it could be confirmed that the test group, which had been administered intra-abdominally with the inventive composition containing 25 mg/ml of α-lipoic acid, shows a lower increase in airway resistance upon methacholine challenge than that in the asthma control group with no administration.

Also, as shown in FIG. 1B, it could be found that the test group, which had been administered orally with the inventive composition containing 0.5% or 1% of α-lipoic acid, shows a lower airway hyperreactivity upon methacholine challenge than that of the asthma control group with no administration.

Particularly, the test group to which α-lipoic acid contained in the inventive composition had been administered intra-abdominally at 64 mg each day, and the test group to which the composition containing 1% of α-lipoic acid had been administered orally, displayed statistically significantly lower airway resistances than that of the asthma control group with no administration. Such lower airway
resistances of the test groups were similar to that of the normal control group.

Accordingly, the inventive composition effectively inhibits bronchial hyperreactivity, indicating that it can be advantageously used for the prevention and treatment of asthma.

Example 2: Anti-inflammatory activity in asthma model mice administered with inventive composition

Anti-inflammatory activity in asthma model mice administered with the inventive composition was examined in the following manner.

1) Bronchoalveolar lavage and construction of lung tissue pieces

Immediately after performing the methacholine challenge test in Example 1, bronchoalveolar lavage was performed as follows. Mice of each group were anesthetized by intra-abdominal administration of 50 mg/ml of ketamine, and then subjected to open-chest and heart puncture procedures, thus collecting about 800 μl or more of blood. Just after exposing their organs, a 24-guage medical catheter was inserted into the organs, and physiological saline solution was slowly injected several times into the organs at 3 ml each time, thus achieving bronchoalveolar lavage.

After performing the bronchoalveolar lavage procedure, lungs were taken out, fixed with formalin, and then embedded into paraffin, thus preparing tissue pieces having a thickness of 3 μm.

2) Pathological analysis

Each tissue piece was stained with hematoxylin-eosin (H&E), and the inflammatory cell number of tissue was observed under an optical microscope to determine lesion score.

As shown in FIG 2A, it could be confirmed that the group administered with the inventive composition containing 25 mg/ml of α-lipoic acid shows a lower lesion
score than that of the asthma control group with no administration.

Particularly, the test group to which α-lipoic acid had been administered intra-abdominally at 64 mg each day displayed a statistically significantly lower lesion score than that of the asthma control group with no administration.

Also, as shown in FIG. 2B, it could be confirmed that the test group administered with the composition containing 0.5% or 1% of α-lipoic acid shows an improvement in inflammation on bronchoalveolar tissue.

3) Serum ovalbumin-specific immunoglobulin analysis

Serum from the mouse blood collected in the part 1) of Example 2 was separated, and then measured for its egg albumin-specific IgE by the ELISA method.

As shown in FIG. 3A, it could be confirmed that the test group administered intra-abdominally with the inventive composition containing 25 mg/ml of α-lipoic acid shows a lower immunoglobulin E level than that of the asthma control group with no administration. Also, as shown in FIG. 3B, it could be found that the group administrated orally with the inventive composition containing α-lipoic acid shows a reduction in ovalbimin specific immunoglobulin E.

Particularly, the test group to which α-lipoic acid contained in the inventive composition had been administered intra-abdominally at 64 mg each day, and the test group to which the composition containing 0.5% or 1% of α-lipoic acid had been administered orally, showed a statistically significantly lower ovalbumin specific immunoglobulin E level than that of the asthma control group with no administration.

4) Cellular analysis on bronchoalveolar lavage fluid

During bronchoalveolar lavage by the method described in the part 1) of Example 2, the lavage fluid was recovered. When the amount of the bronchoalveolar lavage fluid reached 1 ml, it was centrifuged to collect cells. The
cells were subjected to Diff Quick staining, and then observed for alveolar macrophages (AM), lymphocytes (Lym), eosinophils (Eos), polymorphonuclear neutrophils (PMNL) and the like. Also, the ratio of inflammation-related cells, eosinophils, in total blood cells was observed.

As shown in FIG. 4A, it could be found that, in the group administered intra-abdominally with the inventive composition containing 25 mg/ml of α-lipoic acid, the ratio of eosinophils in the lavage fluid was lower than that in the asthma control group with no administration. Furthermore, as shown in FIG. 4B, it could be confirmed that the group administered orally with the inventive composition containing 0.5% or 1.0% of α-lipoic acid shows a significant reduction in the ratio of eosinophils (Eos).

Particularly, the test group to which α-lipoic acid contained in the inventive composition had been administered intra-abdominally at 64 mg each day, and the test group to which the composition containing 0.5% or 1% of α-lipoic acid had been administered orally, exhibited a statistically significantly lower eosinophil level than that of the asthma control group with no administration.

5) Conclusion

The inventive composition removes bronchial allergic inflammation, a typical symptom of asthma, and reduces not only the ratio of eosinophils in bronchoalveolar lavage fluid, an index of bronchial inflammation, but also antigen-specific immunoglobulin E.

Accordingly, the inventive composition effectively inhibits airway inflammation, indicating that it can be advantageously used for the prevention and treatment of asthma.
Industrial Applicability

The inventive composition has the effect of reducing airway hyperreactivity, a typical symptom of asthma.

Also, the inventive composition has the effect of inhibiting inflammation occurring in asthma.

Accordingly, the inventive composition will be useful for the prevention and treatment of asthma.
What Is Claimed Is:

1. A composition for the prevention and treatment of asthma, which contains α-lipoic acid as an active ingredient.

2. The composition of Claim 1, which has bronchial hyperreactivity inhibitory activity.

3. The composition of Claim 2, which has anti-inflammatory activity.

4. The composition of Claim 3, which reduces inflammatory lesions in the airway of asthma patients.

5. The composition of Claim 3, which reduces the ratio of eosinophils in the total blood cells of bronchoalveolar lavage fluid in asthma patients.

6. The composition of Claim 3, which reduces allergen specific immunoglobulin E in asthma patients.

7. Use of α-lipoic acid as an active ingredient in the preparation of a drug for the prevention and treatment of asthma.
[FIG 2]

A.

<table>
<thead>
<tr>
<th>Lesion Score</th>
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- Normal Control
- Asthma
- ALA-1mg
- ALA-4mg
- ALA-16mg
- ALA-64mg

B. Asthma

1% alpha-lipoic acid treated asthma