

DESCRIPTION

KOJI FUNGUS-FERMENTED PRODUCT, FOOD COMPOSITION, COSMETIC COMPOSITION, RAW MATERIAL FOR KOJI FUNGUS-FERMENTED PRODUCT, METHOD FOR PRODUCING KOJI FUNGUS-FERMENTED PRODUCT, AND ENZYME PRODUCTION PROMOTER

TECHNICAL FIELD

[0001]

The present invention relates to a koji fungus-fermented product, a food composition, a cosmetic composition, a raw material for a koji fungus-fermented product, a method for producing a koji fungus-fermented product, and an enzyme production promoter.

BACKGROUND ART

[0002]

Koji is obtained through carrying out a culture by adding a koji fungus which is effective for food fermentation, such as a microorganism belonging to the genus *Aspergillus*, as a seed koji, to a cereal such as rice, soybean, and wheat. Koji has been used in producing fermented food and fermented drink such as miso (Japanese seasoning bean paste), soy sauce, vinegar, pickles, sake, shochu (Japanese spirits), and awamori (Japanese rice brandy), because Koji decomposes a raw material such as a cereal by an action of an enzyme or the like produced within Koji.

[0003]

Koji fungi secrete enzymes such as protease, amylase, and lipase, and these enzymes hydrolyze starch, protein, or lipid contained in a raw material such as a cereal, to cause conversion into a low molecular weight substance, to thus occur

fermentation. Fermented products obtained through a fermentation by a koji fungus are not only improved in taste, but also contain a nutrient in a form that is easy to absorb for a living body, and thus, have attracted attention also from the viewpoint of health.

[0004]

In recent years, active researches have been made on pharmacological actions of fermented foods produced by using koji fungi, and a wide range of pharmacological actions has been reported, such as tyrosine kinase inhibitory activity and inhibitory action against blood pressure rise (Non-Patent Document 1).

[0005]

For example, Patent Document 1 discloses that rice koji produced by using a specific strain of *Aspergillus oryzae* has high lipase and cellulase activities compared with those of conventional rice koji. It is also disclosed that fermented food such as miso and soy sauce produced by using a specific strain of *Aspergillus oryzae* have high antimutagenicity.

[0006]

Meanwhile, euglena (genus name: *Euglena*, Japanese name: Midorimushi) is attracting attention as a biological resource which is highly expected to be used as food, feed, fuel, etc.

Euglena features as much as 59 kinds of nutrients that correspond to most of nutrients necessary for humans to live, such as vitamins, minerals, amino acids, and unsaturated fatty acids, and there have been suggestions for possibility of use thereof as a supplement for well-balanced intake of variety of nutrients, or use thereof as a food supply source in poverty areas where intake of necessary nutrients is not available.

[0007]

It has been said that bulk culture of euglena is difficult,

for reason such that: euglena is located on the most bottom of the food chain, and therefore, preyed by predators; or that euglena needs strict culture conditions such as light or temperature condition, and stirring speed, as compared with those for other microorganisms. However, in recent years, the present inventors established, through earnest research, a technique of bulk culture which opened a way to bulk supply of euglena-derived substances such as euglena and paramylon extracted from euglena.

Euglena is a unique organism that possesses an animal nature of making flagellar movement and at the same time, has chloroplasts as a plant and performs photosynthesis, and euglena itself and substances derived from euglena are expected to have many functions.

[0008]

Patent Document 2 relates to umami (tastiness) food which is effective for prevention and treatment of various immuno-inflammatory disorders or oxidative stress disorders, metabolic syndromes, etc., and discloses that when a yeast containing a polyamine such as spermine and spermidine as an active ingredient is fermented together with hen of the woods (*Grifola frondosa*) and a koji fungus, euglena algal body is added thereto as an auxiliary material.

[0009]

Although Patent Document 2 uses euglena algae as an auxiliary material to carry out a fermentation by a koji fungus, it includes yeast and hen of the woods as essential components. So far, there has been no example of fermenting euglena itself by a koji fungus.

CITATION LIST

PATENT LITERATURE

[0010]

PATENT LITERATURE 1: JP 4049220 B

PATENT LITERATURE 2: WO 2011/081024

NON-PATENT LITERATURE

[0011]

NON-PATENT LITERATURE 1: "Novel Functionality of Koji Fungi",
The Society for Bioscience and Bioengineering, Japan, vol. 93,
no. 8, pp. 491 (2015)

SUMMARY OF INVENTION

TECHNICAL PROBLEM

[0012]

As set forth above, there has been no example of fermenting euglena itself by a koji fungus, and no example of measuring an enzyme titer of a product fermented by a koji fungus with euglena. Nor has there been any example of researching an influence of euglena to a koji fungus and a fermented product, when a euglena is added in a process of fermenting a cereal or the like by a koji fungus. Furthermore, it has been found that a fermentation by a koji fungus is difficult by a normal method, when a euglena is used in a powdered state.

The present invention has been made in view of the above problem, and an object of the present invention is to provide a koji fungus-fermented euglena product, a raw material for a koji fungus-fermented euglena product, a method for producing a koji fungus-fermented euglena product, and an enzyme production promoter.

Another object of the present invention is to provide a food composition and a cosmetic composition containing a koji fungus-fermented euglena product.

Another object of the present invention is to provide a

fermented food obtained by using a koji fungus-fermented euglena product as a seed koji, which will be a novel usage of a koji fungus-fermented euglena product.

SOLUTION TO PROBLEM

[0013]

The present inventors have found, as a result of intensive research, that it is possible to obtain a koji fungus-fermented product having a high enzyme titer, when a mixture of a cereal and a euglena algal body is fermented by using a koji fungus.

Specifically, it has been found that, it is possible to obtain a koji fungus-fermented product having a high enzyme titer, by fermenting a raw material for a koji fungus-fermented product which is obtained by coating a steamed cereal with a mixture of a euglena algal body and a koji fungus.

More particularly, the present inventors clarified that euglena algal body functions as an enzyme production promoter to promote enzyme production of a koji fungus, which has led to the present invention.

[0014]

Therefore, according to the present invention, the problem is solved by a koji fungus-fermented product which is obtained by fermenting a raw material for a koji fungus-fermented product obtained by coating a steamed cereal with a mixture of a euglena algal body and a koji fungus.

In this manner, by coating a steamed cereal with a mixture of a euglena algal body and a koji fungus, it is possible to appropriately ferment a euglena by a koji fungus.

[0015]

In this case, it is preferred that the cereal be a pregelatinized cereal.

In this case, it is preferred that the mixture of a euglena

algal body and a cereal be a steamed cereal coated with the euglena algal body.

In this case, it is preferred that the steamed cereal be at least one kind selected from the group consisting of germinated brown rice, glutinous rice, glutinous brown rice, pearl barley, naked barley, soybean, and rice.

In this case, it is preferred that the mixture of a euglena algal body and a cereal be a granule comprising 20 to 150 parts by mass of a rice powder relative to 100 parts by mass of the euglena algal body.

In this manner, by obtaining a mixture of a euglena powder and a cereal in an appropriate form, it is possible to improve an enzyme titer of a koji fungus-fermented euglena product.
[0016]

In this case, it is preferred that a titer of glucoamylase per gram of the koji fungus-fermented product be 1000 to 2000.

In this case, it is preferred that the euglena algal body be used as an enzyme production promoter for a koji fungus.

By the above structure, a euglena algal body functions as an enzyme production promoter to promote an enzyme production of a koji fungus, and thus, it is possible to provide a koji fungus-fermented product having a high enzyme titer.

In this case, the koji fungus-fermented euglena product may be contained in a food composition or a cosmetic composition.

[0017]

According to the present invention, the problem is solved also by a raw material for a koji fungus-fermented product comprising a koji fungus and a mixture of a euglena algal body and a cereal.

In this manner, it is possible to appropriately ferment a euglena by a koji fungus, by making a mixture of a euglena

algal body with a cereal as a bulking agent.

According to the present invention, the problem is solved also by a raw material for a koji fungus-fermented product obtained by coating a steamed cereal with a mixture of a euglena algal body and a koji fungus.

In this manner, it is possible to appropriately ferment a euglena by a koji fungus, by using a raw material for a koji fungus-fermented product obtained by coating a steamed cereal with a mixture of a euglena algal body and a koji fungus.

[0018]

According to the present invention, the problem is solved by a method for producing a koji fungus-fermented product characterized in carrying out a mixing step for preparing a mixture of a euglena algal body and a cereal, by mixing a euglena algal body with a cereal; a koji fungus addition step for obtaining a raw material for the koji fungus-fermented product by adding a koji fungus to the mixture of a euglena algal body and a cereal; and a fermentation step for fermenting the raw material for the koji fungus-fermented product.

In this case, it is preferred that the mixing step include a step of coating a steamed cereal with the euglena algal body.

In this case, it is preferred that the mixing step include a step of producing a granule comprising 20 to 150 parts by mass of rice powder relative to 100 parts by mass of the euglena algal body.

[0019]

According to the present invention, the problem is solved by a method for producing a koji fungus-fermented product characterized in carrying out: a step of steaming a cereal and mixing a euglena algal body with a koji fungus to prepare a mixture of a euglena algal body and a koji fungus; a mixing step for obtaining a raw material for a koji fungus-fermented product

by mixing the cereal steamed in the previous step with the mixture of a euglena algal body and a koji fungus obtained in the previous step, and; a fermentation step for fermenting the raw material for a koji fungus-fermented product.

According to the present invention, the problem is solved by a method for producing a koji fungus-fermented product characterized in carrying out: a step of steaming a cereal and mixing a euglena algal body with a koji fungus to prepare a mixture of a euglena algal body and a koji fungus; a mixing step for obtaining a raw material for a koji fungus-fermented product by coating the steamed cereal with the mixture of a euglena algal body and a koji fungus obtained in the previous step; and a fermentation step for fermenting the raw material for a koji fungus-fermented product.

According to the present invention, the problem is solved by an enzyme production promoter for a koji fungus, characterized in comprising a euglena algal body as an active ingredient, and in being used to promote an enzyme production of a koji fungus.

In this case, it is preferred that the koji fungus be a yellow koji fungus or a white koji fungus.

In this case, it is preferred that the enzyme be at least one kind selected from the group consisting of a glucoamylase, an α -amylase, and an acid protease.

EFFECTS OF INVENTION

[0020]

According to the present invention, it is possible to provide a koji fungus-fermented product, a raw material for a koji fungus-fermented product, a food composition, a cosmetic composition, a method for producing a koji fungus-fermented product, and an enzyme production promoter.

According to the koji fungus-fermented euglena product of the present invention, it is possible to provide a fermented food by using the koji fungus-fermented euglena product as a seed koji.

In addition, since a koji fungus-fermented product added with a euglena has a higher enzyme titer than that of a koji fungus-fermented product without added with euglena, it is possible to provide an enzyme production promoter for a koji fungus which includes a euglena as an active ingredient.

The koji fungus-fermented euglena product of the present invention can be provided by being included in a food composition or in a cosmetic composition.

BRIEF DESCRIPTION OF DRAWINGS

[0021]

FIG. 1 is a flowchart showing a method for producing a koji fungus-fermented euglena product according to one embodiment of the present invention.

FIG. 2 is a flowchart showing a method for producing a koji fungus-fermented euglena product which uses a steamed cereal, according to one embodiment of the present invention.

FIG. 3 is a schematic view of a device by which a fermentation by a koji fungus of the present invention was carried out.

DESCRIPTION OF EMBODIMENTS

[0022]

Hereinbelow, embodiments of the present invention will be described with reference to FIG. 1 to FIG. 3.

The present embodiment relates to a koji fungus-fermented euglena product, a food composition comprising a koji fungus-fermented euglena product, a cosmetic composition

comprising a koji fungus-fermented euglena product, a raw material for a koji fungus-fermented euglena product, a method for producing a koji fungus-fermented euglena product, and an enzyme production promoter.

[0023] <Euglena>

In the present embodiment, "euglena" includes all of microorganisms categorized in the genus *Euglena* in the categorization of zoology or botany, mutants thereof, and variant species thereof.

Here, the microorganisms of the genus *Euglena* in the zoology refer to microorganisms belonging to the suborder Euglenoidina of the order Euglenida which belongs to the subclass Phytomastigophorea of the class Mastigophorea in the phylum Protozoa. On the other hand, the microorganisms of the genus *Euglena* in the botany belong to the order Euglenales belonging to the class Euglenophyceae in the phylum Euglenophyta.

[0024]

The microorganisms of the genus *Euglena* can specifically listed as follows: *Euglena acus*, *Euglena anabaena*, *Euglena caudata*, *Euglena chadefaudii*, *Euglena deses*, *Euglena gracilis*, *Euglena granulata*, *Euglena intermedia*, *Euglena mutabilis*, *Euglena oxyuris*, *Euglena proxima*, *Euglena spirogyra*, *Euglena viridis*, *Euglena vermiformis*, *Euglena intermedia*, *Euglena piride*, etc.

As euglena cells, it is possible to use those of euglena *gracilis* (*E. gracilis*), particularly, euglena *gracilis* (*E. gracilis*) Z strain, euglena *gracilis* (*E. gracilis*) NIES-49 strain, or the like; or alternatively, a variant strain SM-ZK strain (chloroplast-deficient strain) or a mutant var. *bacillaris* of euglena *gracilis* (*E. gracilis*) Z strain; β -1,3-glucanase, *Euglena intermedia*, *Euglena piride* which are

derived from a gene-modified strain, such as a chloroplast variant strain of these species; or other euglenas such as *Astaia longa*.

[0025]

The genus *Euglena* is widely distributed in fresh water such as ponds and swamps, and is available through a separation therefrom. It is also possible to use any of the genus *euglena* already isolated.

The genus *Euglena* includes all variant strains thereof. The variant strains further include those obtained through a genetic method such as a recombination, a transduction, a transformation, etc.

[0026]

In the culture of *euglena* cells, it is possible to use, as a culture solution, for example, a culture solution added with a nitrogen source, a phosphorus source, or a nutrient salt such as a mineral. Examples therefor include a modified Cramer-Myers medium ($(\text{NH}_4)_2\text{HPO}_4$ 1.0 g/L, KH_2PO_4 1.0 g/L, $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ 0.2 g/l, $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ 0.02 g/l, $\text{Fe}_2(\text{SO}_4)_3 \cdot 7\text{H}_2\text{O}$ 3 mg/l, $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$ 1.8 mg/l, $\text{CoSO}_4 \cdot 7\text{H}_2\text{O}$ 1.5 mg/l, $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ 0.4 mg/l, $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$ 0.2 mg/l, $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ 0.02 g/l, thiamine hydrochloride (vitamin B1) 0.1 mg/l, cyanocobalamin (vitamin B12), (pH 3.5)). Here, $(\text{NH}_4)_2\text{HPO}_4$ can be converted to $(\text{NH}_4)_2\text{SO}_4$ or NH_3aq . Alternatively, it is also possible to use a publicly known Hutner medium or Koren-Hutner medium prepared on the basis of description of *Euglena*, Physiology and Biochemistry (Edited by Shozaburo Kitaoka, Society Publishing Center, Inc.).

[0027]

The culture solution has pH of preferably 2 or more, with the upper limit preferably being 6 or less, more preferably 4.5 or less. By setting the pH to the acidic side, photosynthetic microorganisms can grow more dominantly than other

microorganisms, and thus, it is possible to suppress contamination.

Euglena cells may be cultured by an open pond method that uses sunlight directly, or by a light collecting method that condenses sunlight by a light concentrator and transfers the condensed sunlight through an optical fiber or the like, radiates the transferred light in a culture tank so as to be used in photosynthesis.

Culture of euglena cells can be performed by using, for example, a fed-batch method, and may also be performed by any of liquid culture methods such as flask culture, culture using a fermentation tank, batch culture, semi-batch culture (fed-batch culture), continuous culture (perfusion method), etc.

Separation of Euglena cells is carried out, for example, by a centrifugation, a filtration or a simple sedimentation of the culture solution.

[0028] (Euglena algal body)

In the present embodiment, it is possible to use live euglena cells just as those are separated by a centrifugation, a filtration, a sedimentation, or the like, as the euglena algal body. Although euglena live cells can be used in a state just as those are after harvested from a culture tank, it is preferred that those are washed with water or a physiological saline. It is also possible to use euglena algal body in a state of dispersion liquid, in which euglena algal body is dispersed in a liquid, such as water. In the present embodiment, it is suitable to use a euglena dry algal body which is obtained by subjecting live euglena cells to a freeze-drying or a spray-drying.

It is also possible to use, as the euglena algal body, a mechanically processed product of an algal body which is

obtained by subjecting live euglena cells to a mechanical process such as an ultrasonic wave radiation process and a homogenization process. It is also possible to use, as the euglena algal body, a mechanically processed dry product which is obtained by subjecting the mechanically processed product to a drying process.

[0029] <Raw Material for Koji fungus-Fermented Product of Euglena >

This embodiment carries out a fermentation of a euglena, using, as a raw material, a raw material for a koji fungus-fermented euglena product, which is obtained by adding a koji fungus to a mixture obtained by mixing a euglena algal body and a bulking agent.

[0030] (Cereal as bulking agent)

The bulking agent that can be used in the present embodiment is not particularly limited as long as it is a bulking agent with which a euglena is fermented by a koji fungus. It is possible to use rice in a form of rice powder or steamed rice; brown rice such as germinated brown rice; glutinous rice such as glutinous green rice and glutinous brown rice; wheat such as pearl barley and naked barley; or soybean; etc.

It is preferred to use a rice powder, particularly, a rice powder obtained by pregelatinizing a starch in the rice; or a pregelatinized-rice such as a steamed rice.

As used herein, pregelatinization refers to gelatinization of a starch contained in a cereal such as rice by a moisture and a heat, and is synonymous with gelatinization.

[0031]

Mixing ratio of the bulking agent to the euglena algal body is only required to be within 1 to 150 parts by mass, preferably 1 to 130 parts by mass, more preferably 1 to 120 parts by mass, and particularly preferably 1 to 110 parts by mass,

relative to 100 parts by mass of the euglena algal body.

[0032] (Mixture of euglena algal body and cereal)

Form of the mixture of a euglena algal body and a cereal in the present embodiment is not particularly limited as long as it is a form in which a euglena is fermented by a koji fungus, and for example, it is possible to use a granule of a euglena algal body and a cereal powder, or a steamed cereal coated with a euglena algal body obtained by dredging a euglena algal body over a steamed cereal.

[0033] (Koji fungus)

Koji fungus in the present embodiment may be any koji fungus as long as it is a koji fungus capable of producing an effective enzyme and capable of fermenting a euglena, which can be listed as black koji fungi, white koji fungi, yellow koji fungi, and red koji fungi, and examples thereof include microorganisms belonging to the genus *Aspergillus* or the genus *Monascus*, but not limited thereto.

[0034]

Koji fungi belonging to the genus *Aspergillus* can be listed as follows: *Aspergillus oryzae*, *Aspergillus niger*, *Aspergillus sojae*, *Aspergillus awamori*, *Aspergillus kawachii*, *Aspergillus luchensis*, etc.

Koji fungi belonging to *Aspergillus oryzae* include yellow koji fungi and soy sauce koji fungi. Koji fungi belonging to *Aspergillus niger* include black koji fungi and white koji fungi. Examples of koji fungi belonging to the genus *Monascus* include red koji fungi.

[0035]

In the present embodiment, used as the koji fungus to ferment a euglena is preferably a koji fungus belonging to *Aspergillus oryzae*, *Aspergillus niger*, or *Aspergillus kawachii*, more preferably a koji fungus belonging to *Aspergillus oryzae*,

and particularly preferably a yellow koji fungus, but not limited thereto. Among yellow koji fungi, it is preferred to use AOK139 strain of *Aspergillus oryzae* (FERM AP-20562), or AOK138 strain of *Aspergillus oryzae* (FERM AP-20550).

[0036]

Form of the koji fungus to be used may be, for example, a dispersion of seeds koji in a sterilized water, but is not limited thereto.

Mixing of seeds koji to a mixture of a euglena algal body and a cereal is carried out, for example, by spraying seeds koji on a mixture of a euglena algal body and a cereal. In a case of a bulk production, a spreader can be used.

[0037] (Moisture content)

A moisture content at the time of fermentation of the mixture of a euglena algal body and a cereal in the present embodiment is not particularly limited as long as it is a moisture content at which a euglena is fermented by a koji fungus, and is only required to be within 20 to 80 parts by mass, preferably 30 to 75 parts by mass, and more preferably 35 to 70 parts by mass, when total weight of the mixture of a euglena algal body and a cereal is 100 parts by mass.

[0038] (Other components)

The raw material for a koji fungus-fermented euglena product of the present embodiment can contain a component other than the mixture of a euglena algal body and a cereal, the koji fungus, and the moisture, as long as it does not interfere with a fermentation of a euglena by a koji fungus.

Content of other components contained in the raw material for a koji fungus-fermented euglena product is 10 parts by mass or less, preferably 5 parts by mass or less, more preferably 2 parts by mass or less, relative to 100 parts by mass of the raw material for a koji fungus-fermented euglena product.

[0039] <Method for Producing Koji fungus-Fermented Product of Euglena>

Method for producing a koji fungus-fermented euglena product of the present embodiment is characterized in carrying out a mixing step for preparing a mixture of a euglena algal body and a cereal by mixing a euglena algal body with a cereal; a koji fungus addition step for obtaining a raw material for the koji fungus-fermented product by adding a koji fungus to the mixture of a euglena algal body and a cereal; and a fermentation step for fermenting the raw material for the koji fungus-fermented product.

Hereinbelow, each of the steps are described in detail, with reference to FIG. 1.

[0040] (Mixing step)

In the mixing step, a euglena algal body and a cereal are mixed to each other (Step S1). Specifically, a cereal as a bulking agent is mixed with a euglena algal body to prepare a mixture of a euglena algal body and a cereal.

Although a mixing device may be used in the mixing step, it is preferred to carry out a mixing by hand.

[0041] (Koji fungus addition step)

In the koji fungus addition step, the mixture of a euglena algal body and a cereal obtained in the mixing step is added with a koji fungus (Step S2). As the koji fungus, a seed koji comprising the koji fungus is used. The addition of seed koji is carried out, for example, by spraying seed koji on the mixture of a euglena algal body and a cereal. In a case of a bulk production, a spreader can be used. Incidentally, in the mixing step, in a case where the mixing is carried out by using a sterilized water comprising a seed koji, the mixing step and the koji fungus addition step shall be carried out simultaneously.

In the koji fungus addition step, although a mixing device may be used in stirring, it is preferred to carry out a mixing by hand.

[0042] (Fermentation step)

In the fermentation step, the raw material for a koji fungus-fermented euglena product obtained in the koji fungus addition step is fermented under a predetermined fermentation condition (Step S3).

[0043]

According to the method for producing a koji fungus-fermented euglena product according to the present embodiment, a rice as a bulking agent is mixed with a euglena algal body, to make a fermentation by a koji fungus be carried out appropriately.

[0044] <Method for Producing Koji Fungus-Fermented Product of Euglena Using Steamed Cereal>

The method for producing a koji fungus-fermented euglena product of the present embodiment is characterized in carrying out: a step of steaming a cereal and mixing a euglena algal body with a koji fungus to prepare a mixture of a euglena algal body and a koji fungus; a mixing step for obtaining a raw material for a koji fungus-fermented product by mixing the cereal steamed in the previous step with the mixture of a euglena algal body and a koji fungus obtained in the previous step, and a fermentation step for fermenting the raw material for the koji fungus-fermented product.

Hereinbelow, each of the steps are described in detail, with reference to FIG. 2.

[0045]

In the step of steaming a cereal and mixing a euglena algal body with a koji fungus to prepare a mixture of a euglena algal body and a koji fungus, a step of steaming a cereal (Step S11)

and a first mixing step (Step S12) are carried out.

[0046] (Step of steaming cereal)

In the step of steaming a cereal, a cereal is steamed (Step S11). Specifically, a starch contained in a cereal is pregelatinized, for example, by steaming a cereal such as a rice in a form such as a rice powder and a steamed rice, a brown rice such as a germinated brown rice, a glutinous rice such as a glutinous green rice and a glutinous brown rice, a wheat such as a pearl barley and a naked barley, or a soybean, with an addition of water.

[0047] (First mixing step)

In the first mixing step, a euglena algal body and a koji fungus are mixed with each other to prepare a mixture of a euglena algal body and a koji fungus (Step S12). Specifically, a euglena algal body is mixed with a seed koji comprising a predetermined koji fungus, to prepare a mixture of the euglena algal body and the koji fungus. The mixing is carried out, for example, by spraying the seed koji on the euglena algal body. In a case of a bulk production, it is also possible to use a spreader. Although a mixing device may be used in the first mixing step, it is preferred to carry out a mixing by hand.

[0048]

In the step of steaming a cereal and mixing a euglena algal body with a koji fungus to prepare a mixture of a euglena algal body and a koji fungus, an order of carrying out the step of steaming a cereal (Step S11) and the first mixing step (Step S12) is not limited to carrying out the first mixing step (Step S12) after carrying out the step of steaming a cereal (Step S11). For example, it is also possible to carry out the step of steaming a cereal (Step S11), after carrying out the first mixing step (Step S12). It is also possible to simultaneously carry out the step of steaming a cereal (Step S11) and the first mixing

step (Step S12), in parallel.

[0049] (Second mixing step)

In the second mixing step, a raw material for the koji fungus-fermented product is obtained by mixing the cereal steamed in the step of steaming a cereal with the mixture of a euglena algal body and a koji fungus obtained in the first mixing step (Step S13). Specifically, a raw material for the koji fungus-fermented product is obtained by carrying out the mixing in a manner of dredging the mixture of a euglena algal body and a koji fungus over the steamed cereal. At this time, it is also possible to carry out a step of removing the euglena algal body which is not adhered to the steamed cereal.

[0050] (Fermentation step)

In the fermentation step, the raw material for the koji fungus-fermented euglena product obtained in the second mixing step is fermented under a predetermined fermentation condition (Step S14).

[0051]

The method for producing the koji fungus-fermented euglena product according to the present embodiment makes a fermentation by the koji fungus be carried out appropriately, by mixing a euglena algal body with a koji fungus in advance, and then mixing a steamed rice as a bulking agent thereto.

[0052] <Condition for Fermenting Euglena by Koji Fungus>

In the fermentation step in the method for producing the koji fungus-fermented euglena product of the present embodiment, a fermentation of a euglena by a koji fungus is carried out under a predetermined fermentation condition which appropriately selects conditions such as temperature, humidity, pH, duration, style of fermentation, and fermentation device.

Hereinbelow, each of the fermentation conditions are described in detail.

[0053] (Temperature for fermentation)

Temperature for fermentation in the present embodiment is not particularly limited as long as it is a temperature at which a euglena is fermented by a koji fungus, and is only required to be within 0 to 70°C, preferably 10 to 60°C, more preferably 20 to 50°C, and even more preferably 25 to 40°C.

[0054] (Humidity for fermentation)

Humidity for fermentation in the present embodiment is not particularly limited as long as it is a humidity at which a euglena is fermented by a koji fungus, and is only required to be a relative humidity of 40 to 100% RH at fermenting temperature of 10 to 60°C.

[0055] (Fermentation pH)

Although pH for fermentation in the present embodiment varies depending on type of koji fungus used, it is not particularly limited as long as it is a pH at which a euglena is fermented sufficiently by a koji fungus, and is only required to be within pH 4 to 7.

[0056] (Duration of fermentation)

Duration of fermentation in the present embodiment is not particularly limited as long as it is a duration for which a euglena is fermented sufficiently by a koji fungus, and is only required to be 24 to 72 hours.

[0057] (Style of fermentation, device for fermentation)

Examples of a vessel which can be used in euglena fermentation of the present embodiment by a koji fungus include lidded containers made of glass, metal, plastic, etc. It is also possible to use a container capable of evaporating moisture, such as an open container or a wooden container, as long as it can maintain a moisture content within a predetermined range. In a case of a bulk production, it is possible to use a koji production apparatus, such as a rotary automatic koji

production and culture apparatus.

[0058]

Each of the conditions for fermentation may be constant throughout a duration, and it is also possible to change each of the fermentation conditions depending on duration of a fermentation so as to promote a fermentation, in order to improve an enzyme titer.

[0059] <Koji Fungus-Fermented Product of Euglena>

The koji fungus-fermented euglena product of the present embodiment has a characteristic of having a high enzyme titer.

Here, the koji fungus-fermented euglena product comprises not only a fermented product itself which is obtained by fermenting a raw material for a koji fungus-fermented euglena product by koji fungus, but also a processed product of the fermented product obtained by appropriately processing the fermented product, which includes a solid such as a powder, a dried product, and a dried extract, and a liquid such as a liquid extract, a liquid dispersion, a squeezed juice, and a liquid filtrate.

[0060] (Enzyme titer)

The koji fungus-fermented euglena product of the present embodiment exhibits an enzyme activity according to a koji fungus used. The enzymes can be listed as follows: a protease, an amylase (an α -amylase, a β -amylase, a glucoamylase, an isoamylase, etc.), lipase, cellulase, peptidase, etc., but not limited thereto.

[0061]

The koji fungus-fermented euglena product of the present embodiment is characterized in having a high enzyme titer measured according to an analysis method prescribed by the National Tax Agency JAPAN.

As used herein, having a high enzyme titer refers to, for

example, exhibiting an enzyme titer as shown below.

[0062]

The koji fungus-fermented euglena product of the present embodiment has a glucoamylase titer measured according to an analysis method prescribed by the National Tax Agency JAPAN of 200 U/g to 2000 U/g, preferably 250 U/g to 2000 U/g, more preferably 300 U/g to 2000 U/g.

The koji fungus-fermented euglena product of the present embodiment has an α -amylase titer measured according to an analysis method prescribed by the National Tax Agency JAPAN of 1000 U/g to 5000 U/g, preferably 2000 U/g to 5000 U/g, more preferably 4000 U/g to 5000 U/g.

The koji fungus-fermented euglena product of the present embodiment has an acid protease titer measured according to an analysis method prescribed by the National Tax Agency JAPAN of 4000 U/g to 60000 U/g, preferably 5000 U/g to 60000 U/g, more preferably 10000 U/g to 60000 U/g.

The koji fungus-fermented euglena product of the present embodiment has an acid carboxypeptidase titer measured according to an analysis method prescribed by the National Tax Agency JAPAN of 4000 U/g to 60000 U/g, preferably 5000 U/g to 60000 U/g, more preferably 20000 U/g to 60000 U/g.

As used herein, the unit U/g of enzyme titer refers to a unit representing an enzyme titer per gram of a koji fungus-fermented euglena product. A value of the enzyme titer is a dry matter equivalent which is obtained by converting a measured value of a raw koji in a state of containing moisture into a value when no moisture is contained, or a moisture content is 0%. Moisture content of koji can be measured by using a decompression drying method or a heat drying method prescribed by the National Tax Agency JAPAN.

[0063] <Enzyme Production Promoter for Koji Fungus>

The koji fungus-fermented product of the present embodiment is obtained by carrying out a fermentation using, as a raw material, a raw material for a koji fungus-fermented product which is obtained by adding a koji fungus to a mixture obtained by mixing a rice as a bulking agent to a euglena algal body. Here, the koji fungus-fermented product of the present embodiment exhibits a high enzyme titer as compared with that of a koji fungus-fermented product without added with a euglena algal body. Thus, a euglena algal body has an action of promoting enzyme production of a koji fungus, and is capable of being used as an enzyme production promoter for a koji fungus.

[0064]

Specifically, since the euglena algal body promotes production of a glucoamylase, an α -amylase, an acid protease, and an acid carboxypeptidase of a koji fungus (a yellow koji fungus) which belongs to *Aspergillus oryzae*, it is possible to use the euglena algal body as a glucoamylase production promoter, an α -amylase production promoter, an acid protease production promoter, and an acid carboxypeptidase production promoter for a koji fungus (a yellow koji fungus) which belongs to *Aspergillus oryzae*.

[0065]

Since the euglena algal body promotes production of a glucoamylase, an α -amylase, an acid protease, and an acid carboxypeptidase of a white koji fungus, it is possible to use the euglena algal body as a glucoamylase production promoter, an α -amylase production promoter, an acid protease production promoter, and an acid carboxypeptidase production promoter for a white koji fungus.

[0066] <Food Composition, Cosmetic Composition>

The koji fungus-fermented euglena product of the present embodiment is capable of being used as a composition such as

a food composition, a cosmetic composition, etc.

[0067] (Food composition)

The koji fungus-fermented euglena product according to the present embodiment, and a processed product of the fermented product may be used in any food composition such as, for example, beverage (soft drink, alcoholic beverage, carbonated beverage, milk beverage, fruit juice beverage, tea, coffee, nutritional drink, etc.), seasoning such as soy sauce, soups, creams, various dairy products, processed meat product, processed agricultural product, frozen confectionery such as ice cream, various powdered foods (including beverage), concentrated beverage, food for preservation, frozen food, breads, cereal foods, and confectioneries (candy (throat lozenge), cookie, biscuit, gum, gummi, chocolate, etc.).

[0068]

Alternatively, it is also possible to use the koji fungus-fermented euglena product according to the present embodiment and a processed product thereof in Foods with Health Claims (Foods for Specified Health Function Claims, Foods with Nutrient Function Claims, and Foods with Function Claims), so-called health foods (including beverages), concentrated nutritional supplements, liquid foods, or baby/infant foods.

Here, the Foods for Specified Health Uses are foods comprising a health functional component that influences a biological function, etc., which can display itself as suited to a specific health usage, under a permission of commissioner of the Consumer Affairs Agency.

The Foods with Nutrient Function Claims are foods which are used to supplement a nutrient component (vitamin or mineral), and display a function of the nutrient component. In order to sell as Foods with Nutrient Function Claims, it is necessary that a nutrient component contained in a standard dairy intake

thereof needs be within a range between a predetermined upper limit and lower limit, and it is also necessary to display not only a nutrient function, but also a warning notice, or the like.

The Foods with Function Claims are foods which display its functionality based on a scientific ground, under a responsibility of a trader thereof, with information about grounds for safety and functionality, etc. thereof having been reported to the Commissioner of the Consumer Affairs Agency, before sales.

[0069]

It is possible to freely select one kind or two or more kinds of components which can usually be used in a food composition, and blend them in the food composition according to the present embodiment, in addition to the koji fungus-fermented euglena product and a processed product of the fermented product. For example, it is possible to include any additive which can usually be used in the field of food, such as every kind of seasonings, preservatives, emulsifiers, stabilizers, spices, colorants, antiseptics, pH adjusters, etc.

[0070]

Content of the koji fungus-fermented euglena product and a processed product of the fermented product in a food composition is not particularly limited, and can freely be set according to a purpose.

[0071] (Cosmetic composition)

The koji fungus-fermented euglena product and the processed product of the fermented product according to the present embodiment may suitably be used in a cosmetic composition.

The cosmetic composition is capable of being applied to any form of cosmetic agents. For example, it may be applied

to a skin care cosmetics such as a lotion, a milky lotion, a cream, and a conditioning essence; a makeup cosmetics such as a foundation, a concealer, a makeup base, a lipstick, a blusher, an eye shadow, and an eyeliner, or sunscreen cosmetics, etc.
[0072]

It is possible to freely select one kind or two or more kinds of components which can usually be used in a cosmetic composition and blend them in the cosmetic composition according to the present embodiment, in addition to the koji fungus-fermented euglena product and a processed product of the fermented product of the present embodiment.

For example, it is possible to include any additive which can usually be used in the field of cosmetics, such as, for example, a base material, a preservative, an emulsifier, a colorant, an antiseptic, a surfactant, a UV absorber, an antioxidant, a moisturizer, a UV absorber, a spice, an antiseptic and antifungal agent, an extender, a coloring pigment, an alcohol, water, and the like.

[0073]

In the cosmetic composition according to the present embodiment, content of the koji fungus-fermented euglena product and the processed product of the fermented product according to the present embodiment is not particularly limited, and can freely be set according to a purpose.

[0074] <Fermented food>

The koji fungus-fermented euglena product of the present embodiment is capable of producing a fermented food by being used as a seed koji to ferment a raw material such as beans and cereals.

[0075]

Examples of the fermented food obtained by using the koji fungus-fermented euglena product of the present embodiment as

a seed koji may include miso, soy sauce, sake, vinegar, cheese, yogurt, etc.

[0076]

Raw material to be fermented by using koji fungus-fermented euglena product is not particularly limited as long as it is a material used as a food material, and examples therefor include beans such as soybeans, red beans, broad beans, peas, etc., cereals such as rice, wheat, foxtail millet, Japanese millet, corn, etc., tubers and roots such as sweet potato, potato, etc., root vegetables such as daikon (long white radish), carrot, spinach, etc., and animal-derived raw materials such as milk, fish meat, animal meat, etc.

[0077]

It is possible to appropriately add a general food additive to a fermented food obtained by using a koji fungus-fermented euglena product as a koji fungus. Such additives can be listed as follows: antiseptics, antioxidants, chelating agents, pH adjusters, amino acids, inorganic salts, thickeners, surfactants, spices, colorants, moisture, fats and oils, alcohols, sugars, vitamins, proteins, starches, etc.

[Example]

[0078]

Hereinbelow, the present invention is described specifically, on the basis of concrete Examples, however, the present invention is not limited thereto.

The following Examples carried out fermentation by a koji fungus, using varied forms of mixtures of a rice and a euglena algal body, to study method of fermenting a euglena which increases an enzyme titer of an obtained koji fungus-fermented product.

[0079] <Preparation of Mixtures of Euglena Algal Body and Rice>

A mixture of a euglena algal body and a rice was prepared by using a euglena gracilis powder from Euglena Co., Ltd. as the euglena algal body, and Aspergillus oryzae AOK 139 strain (FERM AP-20562) from AKITA KONNO CO., LTD. as the seed koji. [0080]

Test samples for Example 1, Example 2, Comparative Example 1 and Comparative Example 2 were prepared in the forms and the compositions as shown in Table 1.

[0081] [Table 1]

TEST SAMPLE	FORM	EUGLENA POWDER	BULKING AGENT	STEAMED RICE	AMOUNT OF WATER ADDITION	AMOUNT OF SEED KOJI
COMPARATIVE EXAMPLE 1	POWDER	100g	—	—	60ml	0.035g
COMPARATIVE EXAMPLE 2	GRANULE	100g	—	—	60ml	0.035g
EXAMPLE 1	RICE POWDER GRANULE	70g	30g	—	60ml	0.035g
EXAMPLE 2	COATED ON STEAMED RICE	100g	—	100g	—	0.035g

[0082] (Comparative Example 1)

To 100 g of euglena powder, 60 ml of sterilized water added with 0.035 g of seed koji was sprayed, and the resultant was stirred without kneading, to obtain a euglena powder to which a seed koji is mixed.

[0083] (Comparative Example 2)

To 100 g of euglena powder, 60 ml of sterilized water added with 0.035 g of seed koji was sprayed, and the resultant was kneaded and formed into a dumpling shape, which is then passed through a perforated mesh (5 mm diameter) to obtain a granule of euglena powder to which a seed koji is mixed.

[0084] (Example 1)

A euglena powder 70 g and a rice powder (a raw general rice powder) 30 g as a bulking agent are mixed to each other and stirred, to which a sterilized water 60 ml added with a seed

koji 0.035 g was sprayed, and the resultant was kneaded and formed into a dumpling shape, and passed through a perforated mesh (5 mm diameter), to obtain a rice powder granule of euglena powder to which a seed koji is mixed.

[0085] (Example 2)

Used as a steamed rice was a rice immersed in water which is then drained away therefrom, and steamed for 1 hour and 50 minutes. The obtained steamed rice was dredged and coated with a mixture 0.035 g of a euglena powder and a seed koji mixed to each other and homogenized in advance, and the euglena powder and seed koji which was not adhered to the steamed rice (about 99 g) was removed, to thereby obtain a steamed rice coated with euglena powder.

[0086] <Fermentation of Euglena Algal Body and Rice>

A fermentation by a koji fungus of each of the test samples was carried out using the device shown in FIG. 3, under conditions of temperature of 28 to 30°C and humidity of 75-95% RH.

A petri dish 3 with a lid, in which prepared each test sample was placed, was prepared on a slat 2 in a stainless steel vat 1 with a lid. After 24 hours from culture initiation, the lids of the stainless steel vat 1 and the petri dishes 3 were removed, the contents were lightly stirred, and a filter papers 4 were placed on the petri dishes and the stainless steel vat. After 45 hours from culture initiation, the koji fungus-fermented products were taken out.

[0087] <Test 1: Enzyme Titer Measurement of Koji Fungus-Fermented Euglena Product>

Enzyme titer of the koji fungus-fermented euglena products of Example 1, Example 2, Comparative Example 1 and Comparative Example 2 was measured according to an analysis method prescribed by the National Tax Agency JAPAN. As

Comparative Example 3, enzyme titer of an unprocessed euglena powder was measured.

Results are shown in Table 2.

[0088] [Table 2]

TEST SAMPLE	GLUCOAMYLASE		α -AMYLASE		ACID PROTEASE (pH=3.0)		MOISTURE CONTENT (wt%)
	MEASURED VALUE	DRY MATER BASIS	MEASURED VALUE	DRY MATER BASIS	MEASURED VALUE	DRY MATER BASIS	
COMPARATIVE EXAMPLE 1	46	70	1187	1807	205	313	34.3
COMPARATIVE EXAMPLE 2	38	57	1162	1723	257	381	32.6
EXAMPLE 1	262	391	3217	4794	2234	3330	32.9
EXAMPLE 2	1130	1633	3296	4763	36722	53067	30.8
COMPARATIVE EXAMPLE 3	0	0	UNMEASURABLE		-180	-190	5.4

UNIT OF ENZYME TITER: U/g KOJI

[0089]

The numerical values in Table 2 represent enzyme titer per gram of the koji fungus-fermented euglena products in U/g, by an enzyme titer measurement.

For the glucoamylase activity, 1 unit (U) thereof was defined as an activity for producing 1 mg of glucose from a soluble starch in 60 minutes at 40°C.

The α -amylase activity was represented by an amount of 1% solubilized starch (ml) degraded in 30 minutes at 40°C.

For the acid protease activity, 1 unit thereof was defined as an amount of the enzyme that produces 1 μ g of tyrosine equivalent in 60 minutes at 40°C.

[0090]

The moisture content was obtained according to an analysis method prescribed by the National Tax Agency JAPAN (decompression drying method) by the following formula:

$$\text{Moisture (wt\%)} = \{(a - b)/a\} \times 100$$

,wherein "a" represents a specimen weight before drying

and "b" represents a specimen weight after drying, after about 10 g of specimen was accurately weighed out in a weighing device with a lid, and then dried at 70 to 75°C under a pressure not exceeding 50 mmHg for 1 hour, and weighed after cooled in a desiccator.

The dry matter basis represents a value obtained by converting a measured value of a raw koji in a state of containing moisture into a value when the moisture content is 0%.

[0091] (Results of Test 1)

In Comparative Example 1 and Comparative Example 2, where the euglena powder was not mixed with rice powder, titer of each enzyme was low.

On the other hand, In Example 1 where the euglena powder was mixed with a rice powder as a bulking agent at a mass ratio of 70:30, and the thus obtained rice powder granule was fermented, each enzyme had a large titer as compared with those of Comparative Examples. Values on dry matter basis of glucoamylase, α -amylase and acid protease relative to Comparative Example 1 were 5.6 folds, 2.7 folds, and 10.6 folds, respectively.

[0092]

In Example 2, where the steamed rice coated with the euglena powder was fermented, each enzyme had a large titer as compared with those of Comparative Examples. Values on dry matter basis relative to Comparative Example 1 of glucoamylase, α -amylase, and acid protease were 23.3 folds, 2.6 folds, and 170 folds, respectively.

In particular, titers of glucoamylase and acid protease were found to be increased.

[0093]

From the above results, it has been found that a koji fungus-fermented product having a high titer of each enzyme is

obtained when a mixture obtained by mixing a euglena powder with a rice powder as a bulking agent is fermented by a koji fungus.

As for the unprocessed euglena powder of Comparative Example 3, no activity was observed in each of the enzymes, which suggested that an activity of each enzyme is generated by fermenting a euglena.

[0094]

The following Examples used mixtures of a euglena algal body and various cereals to carry out fermentation by a koji fungus (a yellow koji fungus or a white koji fungus), and measured enzyme titers of the obtained koji fungus-fermented products.

[0095] <Preparation of Mixtures of Euglena Algal Body and Cereals>

Euglena gracilis powder from Euglena Co., Ltd. was used as the euglena algal body, and a yellow koji (*Aspergillus oryzae* AOK139 strain, FERM AP-20562 from AKITA KONNO CO., LTD.) or a white koji (white koji Shirakami from AKITA KONNO CO., LTD.) was used as the seed koji, to prepare mixtures of a euglena algal body and a cereal.

[0096]

The test samples of Examples 3 to 9, and Comparative Examples 3 to 9 were prepared with the cereals and at the compositions as shown in Table 3, using the yellow koji.

[0097] [Table 3]

TEST SAMPLE	CEREAL	EUGLENA POWDER	STEAMED RICE	AMOUNT OF SEED KOJI (YELLOW KOJI)
COMPARATIVE EXAMPLE 3	GERMINATED BLUE BROWN RICE FOR PROFESSIONAL USE	—	100g	0.035g
COMPARATIVE EXAMPLE 4	GLUTINOUS GREEN RICE	—	100g	0.035g
COMPARATIVE EXAMPLE 5	GLUTINOUS BROWN RICE	—	100g	0.035g
COMPARATIVE EXAMPLE 6	PEARL BARLEY	—	100g	0.035g
COMPARATIVE EXAMPLE 7	NAKED BARLEY	—	100g	0.035g
COMPARATIVE EXAMPLE 8	SOYBEAN	—	100g	0.035g
COMPARATIVE EXAMPLE 9	RICE FOR STAPLE FOOD	—	100g	0.035g
EXAMPLE 3	GERMINATED BLUE BROWN RICE FOR PROFESSIONAL USE	100g	100g	0.035g
EXAMPLE 4	GLUTINOUS GREEN RICE	100g	100g	0.035g
EXAMPLE 5	GLUTINOUS BROWN RICE	100g	100g	0.035g
EXAMPLE 6	PEARL BARLEY	100g	100g	0.035g
EXAMPLE 7	NAKED BARLEY	100g	100g	0.035g
EXAMPLE 8	SOYBEAN	100g	100g	0.035g
EXAMPLE 9	RICE FOR STAPLE FOOD	100g	100g	0.035g

[0098]

The test samples of Examples 10 to 16, and Comparative Examples 10 to 16 were prepared with the cereals and at the compositions as shown in Table 4, using the white koji.

[0099] [Table 4]

TEST SAMPLE	CEREAL	EUGLENA POWDER	STEAMED RICE	AMOUNT OF SEED KOJI (WHITE KOJI)
COMPARATIVE EXAMPLE 10	GERMINATED BLUE BROWN RICE FOR PROFESSIONAL USE	—	100g	0.035g
COMPARATIVE EXAMPLE 11	GLUTINOUS GREEN RICE	—	100g	0.035g
COMPARATIVE EXAMPLE 12	GLUTINOUS BROWN RICE	—	100g	0.035g
COMPARATIVE EXAMPLE 13	PEARL BARLEY	—	100g	0.035g
COMPARATIVE EXAMPLE 14	NAKED BARLEY	—	100g	0.035g
COMPARATIVE EXAMPLE 15	SOYBEAN	—	100g	0.035g
COMPARATIVE EXAMPLE 16	RICE FOR STAPLE FOOD	—	100g	0.035g
EXAMPLE 10	GERMINATED BLUE BROWN RICE FOR PROFESSIONAL USE	100g	100g	0.035g
EXAMPLE 11	GLUTINOUS GREEN RICE	100g	100g	0.035g
EXAMPLE 12	GLUTINOUS BROWN RICE	100g	100g	0.035g
EXAMPLE 13	PEARL BARLEY	100g	100g	0.035g
EXAMPLE 14	NAKED BARLEY	100g	100g	0.035g
EXAMPLE 15	SOYBEAN	100g	100g	0.035g
EXAMPLE 16	RICE FOR STAPLE FOOD	100g	100g	0.035g

[0100] (Comparative Examples 3 to 16)

Each 100 g of the steamed cereal was dredged and coated with a seed koji 0.035 g.

(Examples 3 to 16)

Each 100 g of the steamed cereal was dredged and coated with a mixture of euglena powder 100 g and a seed koji 0.035 g which were mixed to each other and homogenized in advance, and the euglena powder and the seed koji which were not adhered

to the steamed cereals (about 99 g) were removed, to thereby obtain cereals coated with euglena powder.

Next, a fermentation by a koji fungus of each of the test samples was carried out using the device shown in FIG. 3, under a condition of temperature of 28 to 30°C and humidity of 75 to 95% RH. After 24 hours from culture initiation, the lids of the stainless steel vat and the petri dishes were removed, the contents were lightly stirred, and a filter paper was placed on the petri dishes and the stainless steel vat. After 45 hours from culture initiation, the koji fungus-fermented products were taken out.

[0101] <Test 2: Enzyme Titer Measurement of Koji Fungus-Fermented Product of Each of Cereals and Euglena>

Enzyme titers of the koji fungus-fermented products of Examples 3 to 16 and Comparative Examples 3 to 16 were measured according to an analysis method prescribed by the National Tax Agency JAPAN.

Results of using the yellow koji are shown in Table 5 (Comparative Examples 3 to 9) and Table 6 (Examples 3 to 9).

[0102] [Table 5]

TEST SAMPLE	GLUCOAMYLASE		α -AMYLASE		ACID PROTEASE (pH=3.0)		ACID CARBOXYPEPTIDASE		MOISTURE CONTENT (wt%)	
	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI
COMPARATIVE EXAMPLE 3	228	244	1553	1717	14461	14395	34415	32561	40.7	7.6
COMPARATIVE EXAMPLE 4	71	81	821	911	7914	7115	17727	15668	40.3	6.9
COMPARATIVE EXAMPLE 5	134	180	1104	1412	11283	11642	20761	23552	38.1	6.9
COMPARATIVE EXAMPLE 6	1106	1396	3827	4612	25486	37971	48425	51032	38.3	6.7
COMPARATIVE EXAMPLE 7	460	532	2219	2712	23691	27972	52700	49840	41.9	7.1
COMPARATIVE EXAMPLE 8	40	35	1973	1809	369	164	1990	2248	58.2	5.9
COMPARATIVE EXAMPLE 9	1142	1259	4339	3973	14518	13361	33282	39012	35.3	6.2

UNIT OF ENZYME TITER: U/g KOJI

[0103] [Table 6]

TEST SAMPLE	GLUCOAMYLASE		α -AMYLASE		ACID PROTEASE (pH=3.0)		ACID CARBOXYPEPTIDASE		MOISTURE CONTENT (wt%)	
	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI
EXAMPLE 3	950	767	3804	5132	23582	25703	41227	32013	34.4	6.5
EXAMPLE 4	577	619	2291	2835	21198	24380	34486	33794	37.2	6.0
EXAMPLE 5	699	787	2222	3205	22526	31968	39658	41275	36.6	5.9
EXAMPLE 6	777	1008	3300	4108	27940	35335	36595	36921	34.9	5.8
EXAMPLE 7	1025	1208	3856	5636	28473	30986	36141	29550	37.8	5.9
EXAMPLE 8	89	85	3201	3398	519	272	1206	1920	53.0	5.5
EXAMPLE 9	1152	1297	4093	4646	24225	32589	30618	27204	30.2	6.1

UNIT OF ENZYME TITER: U/g KOJI

[0104]

Results of using the white koji are shown in Table 7

(Comparative Examples 10 to 16) and Table 8 (Examples 10 to 16) .

[0105] [Table 7]

TEST SAMPLE	GLUCOAMYLASE		α -AMYLASE		ACID PROTEASE (pH=3.0)		ACID CARBOXYPEPTIDASE		MOISTURE CONTENT (wt%)	
	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI
COMPARATIVE EXAMPLE 10	36	43	828	875	9021	8180	24354	27202	41.6	5.5
COMPARATIVE EXAMPLE 11	14	7	433	448	4420	3905	8994	8561	40.7	6.0
COMPARATIVE EXAMPLE 12	24	20	521	503	6304	5978	13016	15198	39.3	5.5
COMPARATIVE EXAMPLE 13	114	166	1395	1689	15106	18489	38081	38309	39.6	5.3
COMPARATIVE EXAMPLE 14	59	92	965	1355	11738	9542	32430	37190	43.1	5.3
COMPARATIVE EXAMPLE 15	31	14	2165	887	123	69	1000	970	58.4	6.4
COMPARATIVE EXAMPLE 16	252	269	1936	1927	8649	8120	18179	21196	34.1	5.1

UNIT OF ENZYME TITER: U/g KOJI

TEST SAMPLE	GLUCOAMYLASE		α -AMYLASE		ACID PROTEASE (pH=3.0)		ACID CARBOXYPEPTIDASE		MOISTURE CONTENT (wt%)	
	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI	RAW KOJI	DRY KOJI
EXAMPLE 10	199	195	1926	3803	13160	15951	27578	22841	32.3	5.3
EXAMPLE 11	76	81	1440	1760	13202	16796	26309	27595	36.8	5.2
EXAMPLE 12	106	124	1655	2090	13629	18194	26207	25983	34.8	5.1
EXAMPLE 13	281	269	2823	3519	25727	28267	17944	31564	35.1	5.2
EXAMPLE 14	272	313	3547	3830	22584	24428	34311	27206	36.3	5.0
EXAMPLE 15	49	35	3979	2935	310	150	1077	962	54.4	5.7
EXAMPLE 16	393	412	3433	4571	21658	27894	30700	28758	29.6	5.4

UNIT OF ENZYME TITER: U/g KOJI

[0107]

The numerical values in Tables 5 to 8 represent enzyme titer per gram of koji fungus-fermented products in U/g, by an enzyme titer measurement.

For the glucoamylase activity, 1 unit (U) thereof was defined as an activity for producing 1 mg of glucose from a soluble starch in 60 minutes at 40°C.

The α -amylase activity was represented by an amount of 1% solubilized starch (ml) degraded in 30 minutes at 40°C.

For the acid protease activity, 1 unit thereof was defined as an amount of enzyme that produces 1 μ g of tyrosine equivalent in 60 minutes at 40°C.

As for the acid carboxypeptidase activity, 1 unit thereof was defined as an amount of enzyme that produces 1 μ g of tyrosine equivalent from carbobenzoxyglutamyltyrosine in 60 minutes at 30°C.

[0108]

Moisture content was obtained according to an analysis method prescribed by the National Tax Agency JAPAN (decompression drying method), as in Test 1.

The numerical values of the dry koji are values obtained by converting a measured value of raw koji in a state of containing moisture into a value when the moisture content is 0%.

[0109] (Results of Test 2)

In cases where each of cereals were fermented by addition of euglena, there was a tendency that titer of each enzyme became high as compared with cases without addition of euglena.

In Examples 10 to 16 which used the white koji fungus, titer of each enzyme was high with use of any cereal, as compared with the corresponding Comparative Examples.

[0110]

From the above results, it has been found that a koji fungus-fermented product having a high titer of each enzyme is obtained when a mixture obtained by mixing a euglena powder with each of the cereals as a bulking agent is fermented by a koji fungus.

It has further been found that euglena has an action of promoting enzyme production of a koji fungus, from the fact that titer of each enzyme was high in Examples in which fermentation was carried out with addition of euglena, as compared with that of Comparative Examples in which only each cereal was fermented.

REFERENCE SIGNS LIST

[0111]

- 1: Stainless steel vat with lid
- 2: Slat
- 3: Petri dish with lid
- 4: Filter paper

CLAIMS

1. A koji fungus-fermented product, which is obtained by fermenting a raw material for a koji fungus-fermented product which is a steamed cereal coated with a mixture of a euglena algal body and a koji fungus.
2. The koji fungus-fermented product according to claim 1, wherein the steamed cereal is at least one kind selected from the group consisting of a germinated brown rice, a glutinous rice, a glutinous brown rice, a pearl barley, a naked barley, a soybean, and a rice.
3. The koji fungus-fermented product according to claim 1 or 2, wherein the koji fungus is a yellow koji fungus or a white koji fungus.
4. A food composition comprising the koji fungus-fermented product according to any one of claims 1 to 3.
5. A cosmetic composition comprising the koji fungus-fermented product according to any one of claims 1 to 3.
6. A raw material for a koji fungus-fermented product, which is a steamed cereal coated with a mixture of a euglena algal body and a koji fungus.
7. The raw material for a koji fungus-fermented product according to claim 6, wherein the steamed cereal is at least one kind selected from the group consisting of a germinated brown rice, a glutinous rice, a glutinous brown rice, a pearl

barley, a naked barley, a soybean, and a rice.

8. The raw material for a koji fungus-fermented product according to claim 6 or 7, wherein the koji fungus is a yellow koji fungus or a white koji fungus.

9. A method for producing a koji fungus-fermented product, characterized in carrying out:

a step of steaming a cereal, and mixing a euglena algal body with a koji fungus to prepare a mixture of a euglena algal body and a koji fungus;

a mixing step for obtaining a raw material for a koji fungus-fermented product by coating the steamed cereal with the mixture of a euglena algal body and a koji fungus obtained in the previous step; and

a fermentation step for fermenting the raw material for a koji fungus-fermented product.

10. The method for producing a koji fungus-fermented product according to claim 9, wherein the steamed cereal is at least one kind selected from the group consisting of a germinated brown rice, a glutinous rice, a glutinous brown rice, a pearl barley, a naked barley, a soybean, and a rice.

11. The method for producing a koji fungus-fermented product according to claim 9 or 10, wherein the koji fungus is a yellow koji fungus or a white koji fungus.

12. An enzyme production promoter for a koji fungus, characterized in

comprising a euglena algal body as an active ingredient, and

being used in promoting an enzyme production of a koji fungus.

13. The enzyme production promoter for a koji fungus according to claim 12, wherein the koji fungus is a yellow koji fungus or a white koji fungus.

14. The enzyme production promoter for a koji fungus according to claim 12 or 13, wherein the enzyme is at least one kind selected from the group consisting of glucoamylase, α -amylase, and acid protease.