

Description

Title of Invention: ORAL COMPOSITION, FLAVOR IMPROVING METHOD, FLAVOR IMPROVING AGENT FOR CHICKEN EXTRACT, AND USE OF HYDROLYZED COLLAGEN TYPE II

Technical Field

[0001] The present invention relates to an oral composition containing chicken extract and hydrolyzed collagen type II. The present invention also relates to a method of improving the flavor of a composition containing chicken extract, the method including adding hydrolyzed collagen type II to a composition containing chicken extract. The present invention also relates to a flavor improving agent for chicken extract. The present invention also relates to use of hydrolyzed collagen type II for production of a composition containing chicken extract.

Background Art

[0002] Collagen known as gelatin has been widely used in the food field. Since collagen which is an animal protein is a main component of dermis and connective tissue, collagen has been recently drawing attention also in the medical field and the cosmetic field. Generally, orally consumed high molecular weight collagen cannot be efficiently utilized in the body. Recently, low molecular weight collagen peptides obtained by hydrolysis of high molecular weight collagen have been developed in order to facilitate ingestion of collagen, and collagen and foods and/or beverages containing such a collagen peptide have also been developed.

[0003] Collagen is categorized into three types, I, II, and III. Collagens type I and type III are mainly found in skin, tendon, and bone. Collagen type II is mainly found in cartilage. For example, Patent Literature 1 discloses a method of adjusting hydrolyzed collagen type II, a therapeutic agent, and a nutritional supplement.

Citation List

Patent Literature

[0004] PTL 1: US Publication No. 6323319

Summary of Invention

Technical Problem

[0005] Chicken extract is considered to be the main nourishment in chicken meat. For example, in Southeast Asia, chicken extract is used as a "traditional medicine" that is beyond the category of food products and food materials.

Consumer's preference for chicken extract has been increasing, and there is a demand

for the development of chicken extract which provides a stronger chicken extract flavor such as umami. Chicken extract is a component obtained by hot water extraction using chicken parts such as chicken, chicken bone, chicken legs and chicken skins as a raw material. The chicken extract of chicken meat has an odor (fishy odor) and there is a demand for a technique that reduces the odor.

[0006] The present invention aims to provide an oral composition containing chicken extract in which the chicken extract has improved flavor.

The present invention also aims to provide a method of improving the flavor of chicken extract.

The present invention also aims to provide a flavor improving agent for chicken extract.

Solution to Problem

[0007] As a result of various studies on improvement of chicken extract, the present inventors found that adding hydrolyzed collagen type II of chicken extract results in reduced odor of the chicken extract and increased umami, favorably improving the taste. The present invention was thus completed.

[0008] Specifically, the present invention is defined as follows.

(1) An oral composition containing: chicken extract; and hydrolyzed collagen type II, the chicken extract containing histidine, carnosine, and anserine, wherein the amount of the histidine is 0.1 to 1 wt%, the amount of the carnosine is 0.01 to 0.3 wt%, and the amount of the anserine is 0.05 to 0.5 wt%.

(2) The oral composition according to (1) above, wherein the hydrolyzed collagen type II is derived from chicken cartilage.

(3) The oral composition according to (1) or (2) above, wherein the hydrolyzed collagen type II has a weight average molecular weight of 3000 to 6000.

(4) The oral composition according to any one of (1) to (3) above, wherein the hydrolyzed collagen type II contains a low molecular weight peptide having a molecular weight of less than 3000 and a high molecular weight peptide having a molecular weight of 3000 or more.

(5) The oral composition according to (4) above, wherein the content ratio by weight of the low molecular weight peptide to the high molecular weight peptide (low molecular weight peptide/high molecular weight peptide) is 85/15 to 97/3.

(6) The oral composition according to any one of (1) to (5), wherein the amount of the hydrolyzed collagen type II in the oral composition contains is 0.1 to 5 wt%.

(7) The oral composition according to any one of (1) to (6) above, wherein the oral composition is a beverage.

(8) The oral composition according to any one of (1) to (7) above, wherein the oral

composition is at least one selected from the group consisting of a composition for health and a composition for cosmetic use.

(9) A method of improving the flavor of a composition containing chicken extract, the method comprising: adding hydrolyzed collagen type II to a composition containing chicken extract.

(10) The method according to (8) above, wherein the chicken extract contains histidine, carnosine, and anserine, and the amount of the histidine in the composition is 0.1 to 1 wt%, the amount of carnosine in the composition is 0.01 to 0.3 wt%, and the amount of the anserine in the composition is 0.05 to 0.5 wt%.

(11) The method according to (9) or (10) above, wherein the hydrolyzed collagen type II is derived from chicken cartilage.

(12) The method according to any one of (9) to (10) above, wherein the hydrolyzed collagen type II has a weight average molecular weight of 3000 to 6000.

(13) The method according to any one of (9) to (12) above, wherein the hydrolyzed collagen type II contains a low molecular weight peptide having a molecular weight of less than 3000 and a high molecular weight peptide having a molecular weight of 3000 or more.

(14) The method according to (13) above, wherein the content ratio by weight of the low molecular weight peptide to the high molecular weight peptide (low molecular weight peptide/high molecular weight peptide) is 85/15 to 97/3.

(15) The method according to any one of (9) to (14) above, wherein the hydrolyzed collagen type II is added such that the amount of the hydrolyzed collagen type II in the composition is 0.1 to 5 wt%.

(16) A flavor improving agent for chicken extract containing hydrolyzed collagen type II as an active component.

(17) The flavor improving agent for chicken extract according to (16) above, wherein the hydrolyzed collagen type II is derived from chicken cartilage.

(18) The flavor improving agent for chicken extract according to (16) or (17) above, wherein the hydrolyzed collagen type II has a weight average molecular weight of 3000 to 6000.

(19) The flavor improving agent for chicken extract according to any one of (16) to (18) above, wherein the hydrolyzed collagen type II contains a low molecular weight peptide having a molecular weight of less than 3000 and a high molecular weight peptide having a molecular weight of 3000 or more.

(20) The flavor improving agent for chicken extract according to (19) above, wherein the content ratio by weight of the low molecular weight peptide to the high molecular weight peptide (low molecular weight peptide/high molecular weight peptide) is 85/15 to 97/3.

(21) Use of hydrolyzed collagen type II for the production of a flavor improving agent for chicken extract.

(22) Use of hydrolyzed collagen type II for the production of an oral composition containing thickened meat extract or a flavor improving agent for chicken extract.

Advantageous Effects of Invention

[0009] The present invention provides an oral composition containing chicken extract which can reduce odor and which can also improve the flavor of chicken extract such as umami. The present invention also provides a method of improving the flavor of a composition containing chicken extract. The present invention also provides a flavor improving agent for chicken extract.

Brief Description of Drawings

[0010] [fig.1]FIG. 1 shows a flow chart briefly describing a method of preparing hydrolyzed collagen type II.

[fig.2A]FIG. 2A shows a visual comparison between flavor (sensory) evaluation of an oral composition according to Example 2 and flavor (sensory) evaluation of an oral composition according to Comparative Example 1.

[fig.2B]FIG. 2B shows a visual comparison between flavor (sensory) evaluation of an oral composition according to Example 3 and flavor (sensory) evaluation of an oral composition according to Comparative Example 1.

[fig.2C]FIG. 2C shows a visual comparison between flavor (sensory) evaluation of an oral composition according to Example 4 and flavor (sensory) evaluation of the oral composition according to Comparative Example 1.

Description of Embodiments

[0011] <Oral composition>

The oral composition of the present invention contains chicken extract and hydrolyzed collagen type II, and the chicken extract contains histidine, carnosine, and anserine.

[0012] <Chicken extract>

The chicken extract for use in the present invention contains histidine, carnosine, and anserine.

Carnosine is β -alanyl-histidine, which is a dipeptide of β -alanine and histidine.

Anserine is β -alanyl-1-methylhistidine in which histidine is methylated. Preferably, the chicken extract for use in the present invention contains 0.1 to 1 wt% histidine, 0.01 to 0.3 wt% carnosine, and 0.05 to 0.5 wt% anserine.

[0013] In the case that the chicken extract for use in the present invention contains salts of histidine, carnosine, and/or anserine, the content amount of the histidine, carnosine and anserine in the chicken extract means a total amount of the histidine and salts thereof

in terms of histidine, a total amount of the carnosine and salts thereof in terms of carnosine and a total amount of the anserine and salts thereof in terms of anserine. Examples of the histidine salts, carnosine salts and anserine salts are not particularly limited, and include the same salts including inorganic salts such as hydroxides, carbonates, and bicarbonates of sodium, lithium, calcium, magnesium and aluminum and ammonia; and organic salts such as mono-, di-, or tri-alkylamine salts such as salts of methylamine, dimethylamine, and trimethylamine, mono-, di-, or tri-hydroxyalkylamine salts, guanidine salt, and N-methylglucosamine salt.

[0014] The chicken extract for use in the present invention may be, for example, an extract that can be obtained by heating chicken used as a raw material in a liquid, or a commercial product. The raw material may contain bone, cartilage, legs, or the like, but preferably, the raw material does not contain the head or internal organs.

[0015] Examples of the commercial product of the chicken extract (CE) include "Brand's Essence of Chicken (BEC) (produced by Suntory Beverage & Food Asia Pte Ltd)", "Scotch(R) Essence of Chicken (produced by Scotch Industrial (Thailand) Co., Ltd.)", "Quaker Essence of chicken (produced by Standard Foods Corporation (Taiwan) Co., Ltd.)", "Chicken stock and broth of SWANSON(R) Produced by Campbell Soup Company (NYSE:CPB)", "Drip Chicken Essence produced by Eu Yan Sang International Ltd. (Singapore)", "Boned Chicken Tonic produced by Eu Yan Sang International Ltd. (Singapore)", "Boiled Essence of Chicken produced by Lao Xie Zhen Co. Ltd. (Taiwan)". Any of such commercial products may be used, but use of Brand's Essence of Chicken (BEC) is preferred.

[0016] When producing chicken extract for use in the present invention by hot water extraction, the chicken extract can be produced by a method that is usually used in this field. For example, normal pressure extraction and/or pressurized extraction is performed using a liquid at a temperature of 100°C or higher, preferably 125°C or higher, and the resulting extract is treated with a membrane or filtered, whereby chicken extract can be produced. Specifically, the extract is obtained by a pre-treatment step (1) in which chicken is heated in a liquid, and a step (2) in which the liquid is replaced with a fresh liquid after the pre-treatment and the chicken is heated again. The heat treatment in each of the step (1) and the step (2) is preferably performed in a solvent. The solvent is preferably water, ethanol, or a mixture of these, for example.

The chicken extract encompasses a liquid extract obtained by the method described above; a diluted solution, concentrate, or dry powder of the liquid extract; and purified products of these. The purified products may be obtained by, for example, subjecting liquid chicken extract to ultrafiltration, membrane treatment, liquid separation operation, or fraction treatment with resin or the like so as to increase the purity. After

increasing the purity of the chicken extract, the purified product may be formed into powder by freeze-drying or spray-drying, for example.

- [0017] The amount of chicken extract (in terms of solids) in the oral composition of the present invention is, for example, preferably 0.1 wt% or more, more preferably 0.5 wt% or more, and preferably 99 wt% or less, more preferably 90 wt% or less. In an embodiment, for example, the amount of chicken extract (in terms of solids) in the composition is, for example, preferably 0.01 to 99 wt%, more preferably 0.1 to 90 wt%.
- [0018] The amount of histidine in the oral composition of the present invention is, for example, preferably 0.00001 wt% or more, more preferably 0.0001 wt% or more, and is preferably 10 wt% or less, more preferably 1 wt% or less. In an embodiment, for example, the amount of histidine in the composition is, for example, preferably 0.00001 to 10 wt%, more preferably 0.0001 to 1 wt%.
- [0019] The amount of carnosine in the oral composition of the present invention is, for example, preferably 0.00001 wt% or more, more preferably 0.0001 wt% or more, and is preferably 10 wt% or less, more preferably 1 wt% or less. In an embodiment, for example, the amount of carnosine in the composition is, for example, preferably 0.00001 to 10 wt%, more preferably 0.0001 to 1 wt%.
- [0020] The amount of anserine in the oral composition of the present invention is, for example, preferably 0.00001 wt% or more, more preferably 0.0001 wt% or more, and is preferably 10 wt% or less, more preferably 1 wt% or less. In an embodiment, for example, the amount of anserine in the composition is, for example, preferably 0.00001 to 10 wt%, more preferably 0.0001 to 1 wt%.
- [0021] The amounts of histidine, carnosine, and anserine in the oral composition can be quantitated by HPLC by a method usually used in this field.
- [0022] <Hydrolyzed collagen type II>
The hydrolyzed collagen type II for use in the present invention is a component extracted from cartilage of livestock such as chickens, pigs, cows, or the like, preferably a component derived from chicken cartilage. The hydrolyzed collagen type II derived from chicken cartilage is preferred because chicken cartilage contain high purity of Type-II collagen.
- [0023] Hydrolyzed collagen type II can be obtained by hydrolysis of collagen type II with an enzyme or the like. The collagen type II can be extracted from cartilage of chickens, pigs, cows, or the like by a known method. The hydrolyzed collagen type II for use in the present invention can be prepared from livestock (preferably chicken) cartilage by a method usually used in this field. For example, the hydrolyzed collagen type II can be obtained by treating the chicken cartilage with an enzyme. Specifically, the hydrolyzed collagen type II can be prepared by a pre-treatment step (3) in which chicken

cartilage is heated in a liquid, and a step (4) in which the chicken cartilage after the pre-treatment step is treated with an enzyme. The enzyme for use in the step (4) is not limited as long as it is one usually used in this field. Examples include collagenase, papain, bromelain, actinidine, ficin, cathepsin, pepsin, chymosin, trypsin, protease, subtilisin, aminopeptidase, exopeptidase, endopeptidase and enzyme preparations obtained by mixing these enzymes. The method of preparing hydrolyzed collagen type II is not limited to the enzyme treatment method.

[0024] Preferably, the hydrolyzed collagen type II has a weight average molecular weight of 3000 to 6000.

Preferably, the hydrolyzed collagen type II contains a low molecular weight peptide having a molecular weight of less than 3000 and a high molecular weight peptide having a molecular weight of 3000 or more, because the effect of reducing the odor and bitter taste of the hydrolyzed collagen type II can be achieved by addition of a low molecular weight peptide and a high molecular weight peptide.

[0025] Herein, the weight average molecular weight of the hydrolyzed collagen type II can be calculated by using the data of a molecular weight distribution which is obtained by a method usually used in this field such as Eurofins HPAEC-PAD method.

[0026] In the hydrolyzed collagen type II of the present application, the content ratio by weight of the low molecular weight peptide to the high molecular weight peptide (low molecular weight peptide/high molecular weight peptide) is preferably 85/15 to 97/3 because the effect of reducing odor and bitter taste of hydrolyzed collagen type II can be achieved by addition of a low molecular weight peptide and a high molecular weight peptide at the specific mixing ratio. The content ratio of the low molecular weight peptide to high low molecular weight peptide is more preferably 90/10 to 96/4.

[0027] Preferably, the oral composition of the present invention contains the hydrolyzed collagen type II in an amount of 0.1 to 5 wt% in the oral composition because the odor of the chicken extract can be reduced and the umami and/or thickness can be sufficiently improved in the oral composition of the present invention. Preferably, the oral composition contains the hydrolyzed collagen type II in amount of 0.5 to 5wt%.

Herein, the "thickness" as one of sensory attributes of the flavor refers to the degree of retention of the sample in the mouth and the degree of smooth feeling in the throat.

More preferably, the oral composition contains the hydrolyzed collagen type II in an amount of 1.5wt% or more, further preferably 2.0 wt% or more, still further preferably 2.5wt% or more, because the odor can be reduced and not only the umami and thickness but also the Meaty flavor of the chicken can be sufficiently improved.

[0028] <Other components>

The oral composition of the present invention may contain one or two or more components in addition to those described above as long as the effects of the present

invention are not impaired.

The oral composition of the present invention preferably contains a herb, a flavor, a masking agent, a sweetener, or the like. The herb, the flavor or a masking agent can impart a suitable aroma, and the resulting oral composition has a better flavor. The sweetener can impart a pleasant taste, and the resulting oral composition has a better flavor. Any herb and any flavoring may be used. Commonly used herbs and flavorings can be used alone or in combination of two or more thereof. Any sweetener can be used. Examples include sugars, sugar alcohols, and high-intensity sweeteners. One or two of these can be used in combination.

[0029] Examples of the sugars include monosaccharides, disaccharides, trisaccharides, and higher polysaccharides (including oligosaccharides). Specific examples include glucose, fructose, galactose, mannose, sucrose, maltose, lactose, and trehalose. Examples of the sugar alcohols include erythritol, xylitol, sorbitol, mannitol, maltitol, and reduced palatinose. Of these, erythritol is more preferred.

[0030] A high-intensity sweetener is a sweetener having a more intense sweet taste than sugars. Specific examples include acesulfame potassium (acesulfame K), sucralose, aspartame, stevia, saccharin, sodium saccharin, and netotame. Of these, acesulfame K and sucralose are preferred. In one embodiment, the oral composition of the present invention preferably contains a sugar or sugar alcohol and a high-intensity sweetener, and more preferably contains acesulfame K, sucralose, and erythritol. The oral composition containing these sweeteners has a better flavor.

[0031] The oral composition of the present invention may contain, in addition to those mentioned above, additive such as acidulants, antioxidants, stabilizers, preservatives, flavorings, masking agents, emulsifiers, pigments, seasonings, pH adjustors, and nutritional enhancers, as long as the effects of the present invention are not impaired.

[0032] The pH of the oral composition of the present invention is preferably 5.0 or less, more preferably 3.0 to 4.5, still more preferably 3.3 to 4.0, in terms of antiseptic properties. Herein, the pH is measured at 25°C. The pH can be adjusted using an acidulant. The acidulant is preferably an acid or its salt that can be used in foods and beverages. Examples include acids such as citric acid, phosphoric acid, lactic acid, malic acid, tartaric acid, succinic acid, fumaric acid, gluconic acid, phytic acid, and their salts. These acidulants may be used alone or in combination of two or more thereof. Only free acids or only their salts may be used, or free acids and their salts may be used in combination as the acidulant. In terms of flavor of the oral composition, the acidulant preferably contains citric acid or its salt, and more preferably contains citric acid or its salt and phosphoric acid or its salt.

[0033] The amount of the acidulant can be set according to the type of the acidulant. For example, the total amount of acid or its salt used as an acidulant in terms of free acid

amount of the acid or its salt in the oral composition is preferably 100 to 5000 mg/100 mL, more preferably 300 to 3000 mg/100 mL. The above amount refers to the total amount when the composition contains a plurality of acidulants. Herein, the expression "amount in terms of free acid amount" or an expression similar to that refers to the amount of free acid when the acid is in the form of a free acid. Alternatively, when the acid is in the form of a salt, the expression refers to a value obtained by multiplying the molar number of the salt by the molecular weight of the corresponding free acid.

[0034] The method of producing the oral composition of the present invention is not limited. For example, the method preferably includes a step of mixing the components (mixing step). The method also preferably includes steps such as a step of adjusting the pH of the composition and/or a step of adjusting the viscosity of the composition, if necessary.

In the mixing step, preferably, the components are mixed by adding an aqueous medium, if necessary. The aqueous medium is preferably water. The order of mixing the components is not limited as long as the components are uniformly mixed. In one embodiment, when adding a volatile component (e.g., flavoring) and an easily decomposable component (e.g., vitamin C), these components are preferably added at the end.

[0035] The oral composition of the present invention is preferably used as a beverage (a beverage composition).

The oral composition of the present invention can be packed in a container. The form of the container is not limited. The oral composition can be packed in a sealed container such as a bottle, can, PET bottle, paper package, aluminum pouch, or a plastic pouch, whereby a packed beverage or the like can be obtained.

[0036] Preferred examples of the beverage include tea-based beverages, coffee beverages, alcohol beverages, non-alcohol beer, carbonated beverages, functional beverages, fruit and vegetable beverages, dairy beverages, soy source beverages, and flavored water.

[0037] The oral composition of the present invention is preferably used for at least one application selected from the group consisting of health application and cosmetic application. Here, the health application can include use for locomotive syndrome.

The oral composition of the present application is preferably used for mobility.

[0038] <Flavor improving method>

The present invention also encompasses a method of improving the flavor of a composition containing chicken extract, the method including adding hydrolyzed collagen type II to a composition containing chicken extract. Adding the hydrolyzed collagen type II to a composition containing the chicken extract can reduce the odor of the chicken extract, increase the flavor such as umami, and suitably improve the flavor. In one embodiment, the present invention can improve the umami and/or thickness which

is one of sensory attributes of the flavor of chicken extract. In another embodiment, the present invention can improve the umami and/or thickness which is one of sensory attributes of the flavor of chicken extract, and/or can improve the chicken meaty flavor. The method and timing of adding hydrolyzed collagen type II to a composition containing the chicken extract are not limited, as long as the composition containing the chicken extract ultimately contains the hydrolyzed collagen type II. Preferred embodiments, amounts, and the like of the chicken extract and the hydrolyzed collagen type II are the same as those of the oral composition described above. Further, the composition containing chicken extract for use in the flavor improving method may also contain components other than the chicken extract and the hydrolyzed collagen type II, as is the case with the oral composition.

[0039] <Flavor improving agent>

The present invention also encompasses a flavor improving agent for chicken extract containing hydrolyzed collagen type II as an active component. Adding the hydrolyzed collagen type II to the chicken extract can reduce the odor of the chicken extract, increase the flavor such as umami, and suitably improve the flavor. In one embodiment, the present invention can improve the umami and/or thickness which is one of sensory attributes of the flavor of chicken extract. In another embodiment, the present invention can improve the umami and/or thickness which is one of sensory attributes of the flavor of chicken extract, and/or can improve the chicken meaty flavor. Preferred embodiments, amounts, and the like of the chicken extract and the hydrolyzed collagen type II are the same as those of the oral composition described above.

[0040] According to an embodiment, the present invention may provide a method of producing an oral composition including a step of mixing chicken extract with hydrolyzed collagen type II. The chicken extract and the hydrolyzed collagen type II may be added in any order in the method of producing an oral composition.

The present invention may also provide a use of hydrolyzed collagen type II for the production of a flavor improving agent for chicken extract.

The present invention may also provide a use of hydrolyzed collagen type II for the production of an oral composition containing thickened meat extract or a flavor improving agent for chicken extract.

Preferred embodiments, amounts, and the like of the chicken extract and the hydrolyzed collagen type II are the same as those of the oral composition described above.

Examples

[0041] The present invention is more specifically described below with reference to

examples. The present invention is not limited to these examples.

[0042] Raw materials, reagents, and the like used in the examples and the comparative examples are described below.

[0043] <Preparation of hydrolyzed collagen type II (hereinafter also referred to as "HCII") powder>

FIG. 1 shows a flow chart briefly describing a HCII preparation method. First, frozen chicken cartilage was thawed in water at 40°C, and cleansed in water at 40°C (1 hour). Next, the wash water was discarded, and fresh water was poured into a 1200L pot as to be 3 times amount to the chicken cartilage. The water was heated to the optimal working temperature for enzyme treatment, and the washed chicken cartilage was immersed therein to perform enzyme treatment for several hours. After the enzyme treatment, the pot containing the chicken cartilage was heated to 90°C or higher, and the temperature was held at 90°C or higher for 30 minutes, whereby the enzyme used in the earlier enzyme treatment was inactivated. The resulting mixture (the liquid and the enzyme-treated chicken cartilage) was filtered. The resulting liquid was concentrated. Lastly, the resulting concentrate was spray-dried at 200°C, whereby HCII powder was prepared.

[0044] <Measuring for the molecular weight of the HCII powder>

The molecular weight distribution of the HCII obtained was measured by Eurofins HPAEC-PAD method. Table 1 below shows the molecular weight distribution of the HCII obtained. The HCII obtained had a weight average molecular weight of 4582 which was obtained by calculating with usually used method in this field.

[0045]

[Table 1]

Molecular weight Mass Range	Percentage of Molecules	Assumed Mass of Molecule	Total Mass Type of Molecule	Weight Fraction Type of Molecule	Weight Molecular Weight
	Ni(%)	Mi(Da)	NiMi	(NiMi)/Σ(NiMi)	WiMi
<500 D	57.0	500	285	0.221	110.47
500 - 1000 D	23.0	750	172.5	0.134	100.29
1000 - 3000 D	14.0	2000	280	0.217	434.11
3000 - 5000 D	1.0	4000	40	0.031	124.03
5000 - 10000 D	1.1	7500	82.5	0.064	479.65
> 10000 D	4.3	10000	430	0.333	3333.33
			Σ(NiMi)=1290		Σ(WiMi)=4581.88

[0046] <Chicken extract>
Commercially available chicken extract (CE) (Brand's Essence of Chicken (BEC)
(manufacturer: Cerebos Pacific Limited)) was used. In 1 ml of CE, the amount of

carnosine was 0.94 mg/ml, and the amount of anserine was 1.9 mg/ml. In 100ml of CE, the amount of histidine was 0.45g/100ml.

[0047] <Quantification of histidine, carnosine, and anserine>

Quantification of histidine, carnosine, and anserine in the chicken extract was performed by HPLC under the following conditions.

<Analysis conditions of HPLC>

Device: high performance liquid chromatography system with a UV detector (Agilent 1100 Series available from Agilent)

Column: Zorbax 300-SCX 4.6 mm ID × 250 mm available from Agilent.

Mobile phase: 50 mM potassium dihydrogen phosphate

Flow rate: 1.0 mL/min

Flow channel: channel A (50 mM potassium dihydrogen phosphate), channel B (acetonitrile), channel D (deionized water)

UV detector wavelength: 210 nm

Sample injection volume: 10 µL

[0048] <Examples 1 to 4>

Oral compositions (solutions) of Examples 1 to 4 were prepared according to the amounts shown in Table 2 below.

Specifically, HCII powder in an amount shown in Table 2 was added to chicken extract (CE) (68mL) and the mixture was sufficiently stirred, whereby a liquid oral composition was prepared in each of Examples 1 to 4. Each of the resulting liquid oral compositions of Examples 1 to 4 was dispensed into a test tube, followed by sterilization at 121°C for 8 minutes. Table 2 shows the HCII concentration of each of the liquid oral composition of Examples 1 to 4.

[0049] <Comparative Example 1>

A liquid oral composition according to Comparative Example 1 was prepared by the same method as in Example 1, except that the HCII powder was not added.

[0050]

[Table 2]

	Example 1	Example 2	Example 3	Example 4	Comparative Example 1
HClI (g)	0.5	1.0	2.0	3.0	-
HClI Concentration of the composition (mg/g)	7.09	14.085	27.778	41.096	0.00

[0051] <Flavor evaluation>

Five expert panelists evaluated the flavor of the liquid oral compositions (about 50 mL, each) according to Examples 1 to 4 and Comparative Example 1 which were placed in plastic cups at room temperature. The evaluation criteria are as follows.

[0052] <Evaluation criteria>

Table 3 below shows sensory tributes (soy sauce aroma, fishy odor, chicken flavor, meaty flavor, caramelized flavor, salty, umami, bitter, thickness, and astringent) of the flavor. A reference sample to be used for evaluation of each sensory attribute was prepared as below. A sample was diluted in water to a concentration shown in "Reference samples and intensity" of Table 3 below, and the resulting diluted solution was used as a reference sample. Specifically, for example, one of the sensory attributes (soy sauce aroma) of the flavor is defined as the aroma associated with smell of soy sauce. The score of the soy sauce is 5 when the diluted soy sauce contains 25% soy sauce, and the score of the soy sauce is 10 when the diluted soy sauce contains 50% soy sauce. These scores were used as evaluation criteria.

[0053]

[Table 3]

Sensory attributes	Definitions	Reference samples and intensity
Soy Sauce aroma	The aroma associated with smell of soy sauce	- 25% soy sauce (5) - 50% soy sauce (10)
Fishy aroma	Refer to raw fish smell in fish market	NR
Chicken flavor	The flavour intensity reminiscent of cooked chicken meat like steamed chicken breast without seasoning	- 25% water from cooked chicken fillet (1) - 50% water from cooked chicken fillet (5) - 100% water from cooked chicken fillet (10)
Meaty flavor	Flavour characteristic associated with cooked meat	- 25% water from cooked chicken fillet (1) - 50% water from cooked chicken fillet (5) - 100% water from cooked chicken fillet (10)
Caramelised flavor	The character associated with burnt soy sauce	NR
Salty	One of the basic taste, common to sodium chloride	- 0.2% sodium chloride in water (1) - 0.5% sodium chloride in water (5) - 1% sodium chloride in water (10)
Umami	One of the basic taste, common to MSG. The taste and mouth-filling sensation of compounds such as glutamates that is savoury, brothy, meaty rich, full, and complex, common to many foods such as soy sauce, stocks, ripened cheese like parmesan, shellfish eg. lobster, scallops and mushrooms particularly porcini	- 0.5% MSG in water (1) - 1% MSG in water (5) - 2% MSG in water (10)
Bitter	Refer to quinine	NR
Thickness	The degree of retention of the samples in the mouth and the degree to which the samples flow into the throat	- BEC without caramel (1) - BEC with caramel (5)
Astringent	The degree to which the samples give a drying mouthfeel like after eating unripe banana, strong black tea or tannin in water	- 25% water from cooked chicken fillet (1) - 50% water from cooked chicken fillet (5) - 100% water from cooked chicken fillet (10)

[0054] <Selection and training of five expert panelists>

Five panelists who live in Singapore participated in the evaluation. A group discussion on the differences and similarities between the liquid oral composition

(Examples 1 to 4 and Comparative Example 1) were led by a panel leader during evaluation. A list of sensory attributes that described the flavor (sensory characteristics) of the liquid oral composition, focusing on the attributes of the flavor of the chicken extract believed to be influenced by HCII, was developed by the panelists. During the 2-h training session before the actual evaluation session, besides term generation, the panelists also learned to use a 10-point scale to rate attribute intensities. For example, evaluation criteria for the sensory attribute "Salty" are as follows: the score of the salty is 1 when the diluted sodium chloride contains 0.2% sodium chloride; the score is 5 when the diluted sodium chloride contains 0.5% sodium chloride; and the score is 10 when the diluted sodium chloride contains 1% sodium chloride. Each sample was tested at least twice during this training session.

[0055] <Evaluation>

For the evaluation of the flavor of the liquid oral compositions according to Examples 1 to 4 and Comparative Example 1, the five panelists smelled each liquid oral composition (about 50 mL) at room temperature, held the liquid oral composition in the mouth for 10 seconds, and rated the aroma, taste, flavor, and texture/mouthfeel attributes described in Table 3 above. They then completed the rating for each sensory attribute on a 10-point scale. Human sensory analyses were conducted following the spirit of the Helsinki Declaration and informed consent was obtained from all panelists.

[0056] <Analysis of evaluation results>

Statistical analyses were conducted on the results of flavor (sensory) evaluation obtained from the five panelists, using GraphPad Prism version 5.0 (San Diego, California, USA). Table 4 below shows results of the statistical analysis. The results are expressed as mean $\pm$ standard deviation. The obtained results were assessed by two-way analysis of variance followed by post-hoc Bonferroni analysis. Data was considered to be significant when $p < 0.05$.

[0057] FIG. 2A, FIG. 2B, and FIG. 2C show visual comparisons between the flavor (sensory) evaluation on the liquid oral compositions according to Examples 2, 3, and 4 and the flavor (sensory) evaluation of the liquid oral composition according to Comparative Example 1.

[0058]

[Table 4]

	Comparative Example 1	Example 1	Example 2	Example 3	Example 4
Sensory attributes	CE	CE with HCII 0.5g	CE with HCII 1g	CE with HCII 2g	CE with HCII 3g
Concentration of HCII (mg/g)	0	7.09	14.085	27.778	41.096
Salty aroma	3 ± 1.23	3 ± 1.41	3.6 ± 1.82	4.2 ± 1.79	5 ± 1.87
Fishy aroma	6.4 ± 2.51	5 ± 2.12	4.8 ± 1.79	5 ± 2	4.8 ± 2.77
Chicken flavor	5.6 ± 2.19	5.8 ± 1.64	6.8 ± 1.64	7.2 ± 2.05	7.8 ± 2.49
Meaty flavor	5 ± 0.71	6.8 ± 0.84	7.6 ± 0.89	8.4 ± 1.14 **	8.8 ± 1.1 ***
Caramelised flavor	4.8 ± 2.17	4.8 ± 2.39	5.6 ± 2.41	6.4 ± 2.89	6.6 ± 3.13
Salty	4.6 ± 2.07	4.8 ± 1.3	5.6 ± 1.52	6.4 ± 1.82	7 ± 2.35
Umami	4.4 ± 1.52	6 ± 1	7.2 ± 0.45 *	8.2 ± 0.84 ***	8.8 ± 1.1 ***
Bitter	5 ± 2.35	5.4 ± 1.52	5.8 ± 2.05	7 ± 2.45	7.6 ± 2.07
Thickness	2.6 ± 1.52	4 ± 1	5.4 ± 0.89 *	7 ± 1.87 ***	7.8 ± 1.92 ***
Astringent	5.2 ± 1.92	4.8 ± 1.92	5.2 ± 2.39	6 ± 2.24	6.8 ± 2.59

1 Data represented as mean ± SD

2 * p < 0.05, **p < 0.01, ***p < 0.001

[0059] The results in Table 4 above show that the liquid oral composition according to Example 1 in which 0.5 g of HCII was added to CE reduced "fishy odor" and improved "umami" in the sensory attributes. Further, the liquid oral composition according to Example 2 in which 1 g of HCII was added to CE significantly enhanced the intensity of "umami" in the sensory attributes and gave a thicker mouthfeel as a whole compared to the liquid oral composition according to HCII-free Comparative Example 1. The same can be observed also in FIG. 2A. Further, the results in Table 4, FIG. 2B, and FIG. 2C show that the liquid oral composition according to Example 3 in which 2 g of HCII was added to CE and the liquid oral composition according to

Example 4 in which 3 g of HCII was added to CE significantly enhanced the meaty flavor of CE in addition to the improvement in the umami and the thicker mouthfeel. Yet, adding HCII at different concentrations to CE did not significantly change the intensity of other attributes.

Umami taste has been defined not only as the 'MSG-like' but also the 'mouth-filling' sensation of compounds such as glutamates that is savory, flavorful, brothy, meaty, rich, full and complex, which is identified in many common foods (such as soy sauce and stocks (liquids obtained by boiling bones or meat in water, which are used to make soups and sources), ripened cheese, shellfish such as lobsters, mushrooms such as porcini mushrooms, cashews and asparagus. Hence, the enhancement of umami in CE by addition of 1 g of HCII to CE implies an enhancement in other sensations such as richness and complexity of CE.